



Geo-Spatial Distribution of *Brucella melitensis* Infection in Selected Local Government Areas of Katsina and Sokoto States, Nigeria

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SUMMARY

Brucellosis due to *Brucella melitensis* infects mostly small ruminants and has been reported to be the most invasive and pathogenic species for humans. A cross-sectional sero-geospatial study was conducted in 4 and 3 Local Government Areas (LGAs) of Katsina and Sokoto States, Nigeria respectively. Sera were analysed using c-ELISA while a Global Positioning System (GPS) receiver was used to take geographic coordinates of all sampling points. Data were analysed with SPSS version 20. Chi-square test was used to measure associations among categorical variables. ArcGIS 10.3 was used to map the geospatial pattern of distribution of *B. melitensis*. Three-dimensional analysis was also performed using the Inverse Distance Weighted (IDW) interpolation to determine the distribution pattern of *B. melitensis* in unsampled LGAs. Six (10.52%), 11 (20.0%), 11 (2.0%) and 23 (23.0%) sera were positive from Bakori (n=57), Baure (n=56), Daura (n=55) and Funtua (n=101) LGAs of Katsina State respectively while 4 (12.5%), 1 (5.6%), 13 (14.8%) were positive from Illela (n=22), Tambuwal (n=19) and Yabo (n=90) LGAs respectively in Sokoto State were positive for *B. melitensis* antibodies. All the variables tested were not statistically significant ($p \leq 0.05$). The geospatial maps for both States were produced to show the prevalence of *B. melitensis* using c-ELISA throughout the LGAs sampled and extrapolation was done for other unsampled LGAs. Small ruminants in the study areas harboured antibodies to *B. melitensis*. The maps may serve as an excellent tool for active surveillance and control strategies for livestock diseases like brucellosis in the study area.

Key words: *Brucella melitensis*, Small Ruminants, Nigeria, c-ELISA, GIS

INTRODUCTION

Brucellosis is a disease regarded by the Food and Agriculture Organization (FAO), the Office International des Epizooties (OIE) and the World Health Organization (WHO) as one of the most prevalent zoonoses affecting several developing countries (Elemo and Geresu, 2018). Brucellosis caused by *Brucella melitensis* remains one of the most common zoonotic diseases worldwide and is responsible for about 500,000 reported human cases annually (Saleem *et al.*, 2010). Knowledge of the spatial patterns of brucellosis in livestock could be of immense benefit in the prevention and control of the disease in man and other livestock populations. The Geographic Information System (GIS) as a computer based system captures, stores and links geographically referenced data with graphic map features and allows a wide range of information processing and display operations which include map production, analysis, and modeling (Anenucci *et al.*, 1991). Its application in disease surveillance aimed at prevention and control of health related problems has been in use for several years in developed countries (Haghoost, *et al.*, 2007). In Nigeria, however, its use is still in a rudimentary stage. Though, advancement in molecular biology and genomics has given several sophisticated tools for rapid and confirmatory diagnosis of many diseases, disease surveillance using the GIS through monitoring and networking approaches are much more important for implementing effective prevention and control strategies (Deb and Chakraborty, 2012; Dhama *et al.*, 2013). This technology has proved to be a powerful tool for appropriate and timely responses to disease outbreaks. Its application could be used to locate clusters of zoonotic diseases like brucellosis in order for appropriate control measures to be put in place to prevent its spread. This study was set to determine the sero-prevalences of *B. melitensis* in selected LGAs in Katsina and Sokoto States, Nigeria and also to map the geospatial pattern of distribution of *B. melitensis* in the selected LGAs and predict

the sero-prevalence rates in other LGAs where sampling was not done.

MATERIALS AND METHODS

Study Area: This study was conducted in Katsina and Sokoto States that are located in the North-West geopolitical Zone of Nigeria. Katsina State is located between latitudes 11° 20' and 13° 20' N and longitudes 6° 45' and 8° 15' E. It shares its Northern border with Maradi province in Niger Republic and Kaduna State to the South, Jigawa and Kano States to the East and with Zamfara State to the West (Mohammed and Danjuma, 2014) (Fig. 1). The vegetation of Katsina State is the Sudan Savannah type in the Eastern part and Sahel in the Northern part. The State is made up of 34 Local Government Area (LGAs) and occupies a land mass of about 23,850 square kilometres with a human population of about 5,801,586 (NPC, 2006).

Sokoto State is located between longitudes 11° 30' and 13° 50' E and latitudes 4° and 6° 40' N (Fig.1). It shares common border with the Tahowa Province of Niger Republic to the North, Kebbi State to the West and South, Zamfara State to the East and South (SERC, 2005). The State also falls in the Sudan Savannah in the Southern part and Sahel in the Northern part. The State comprises of 23 LGAs and occupies a land mass of about 25,973 square kilometres with a human population of about 3, 696,999 (NPC, 2006).

A large percentage of the population in these two states is involved in arable farming and livestock rearing as full or part time occupation (NPC, 2006). Annual mean populations of goats and sheep in Katsina and Sokoto States are 2,079,178, 1,696,461, and 2,466,484, 2,566,245 respectively (RIM, 1992).

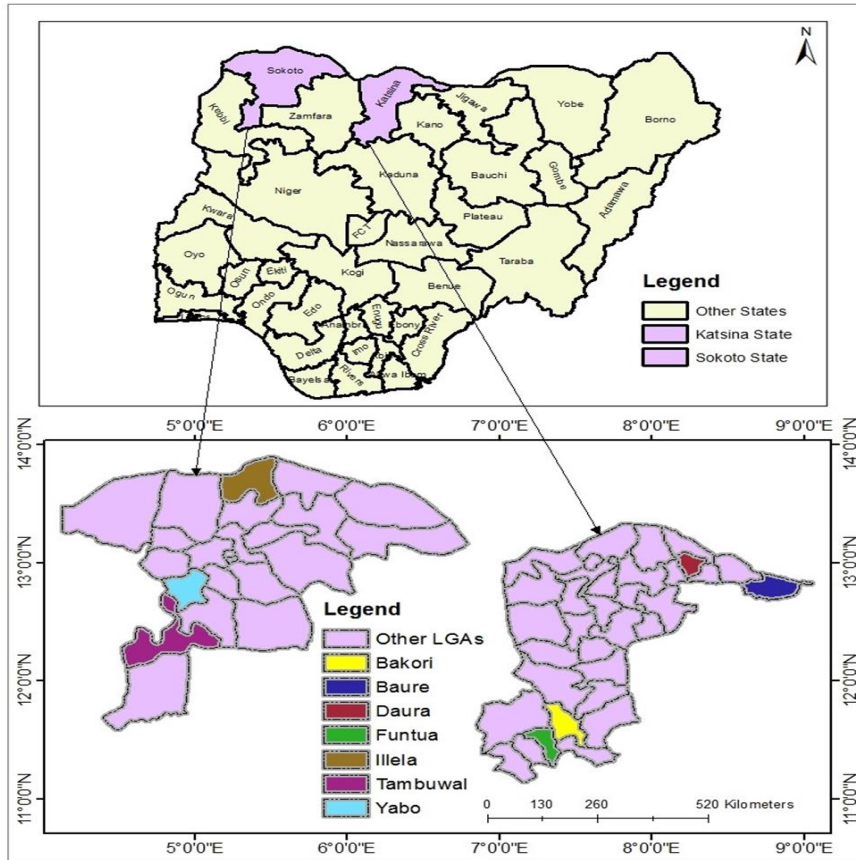


Fig 1: Nigeria showing Sokoto and Katsina States and the LGAs sampled in each of the two States

Study population

Sheep and goats in pastoral holdings were used in this study. The LGAs were selected by random sampling without replacement. Herd selection was also by random sampling and willingness of the farmers to participate in the study. The study involved 269 and 131 small ruminants from Katsina and Sokoto States respectively.

Samples collection

Blood samples collection and handling
 Approximately 5 mL of blood were obtained via a jugular venipuncture from each animal to be sampled using a 10 mL syringe with 21G needle. The blood samples were then transferred into a well-labeled 10 mL plain blood-collecting tubes and placed in a slanting position under shade to allow it to clot. The samples were then transported to the laboratory in leak proof ice-packed

for 5 minutes to allow proper separation of serum from the clotted blood. The serum was then decanted into 5 mL plastic tubes which were properly labelled as for those of the corresponding tubes, after which they were stored in the freezer at -20°C until used.

Geographical Information System

A Global Positioning System (GPS) receiver (*etrex*® Taiwan) was used to take geographic coordinates (latitude and longitude) of all sampling points.

Analysis of Samples

The c-ELISA test kits were sourced from Central Veterinary Laboratory (CVL), Weybridge, Surrey, United Kingdom (U.K.). For the purpose of analysis for *B. melitensis* antibodies using the c-ELISA, 400 serum samples were tested using c-ELISA. The c-ELISA analysis was carried out

according to the manufacturers' instructions and results were read using an ELISA reader (UNIEQUIP® Germany).

These geographic coordinates of the sampling points were arranged in a Microsoft® Excel Spreadsheet and saved as comma separated values (CSV). This was then imported into the ArcGIS 10.3 (Ver.10.3, ESRI Inc., CA, USA) software and then overlaid on the shapefile of the LGAs which is the base map of the study area. The 'symbolology tab' in the ArcGIS was then used to map the distribution of *B. melitensis* prevalence by c-ELISA in the area.

Statistics

Data generated from serological aspects of the study were presented as table 1s. Analysis of data was done with SPSS version 17.0 (2009) where Chi-square (χ^2) and Fishers exact test (FET) were used to test for association among categorical variables. A multi-variable adjusted logistic regression was carried out using all the variables that were statistically significant at the uni-variable analysis. Statistical significance was set at $p \leq 0.05$. All geographical analyses were done using the ArcGIS software version 10.3 and its extension package ArcView Spatial Analyst (Environmental System Research Institute, Inc. Redlands, USA).

RESULTS

A sero-prevalence of 4.0% (51/269) and 13.0% (17/131) were obtained from Katsina and Sokoto States respectively. The difference was not statistically significant ($p > 0.05$) (Table 1).

The highest sero-prevalence rate of 23 (23.0%) was obtained in Funtua LGA while the lowest value of 11 (2.0%) was obtained in Daura LGA. The difference in the prevalence across the LGAs

involved in the study was, however, not statistically significant ($p > 0.05$) (Table 1).

The sero-prevalence of 40 (23.7%) obtained for *B. melitensis* antibodies in sheep was almost double of that obtained in goats 12.1 (12.1%). Here again, the difference was not statistically significant ($p > 0.05$) (Table 1).

In the case of breed, the highest sero-prevalence of 37 (20.4%) was obtained in Kano Brown goats while the lowest value of 0 (0.0%) was obtained

TABLE 1: Sero-prevalence of *B. melitensis* positive sheep and goats using c-ELISA based on State, LGA, species, breed, sex and age in Katsina and Sokoto States, Nigeria

Variable	Level	No. of sera Tested	No. positive (%)	95% C.I on Sero-prevalence	Pearson's Chi-square (χ^2)	P-value
State	Katsina	269	51 (4.0)	3.0 – 5.3	2.239	0.135
	Sokoto	131	17 (13.0)	7.7 – 20.0		
LGA	Bakori	57	6(10.52)	3.9 - 21.5	6.679	0.323
	Baure	56	11 (20.0)	10.2 - 32.4		
	Daura	55	11(2.0)	10.4 - 33.0		
	Funtua	101	23(23.0)	15.0 - 32.1		
	Illela	22	3(14.0)	29.0 - 35.0		
	Tambuwal	19	1(5.2)	0.1 - 26.0		
	Yabo	90	13 (14.4)	8.0 - 23.4		
Species	Sheep	169	40 (23.7)	17.5 – 30.8	0.061	0.804
	Goat	231	28 (12.1)	8.2 – 17.0		
Breed	Kano Brown	181	37 (20.4)	14.8 – 27.1	5.457	0.244
	Sahel (goat)	12	0 (0.0)	0.0 – 26.5		
	Sokoto Red	50	5 (10.0)	3.3 – 21.8		
	Uda	27	4 (14.8)	4.2 – 33.7		
	Yankasa	130	22 (16.9)	10.9- 24.5		
Sex	Female	337	59 (17.5)	13.6 – 22.0	0.385	0.535
	Male	63	9 (14.3)	6.7 – 25.4		
Age Group (Yrs.)*	Young	90	12 (13.3)	7.1 – 22.1	2.248	0.325
	Mature	175	28 (16.0)	10.9 – 22.3		
	Old	135	28 (20.7)	14.3 – 28.6		

* <1 yr. = Young; 1-3.0 yr. = Mature; >3.0 yr. = Old. ($p \leq 0.05$) regarded as significant

in the Sahel breed of goats. Sero-prevalence of 5 (10.0%), 4 (14.8%) and 22 (16.9%) were obtained for Sokoto Red goats, Uda and Yankasa sheep respectively. Similarly, there was no statistical significant difference ($p > 0.05$) between the breeds of animals (TABLE 1).

With regards to the sero-prevalence by sex, a value of 59 (17.5%) obtained for female animals was slightly higher than the 9 (14.3%) obtained in the males. The difference was also not statistically significant ($p > 0.05$) (TABLE 1).

Based on age, the highest sero-prevalence of 28 (20.7%) was obtained for animals aged three years or older while the lowest sero-prevalence of 12 (13.3%) was obtained in animals less than one year old. The difference between the age groups was also not statistically significant ($p > 0.05$) (TABLE 1).

The geospatial map showing the geospatial burden of *B. melitensis* in sheep and goats in LGAs under study in Katsina State is presented in Fig 2. The map for Katsina State showed that various levels of *B. melitensis* infection were present in all of the LGAs sampled (Fig 2). The highest prevalence displayed (red) was located in

LGA, except for a very tiny area in the southern and western parts of Daura LGA (dark green) which had a relatively high prevalence displayed. Also, a map showing the predicted sero-prevalence was produced using Inverse Distance Weighted (IDW) interpolation in other LGAs not sampled in Katsina State is presented in Fig 3. Similarly, the map showing the geospatial burden of *B. melitensis* in sheep and goats in Sokoto LGAs is presented in Fig 4. The map showed that *B. melitensis* infection was also present at various rates in all the LGAs sampled. The highest prevalence (red) was obtained in the North-Eastern part of Illela LGA and majority of Yabo LGA while the least was observed in Eastern part of Tambuwal LGA. The map showing the predicted sero-prevalence based on interpolation in other LGAs in the state which were not part of the sampled areas is shown in Fig 5.

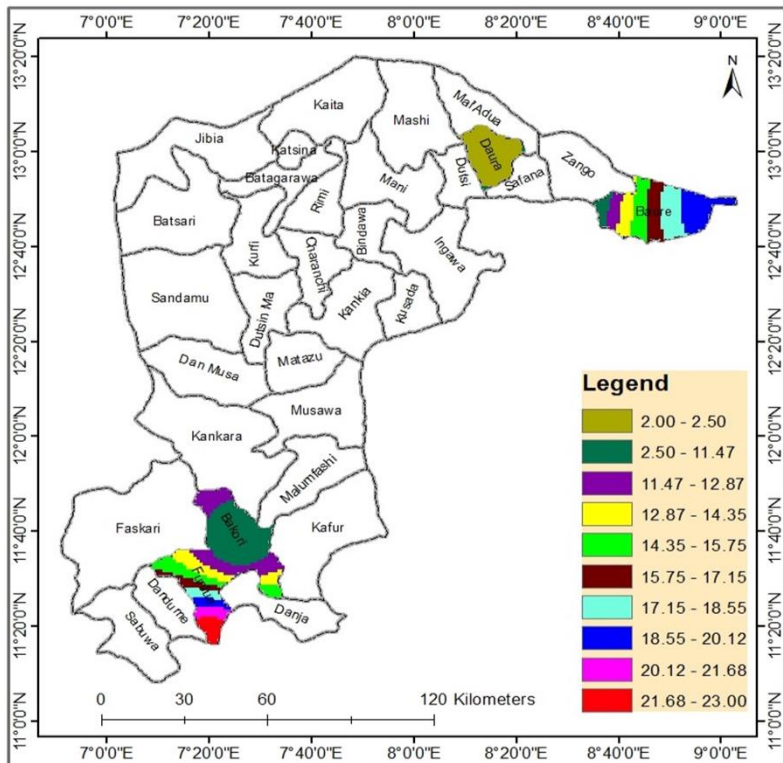


Fig 2: Spatial distribution of *Brucella melitensis* in 4 LGAs in Katsina State

the southern part of Funtua LGA while the lowest prevalence was in majority of Daura

DISCUSSION

In this study, we determined the sero-prevalences of *B. melitensis* in selected LGAs in Katsina and Sokoto States, Nigeria. The sero-prevalence obtained was used to produce GIS maps showing the level of infection in sampled LGAs and also to predict the sero-prevalence rates in other LGAs where sampling was not done. Sero-prevalence of 4.0% and 13.0% were obtained for Katsina and Sokoto States respectively based on c-ELISA. Evidence of *B. melitensis* antibodies in the animals under study may be of public health importance as it is the most pathogenic species for man resulting in long term debilitation (Nasinyama *et al.*, 2014). In most villages in Northern Nigeria, small ruminants make up the bulk of animal protein supply. Apart being routinely consumed,

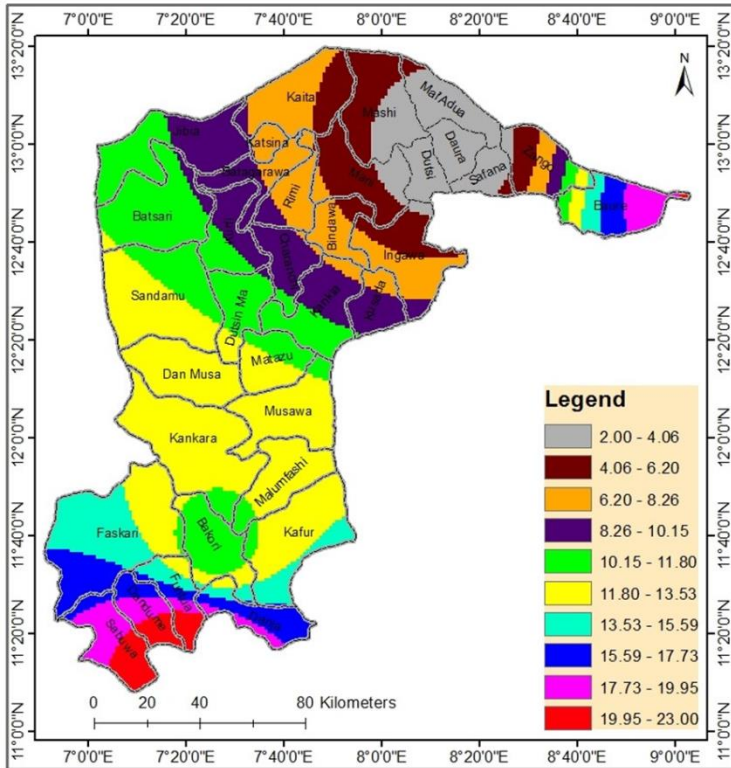
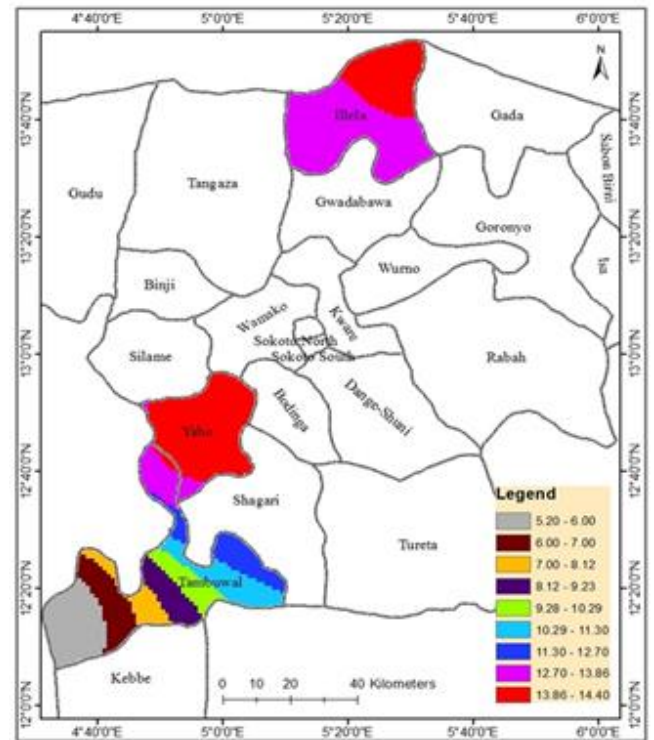


Fig 3: Spatial Interpolation map showing *Brucella melitensis* distribution in all LGAs in Katsina State

their meat are also being processed for delicacies like “balangu and kilishi”. These delicacies may serve as a source of infection as the processing may not completely eliminate *Brucella* organism contained in them. As for in-contact animals, ingestion of the organism could be possible in common grazing fields and watering points during communal grazing as usually practiced in these settings. Infection with this organism may result in great economic loss to the small ruminant production industry as a result of incessant abortions in the herds and also possible infertility. This may consequently lead to a reduction in overall protein supply in affected State and in Nigeria as a whole. There is also the possibility of



spreading the infection to the neighbouring

Fig 4: Spatial distribution of *Brucella melitensis* in 4 LGAs in Sokoto State

Niger republic through the porous borders we share. These findings in this research are quite lower than the overall sero-prevalence of 23.8% obtained by Ya’u (2014) from small ruminants in Bauchi State. It is, however, comparable to the 13.5% obtained in goats in the same Bauchi State by the same author using the same serological test. Nevertheless, Dogo *et al.* (2016) reported a lower sero-prevalence (2.5%) from goats sampled from Giwa LGA in neighbouring Kaduna State. The seeming differences in sero-prevalence in these locations could have something to do with variations in climatic conditions and vegetation in the different locations which could be supporting factors for *Brucella* organisms off the host. This is supported by the reports of Davis and Casey (1973), Bale *et al* (1982) and Ogugua *et al.* (2015) that *Brucella* organisms can survive for durations of time depending on the environmental temperature along with some other factors.

The GIS technique has been found to be a valuable tool that can be used to provide descriptive colour maps of disease burden in various areas and thus can serve as visualized descriptive augmentations to estimated sero-positivity of diseases in pastoralists' herds/villages. The geospatial maps describing *B. melitensis* infected areas in Katsina and Sokoto States as shown in this study can undoubtedly support the surveillance/monitoring and possibly control/eradication programmes for brucellosis in these states. The IDW explicitly makes the assumption that, things that are close to one another are more alike than those that are farther apart. Therefore, measured point has a local influence that diminishes with distance (Li *et al.*, 2014). This technique can be used for prediction of disease prevalence in areas where samples were not collected based on data obtained from some other areas. It thus, could be a very handy tool, especially in low income countries where sampling of a very large area may be very difficult due to lack/limited resources to cater for the logistics involved in sampling and also to purchase the necessary reagents for laboratory analysis of such samples. It can also be of immense value where accessibility to some areas of the country may be difficult or near impossible due to political or geographical hitches as obtained in some African countries. Furthermore, the technology can be used when dealing with highly infectious agents where the researcher may be exposed to the agent by frequent sampling. The results obtained in this study using GIS agrees with Rinaldi *et al.* (2009) and Haghdoost *et al.* (2007) who reported that the GIS is capable of integrating several spatial databases into a single environment for possibilities of improving surveillance and control programmes for infectious diseases and zoonoses. Musa *et al.* (2013) also reported that the most important application of GIS in the field is in the identification of disease clusters. Despite the fact that the technique has been adopted in the study of various zoonoses in the world (Kshirsagar *et al.*, 2013), its application is still in its infancy stage in

Nigeria, especially in the northern part of the country (Alhaji *et al.*, 2016). Amongst the few work done using this technique in Nigeria is the mapping out the burden of contagious bovine pleuropneumonia (CBPP) carried out by Alhaji *et al.* (2016) in north-central Nigeria.

ACKNOWLEDGEMENT

The authors wish to express their appreciation to the Directors Veterinary Services in Katsina and Sokoto States, pastoralists, veterinary personnel from the LGAs involved in this study. Head of Department and laboratory staff, Department of Public Health and Preventive Medicine, Faculty of Veterinary Medicine, Ahmadu Bello University, Zaria, Nigeria.

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