Serological Survey of Brucella Infection in Small Ruminants in Yobe North, Yobe State, Nigeria

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ABSTRACT
Brucella infection is a zoonotic bacterial disease which affects variety of animals and humans. A cross-sectional survey was conducted in Jakusko Yobe North to determine the seroprevalence and risk factors associated with brucellosis. A total of 250 sera samples were collected aseptically from the jugular vein of sheep of both sex and different age groups, sera were screened against Brucella spp. using Rose Bengal Plate test (RBPT). The seroprevalence of 10.4% was recorded out of 250 sheep sampled, with the prevalence rates of 12.1% and 8.3% for female and male sheep respectively. There was no significant difference between sex and Brucella infection (OR = 0.6565, 95% CI = 0.2806 – 1.536, p = 0.443). Seroprevalence obtained was higher in sheep above two years (14.6%) but the seroprevalence was lower in sheep below two years (5.8 %), there was significant statistical difference between the age of sheep and Brucella infection (OR = 0.3619, 95% CI = 1463 – 0.8951, p = 0.0364). Though, brucellosis was reported in all the locations of sheep sampled, but there was no significant difference between the location of sheep. The study indicated the presence of Brucella infection in Jakusko and this is the first report of brucellosis in the study area, which is of public health significant because of the close contact between people and their animals. The information obtained will be useful in setting control, prevention, and eradication program among herdsmen/farmers.

Keywords: Seroprevalence, Brucella, Sheep, Yobe, RBPT
INTRODUCTION
Brucellosis is an important bacterial zoonotic disease that mostly affects variety of animals as well as humans. The disease causes economic loses worldwide as recognized since ancient times [1]. It is one of the most prevalent and neglected bacterial zoonotic disease by Food and Agriculture Organization and World Health Organization which have public health significance globally [2] Brucellosis is a multiple species disease, infection and infestation [3]. Brucellosis is a disease caused by the Brucella organism, Brucella melitensis and B. abortus are the most important species that have high potential for human infection [4] that mainly affect small ruminants and cattle respectively [5]. The natural reservoirs of the B. melitensis species are basically goats and sheep but occurs occasionally in cattle and swine. However, B. ovis is primarily affect sheep [6]. The disease is transmitted to humans through consumption of unpasteurized raw milk or other dairy products but in animals, transmission is by direct contact with the discharges from infected animals and ingestion of feed and water and contaminated by Brucella organism [7]. In humans, brucellosis can be a serious, debilitating and sometimes chronic disease that may affect a variety of organs with signs as joint pain, undulating fever and general body malaise [8]. The disease cause abortion, infertility, and consequently, reduction of milk yields in livestock [5]. Farouk et al. [9] demonstrated Brucella antibodies in cattle milk in Jigawa State where they reported the prevalence of 7.4% and 3.4% in marketed milk and in fresh milk. Brucellosis has also been reported in livestock and humans in many parts of Nigeria [10, 11, 12, 13, 14]. Small ruminants are the major source of meat supply in Yobe State, they play a vital role in the human diet and contribute a significant part of the total calories intake. The objective of this study was to determine the seroprevalence of brucellosis in sheep in Jakusko Local Government Area, Yobe State.

MATERIALS AND METHODS
Study Area
The study was carried out in Jakusko Local Government Area of Yobe State, Nigeria. Yobe State located in the Northern part in North-Eastern part of Nigeria. Jakusko is located at latitude and longitude 12º 22′ 09′N and 10º 46′23′E respectively. Yobe State is located in the North-Eastern part of Nigeria having total land area measuring up to 45,502 km². Yobe State is hot and dry with the exception of some part which has a milder climate. The dry zone has slightly harsh climatic condition with a dry season starting from November to April with average daily peak temperature especially in April and May of 34.4–37.8°C. Yobe States shares international border with Niger Republic which enhances trans-border movement of livestock between the two countries [15].

Study Design
A cross-sectional survey was used in the study, the study was conducted between January to April, 2020 in two major towns (Amshi and Jakusko) in Jakusko Local Government Area, Yobe State Northeastern part of Nigeria using simple random technique in picking the sheep. Two hundred and fifty blood samples were randomly collected from sheep to determine the seroprevalence of brucellosis and investigate the risk factors associated with the infection.

Sample Size Determination and Sample Collection
The sample size for this study was determined using [16] formula, with prevalence of 6.0% [17]
where:

\[ n = \frac{Z^2 pq}{d^2} \]

- \( n \) was the sample size,
- \( z \) was the standard normal deviate for the 95% confidence interval (1.96),
- \( p \) was the prevalence 6.0% [17]
- \( d \) was the desired precision (0.05), and \( q \) was \( 1 - p \)

\[ n = \frac{1.96^2 \times 0.06(1 - 0.06)}{(0.05)^2} \]

\( n = 86 \)

Based on the calculation using the formula above, minimum of 86 samples was required for this research, though the samples were increased to 250 to increase the precision of the test. Five (5) ml of blood sample was aseptically collected from each sheep identified in the flock, using 21-gauge syringe and needle, each sample was numbered. The collected blood samples were transported to the Veterinary Public Health Laboratory, University of Maiduguri in a cooler with ice-pack. These samples were then centrifuged at 3000g for 5 minutes, clear sera were obtained and decanted into two (2) ml cryovial tubes and stored at -20 °C till tested.

Serology
Serological test was carried out using Rose Bengal Plate Test (RBPT) antigen. The RBPT was carried out and the results were recorded in accordance with the method described by Alton et al. [18]. The test was conducted in the Bacterial Research Laboratory in the Department of Veterinary Public Health and Preventive Medicine, Faculty of Veterinary Medicine, University of Maiduguri.

The RBPT antigen was obtained from the Animal and Plant Health Agency (Addlestone, Surrey, U.K.). Briefly, 30 μL of serum were dispensed on the white ceramic tile which was then mixed with 30 μL of RBPT antigen using applicator stick. The mixture was shaken gently at room temperature for four (4) minutes and any visible agglutination or the appearance of a typical rim was recorded as a positive result; while the absence of agglutination was recorded as negative [19].

Data analysis
The data generated from this study were stored in Microsoft Excel spreadsheet, statistical analyses were performed using Statistical Package for Social Sciences (SPSS) (version 21.0; Inc., Chicago, IL, USA) at 0.05% level of significance. Prevalence was calculated by dividing the number of positive and the total number, then expressed as a percentage. The relationship between variables with seropositivity of disease was determine using chi-square (\( \chi^2 \)) and Fisher’s exact analysis to test for the association. Strength of association was calculated using odds ratio (OR) at 95% confidence interval (CI).

RESULTS
Out of 250 sheep tested, 26(10.4%) was seropositive to Brucella infection. Out of the 141 female sheep tested, 17(12.1%) were seropositive, while out of 109 male sheep tested, 9(8.3%) were seropositive. There was no statistically significant difference \( (p = 0.443) \) between the male and female sheep tested. Age distribution, showed that seroprevalence of brucellosis recorded was higher in sheep greater than two years (14.6%) while the least seroprevalence recorded was in sheep less than two years (5.8%). There was statistically significant difference \( (p = 0.036) \) between the age of sheep tested. Based on location, the seroprevalence of brucellosis detected was higher in Jakusko (11.2%) than sheep that were sampled from Amshi (9.6%), though there was no statistically significant difference \( (p = 0.835) \) between the location of sheep tested (Table 1).
### Table 1: Seroprevalence of brucellosis in sheep in Jakusko, Yobe State based on sex, age and location

<table>
<thead>
<tr>
<th>Variable</th>
<th>Category</th>
<th>Number examined</th>
<th>Number positive (%)</th>
<th>Number negative (%)</th>
<th>OR 95% CI lower</th>
<th>OR 95% CI upper</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>Male</td>
<td>109</td>
<td>9 (8.3)</td>
<td>100 (91.7)</td>
<td>0.6565</td>
<td>0.2806</td>
<td>1.536</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>141</td>
<td>17 (12.1)</td>
<td>124 (87.9)</td>
<td>1 (ref)</td>
<td>1 (ref)</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>≤ 2 years</td>
<td>120</td>
<td>7 (5.8)</td>
<td>113 (94.2)</td>
<td>0.3619</td>
<td>0.1463</td>
<td>0.895</td>
</tr>
<tr>
<td></td>
<td>&gt; 2 years</td>
<td>130</td>
<td>19 (14.6)</td>
<td>111 (85.4)</td>
<td>1*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Location</td>
<td>Amshi</td>
<td>125</td>
<td>12 (9.6)</td>
<td>113 (90.4)</td>
<td>0.8420</td>
<td>0.3729</td>
<td>1.901</td>
</tr>
<tr>
<td></td>
<td>Jakusko</td>
<td>125</td>
<td>14 (11.2)</td>
<td>111 (88.8)</td>
<td>1*</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td>250</td>
<td>26 (10.4)</td>
<td>224 (89.6)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1* = reference

### DISCUSSION

The overall seroprevalence of brucellosis recorded in this study was lower than 16.8% reported by Adamu et al. [13] from Kaduna State and 24.0% reported from Libya by Ahmed et al. [19], but the seroprevalence was comparable to 10.7% reported in Zaria by Buhari et al. [20] and 10.9% reported from Sokoto by Junaidu et al. [10]. However, the seroprevalence was higher than 8.2% reported from Giwa by Dogo and Maikai [21], 2.5% reported from Sudan by Abdallah et al. [22] and 8.6% from Kenya by Nakeel et al., [23]. The different seroprevalence rates could be due to the differences in the sample sizes in the various studies, differences in the animal production systems and husbandry and differences in the geographical regions and ecological settings. The seroprevalence of was high in female than male animals tested, this agreed with the works reported by Junaidu et al. [10] from Sokoto, Adamu et al. [13] from Kaduna, Buhari et al. [20] from Zaria and Sorsa et al. [24] from Southern Ethiopia who reported high seroprevalence in female's animals than in male. The high seroprevalence recorded in female animals than male is associated with keeping female animals for relatively longer period in the flock for breeding and could also be due to high concentration of erythritol in the uterus of female which stimulates the growth of the *Brucella* organisms [25]. The high seroprevalence was recorded in animals older than two years compare to animals' below two years. This agreed with the findings of Aworh et al. [26] and Adamu et al. [13] in Nigeria, it also agreed with the report of Ebid et al. [27] in the Arabian Gulf region. The report was in contrast with the work reported by Bashitu et al. [28] in Ethiopia and Buhari et al. [20] in Nigeria. This was because younger animals tend to be more resistant to infection and frequently clear an established infection, although latent infections can occur [29]. This may result from the fact that sex hormones and erythritol, which stimulate the growth and multiplication of *Brucella* organisms, tend to increase in concentration with age and sexual maturity [30]. High seroprevalence was recorded in flocks from Jakusko compared to animals from Amshi, though there was no statistically significant association between the location of the animals tested and seropositivity of *Brucella* specie. In conclusion, this study established the presence of *Brucella* antibodies in sheep in indigenous sheep of Jakusko Local
Government Area of Yobe State, with higher seroprevalence in female sheep (12.1%) and in sheep greater than two years (14.6%). The high seroprevalence rates obtained in this study confirmed the presence of this organism the indigenous flocks of sheep and it is of public health concern since people interact with their animals.

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REFERENCES


