Journal of Biosciences and Medicines, 2021, 9, 105-123
https://www.scirp.org/journal/jbm
ISSN Online: 2327-509X
ISSN Print: 2327-5081

Seroprevalence of *Brucella* Antibodies and Risk Factors Associated with Human Brucellosis in High-Risk Occupational Groups of the Noun Division in the West Region of Cameroon

Rolin Mitterran Ndefo Kamga[1], Barberine Silatsa Assongo[1], Eugenie Melaine Kemta Magang[1], Amadou Fouapon[2], Moussa Salihou[2], Jules-Roger Kuiate[3], Gustave Simo[1]*

[1]Molecular Parasitology & Entomology Unit, Department of Biochemistry, Faculty of Science, University of Dschang, Dschang, Cameroon
[2]Divisional Delegation of Livestock, Fisheries and Animal Industries, Noun, Cameroon
[3]Laboratory of Microbiology and Antimicrobial Substances, Department of Biochemistry, Faculty of Sciences, University of Dschang, Dschang, Cameroon
Email: rolinmitterrank@yahoo.fr, sabarberine@yahoo.fr, eugeniemagang94@yahoo.com, amadoumfouapon@yahoo.com, salihmoussa1@gmail.com, jrkuiate@yahoo.com, *gsimoca@yahoo.fr, *gustave.simo@univ-dschang.org

How to cite this paper: Ndefo Kamga, R.M., Silatsa Assongo, B., Kemta Magang, E.M., Fouapon, A., Salihou, M., Kuiate, J.-R. and Simo, G. (2021) Seroprevalence of *Brucella* Antibodies and Risk Factors Associated with Human Brucellosis in High-Risk Occupational Groups of the Noun Division in the West Region of Cameroon. *Journal of Biosciences and Medicines*, **9**, 105-123.
https://doi.org/10.4236/jbm.2021.93011

Received: January 21, 2021
Accepted: March 28, 2021
Published: March 31, 2021

Abstract

Brucellosis is an anthropozoonotic disease with an important public health impact. Although the transmission of *Brucella* from animals to humans can occur in different epidemiological settings of sub-Saharan African countries, little data has been published on human brucellosis. This study aimed to detect *Brucella* antibodies and the risk factors associated to brucellosis among high-risk occupational groups of people in the Noun Division of Cameroon. For this study, a structured questionnaire was used to assess risk factors associated with human brucellosis. Thereafter, blood samples were collected from high-risk occupational groups of people in four villages. Plasma was extracted from each sample and *Brucella* antibodies were detected using Rose Bengal Plate Test (RBPT) and indirect Enzyme-Linked Immunosorbent Assay (i-ELISA). Of the 273 participants enrolled, the overall seroprevalence of *Brucella* antibodies was 12.45% with RBPT and 10.26% with i-ELISA test. This seroprevalence was significantly ($P = 0.04$; $X^2 = 9.73$) higher among livestock herdsmen (15.8%), slaughterhouse workers (9.8%), butchers (4.8%), participants having no educational level (14.3%) and those experiencing above 5 years of risky activity (15%). Raw milk consumption (*OR*: 4.8; $P = 0.001$), no formal education (*OR*: 6.4; $P = 0.03$) and assistance of animal during parturition (*OR*: 7.2; $P < 0.0001$) appeared as factors that may increase the risk of *Brucella* infections. The detection of *Brucella* antibodies indicates the risk of human brucellosis in some groups of people of the Noun division.

Consuming unpasteurized milk, participating in parturition and lacking knowledge on brucellosis appeared as risk factors associated with human brucellosis in western Cameroon. It raises the need of developing and implementing control measures for human and animal brucellosis.

Keywords

Brucellosis, Risk Factors, High-Risk Occupational Groups, Cameroon

1. Introduction

Brucellosis is an infectious and contagious zoonotic disease classified among the top seven neglected bacterial disease of both humans and animals [1] [2]. It is caused by bacteria of the genus *Brucella*. Different *Brucella* species, including *Brucella abortus*, *Brucella melitensis*, *Brucella suis*, *Brucella ovis* and *Brucella canis*, are responsible for brucellosis in animals and humans [3]. These bacteria are transmitted to humans through the consumption of infected and unpasteurized dairy products or by direct contact with infected animal secretions and excreta [4]. Brucellosis induces important socio-economic impacts and constitutes a serious public health threats for humans and animals. As a neglected zoonotic disease, less attention is given to it by scientists and stakeholders, especially in low-income countries [5].

Brucellosis is one of the major constraints of livestock productivity with significant economic losses estimated yearly to about 427 million US$ in sub-Saharan Africa [1] [6] [7]. Many studies undertaken to generate epidemiological data on brucellosis have been focused on *Brucella* antibodies in various animal species [8] [9] [10] [11] [12]. In these studies, the seroprevalence of *Brucella* antibodies ranges from 3% to 41% [12] [13]. These variations were observed within and between African countries. In Cameroon for instance, Kamga *et al.* (2020) [14] have recently reported some variations in the seroprevalence of *Brucella* antibodies in domestic animals according to agro-ecological zones, highlighting the circulation of *Brucella* species in different agro-ecological settings. In addition, the presence of *Brucella* antibodies in livestock indicates the transmission of *Brucella* to different animal species and probably to humans; pointing out the risk of human brucellosis in Cameroon like in many developing countries. As the risk of human brucellosis depends of the livestock diseases and also the contact frequency between human and livestock, the detection of *Brucella* antibodies in animals of many sub-Saharan countries suggests possible transmission of *Brucella* to humans.

Human brucellosis is characterized by non-specific symptoms such as undulating fever, fatigue, headache, backache, joint pains, musculo-skeletal pains, sweating, arthralgia, malaise and body wasting [15]. These non-specific signs make the clinical diagnosis of brucellosis challenging in sub-Saharan Africa due to misdiagnosis to malaria or other infectious diseases [15] [16]. Globally, an es-

timate of 500,000 new cases and 25,000 deaths due to *Brucella* infections occur every year [17] [18]. Although sporadic cases of brucellosis are often reported in some sub-Saharan African countries, its prevalence may reach 41% in some areas [12] [19] [20] [21]. However, it is important to point out that many human brucellosis cases can pass undetected due to the lack of surveillance program in many sub-Saharan countries [22]. Raising awareness for the design and implementation of control program for humans and animal brucellosis requires generating data on *Brucella* infections in both human and animals. Data already generated on the seroprevalence of *Brucella* antibodies in animals indicate the risk of human brucellosis in many sub-Saharan countries. In Cameroon, little investigations have been undertaken on humans' brucellosis [23]. Investigations performed on abattoir workers of the Adamawa and East regions of Cameroon reported a seroprevalence of *Brucella* antibodies of 12.15% and 28.10% respectively [24] [25]. The difference observed in this seroprevalence suggests that the risk of human brucellosis could vary according to epidemiological settings. Understanding the real epidemiological situation of human brucellosis may require additional studies and data within different epidemiological settings. In this framework, estimating the contact or the seroprevalence of *Brucella* antibodies in humans, especially in people who are regularly in close contact with livestock or derived products, constitutes one step of the process that will help to understand the epidemiological situation of brucellosis in each setting. Moreover, knowledge on the risk factors associated with brucellosis could also help to understand the transmission of *Brucella* and to design control measures for this neglected zoonotic disease.

This study was designed to determine the seroprevalence of *Brucella* antibodies and risk factors associated with *Brucella* infections among high-risk occupational groups of people of the Noun division in the west region of Cameroon.

2. Material and Methods

2.1. Study Area and Study Population

The study population was individuals performing activities that exposed them to the risk of acquiring *Brucella* infections [26] [27]. For this study, the activities considered as high-risk factors for human brucellosis are livestock herdsmen, butchers, slaughterhouse's workers, veterinarians, meat and milk sellers. People performing these activities are considered as high-risk occupational groups (HROG) for brucellosis.

This study was performed in the Noun Division of Cameroon. It is a semi-rural zone with an estimated population of 434,542 inhabitants. It covers about 7687 km^2 with a vegetation characterized by savannah and degraded forest [28]. The sampling was done at Magba (5˚57'00N; 11˚13'00E), Koutaba (5˚42'29N; 10˚41'02), Foumbam (5˚43'N; 10˚55'E) and Massangam (5˚25'29'N; 11˚0'1E) (**Figure 1**). The division has a dense hydrographic network with many streams, rivers and dams rendering the locality a suitable transhumance area for farmers

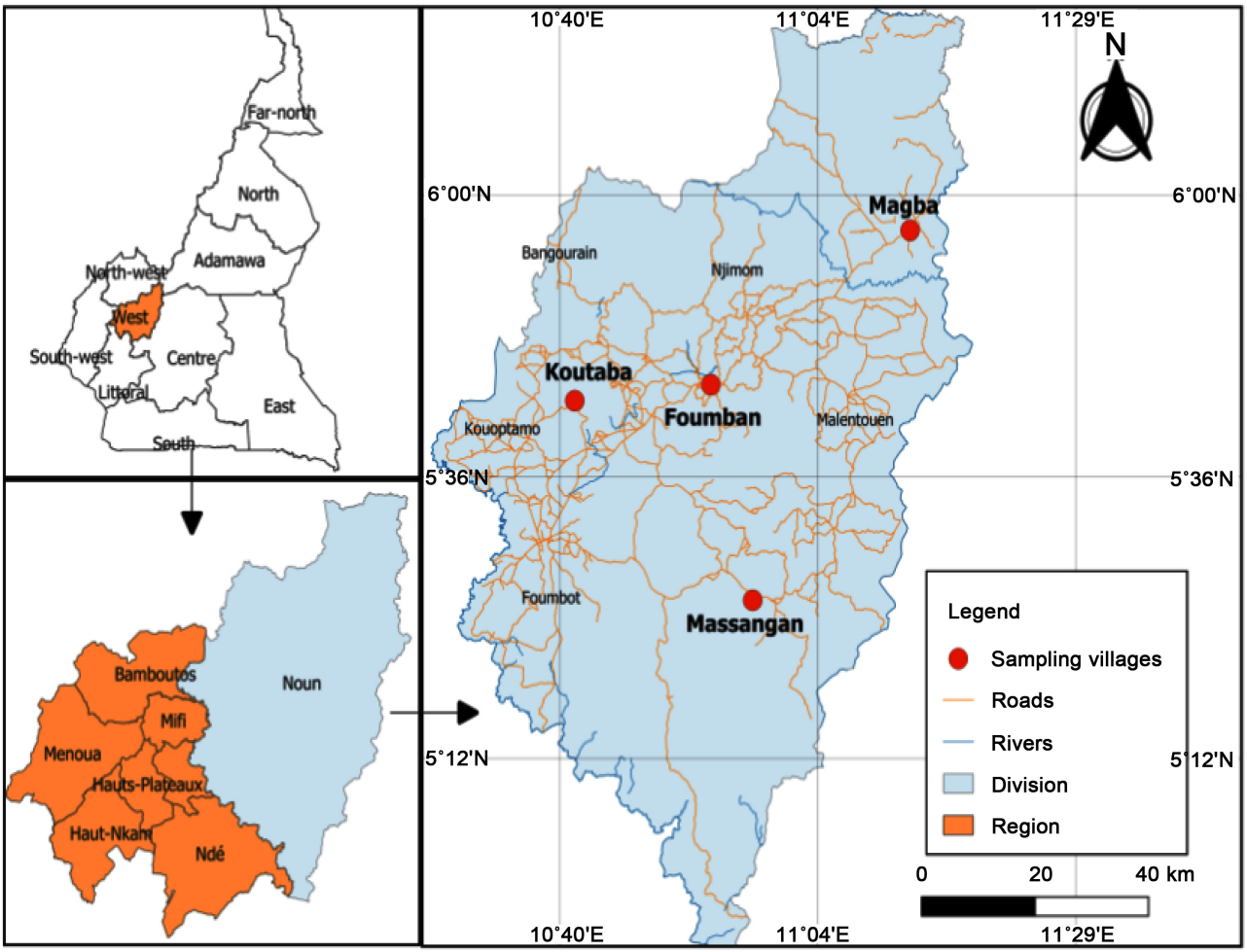

Figure 1. Map of the Noun division showing villages where samples were collected (Map created with QGIS v. 3.8 software (https://www.qgis.org).

during the dry season. Inhabitants of the Noun division practice agriculture, trade, breeding of cattle and small ruminants. In most villages, human/livestock contacts are common. Foumban which is the head-quarter of the Noun Division is the most populous village with an estimated population of 106,309 inhabitants [29]. The main slaughterhouse and cattle market of the Noun division is located at Foumban. Recently, Kamga *et al.* (2020) [14] reported *Brucella* antibodies in several domestic animal species of this locality.

2.2. Ethics Approval and Consent to Participate

The protocol of this study was approved by the Ethical Committee of the Ministry of Public Health of Cameroon of the 23 October 2018 with reference number N˚2018/10/1117/CE/CNERSH/SP. The local administrative and traditional authorities of each sampling site were also informed and gave their approval. Subsequently, the review board of the Molecular Parasitology and Entomology Sub-Unit of the Department of Biochemistry of the Faculty of Science of the University of Dschang gave their approval. Informed consent was written be-

cause each adult enrolled in this study gave its approval by signing an informed consent form and a Certificate of Confidentiality. For all participants below 18 years, the written informed consent was obtained from their parents or guardians. Moreover, a signed assent form was also obtained from participants of 10 to 17 years. During the analyses, data for each participant were anonymized.

2.3. Sampling Size Estimation and Questionnaire Survey

The sample size was estimated using a cross-sectional studies formula as described by [30]:

$$n = Z^2 P_{\exp}\left(1 - P_{\exp}\right) \Big/ L^2$$

where:

n: is the minimum sample size required;

Z: is the critical value for a given confidence interval which is 1.96 at 95% confidence interval;

P: is the expected prevalence;

L: is the margin of error (the margin of error is 0.05).

To estimate this sample size, a human brucellosis prevalence of 12.5% outlined by [24] in abattoir workers of Ngaoundere in the Adamawa region of Cameroon was used. Based on this, the estimated minimum sample size was 165 individuals. However, a total of 273 participants were enrolled based on their cooperation.

2.4. Recording of Socio-Demographic Factors

For each participant who agreed to participate in this study by signing an informed consent form, socio-demographic parameters and other information related to the risk factors that can be associated with brucellosis were recorded using a structured questionnaire adopted from [31] [32] [33] [34]. The collected information's were age, sex, professional activities (butcher, veterinarians, meat seller, farmers and livestock keepers), awareness about brucellosis, assistance during animal parturition, knowledge about zoonotic diseases, consumption of raw milk, educational level and longevity in the profession. The questionnaire was translated as needed to local language (*Bamoun* or *Foufoulde*) in order to minimize bias by the interviewer.

2.5. Blood Samples Collection and Plasma Preparation

For this cross-sectional study carried-out from October to November 2019, 5 ml of blood sample were collected from each selected participant (high-risk occupational groups). This collection was performed by peripheral vein puncture with disposable needles. The Venoject EDTA-coated tubes containing blood sample were labelled and carefully packed. In the field, blood samples were stored at 4˚C in an electric cooler before being transported to the laboratory where they were stored at 4˚C for less than one week before plasma extraction. Plasma was extracted by centrifugation of each blood tube at 8000 xg for 10 min. Thereafter,

500 µl of plasma was collected from each tube and then, transferred into 2 ml sterile micro-tubes that were subsequently stored at −20˚C until use.

2.6. Detection of *Brucella* Antibodies

All plasma samples were accurately tested for *Brucella* antibodies using a combination of two serological tests: the Rose Bengal Plate Test (RBPT) and the indirect Enzyme-Linked Immunosorbent Assay (i-ELISA). The RBPT and the i-ELISA were performed of plasma samples that were stored at −20˚C for less than two months.

2.6.1. Detection of *Brucella* Antibodies by Rose Bengal Plate Test (RBPT)

The detection of *Brucella* antibodies in the plasma was performed with the Rose Bengal Plate Test (RBPT) (ID.Vet, Innovative Diagnostics, France) as described by [35]. Briefly, an aliquot of each plasma sample as well as the RBPT reagents were allowed to thaw at room temperature (22˚C ± 4˚C) for approximately 25 minutes as recommended by the manufacturer. Thereafter, 30 µL of plasma and an equal volume of RBPT antigen were put in each circle of the RBPT plate and then mixed. After 4 minutes of rocking the plate at room temperature, the plate was observed by unaided eyes to see if there is any agglutination. Any visible agglutination on the plate by unaided eyes was considered positive; this means that the plasma sample contains *Brucella* antibodies [36]. If no agglutination was observed, the test was considered negative (no antibodies against *Brucella* in the plasma tested).

2.6.2. Detection of *Brucella* Antibodies by Indirect Enzyme-Linked Immunosorbent Assay (i-ELISA)

The i-ELISA test was performed to confirm RBPT results. The i-ELISA tests were performed in a polystyrene plate of 96-wells pre-coated with purified antigens of *Brucella* spp. Plasma samples were tested for the presence of antibodies against *Brucella* spp using multi species commercial indirect ELISA test kit (ID-Screen Brucellosis Serum Indirect Multispecies, ID VET, product code BRUS-MS-1014, Gabrels, France) [37]. The tests were performed according to the instructions of the manufacturer (ID-VET, 2008). Before each i-ELISA test, reagents and plasma samples were allowed to thaw at room temperature (22˚C ± 4˚C) and 100 µL of diluted buffer were added to each well. Ten (10 µl) micro-liters of positive control and equal volume of negative control provided by the manufacturer were introduced into two different wells of the plate and 10 µL of each plasma sample were introduced in the remaining wells. Each plate was sealed and manually homogenized gently. After incubation at room temperature for 45 minutes, each plate was washed 3 times with PBS-Tween and 100 µL of multispecies horseradish peroxidase (HRP) conjugate were added to each well. Each plate was subsequently incubated for 30 minutes at room temperature and washed 3 times to eliminate the excess of conjugate. Thereafter, 100 µL of the substrate solution (tetramethylbenzidine in substrate buffer containing H_2O_2)

were added to each well and the plate was incubated in the dark for 15 minutes at room temperature. The reaction was stopped by adding 100 µL of 1 N hydrochloric acid (HCl). The optical density in each well was measured at 450 nm using a micro plate photometer (Bio Tek ELX800 absorbance reader). For each tested sample, its result was expressed as a percentage of optical density (%OD) that was calculated using the following formula:

$$\%\mathrm{OD} = 100 \times (S - N) / (P - N)$$

where S, N and P are ODs of the sample, the negative and positive controls respectively. Sample with a % OD ≥ 120% where considered positive.

2.7. Data Analysis

The Statistical Package for Social Sciences (SPSS), version 21.0, software package was used to perform the statistical analysis of all collected data. Pearson chi-square test was used to compare the seroprevalence of *Brucella* antibodies between villages, activities performed by participants, sex, age, longevity in the activity and educational school level of the study population. Logistic regression model was used to determine which risk factor is associated with the presence of *Brucella* antibodies among the study participants. Odds ratio (OR) and 95% Confidence interval (CI) were noted. A p-value below 0.05 was considered as statistically significant.

3. Results

3.1. Socio-Demographic Characteristics of the Study Population

Of the 273 participants enrolled in this study, 186 (68.13%) were from Foumban, 54 (19.8%) from Koutaba, 21 (7.7%) from Magba and 12 (4.4%) from Massangam (Table 1). Among these participants, 101 (36.99%) were livestock keepers/famers, 84 (30.76%) butchers, 61 (22.34%) slaughterhouse workers, 11 (4.03%) veterinarians and 16 (%) meat and milk sellers (Table 1). Sixteen (5.8%) of these participants were female while 257 (94.1%) were males. The mean age of participants was 38.5 ± 14.45 years (inter quartile range (IQR): 15 - 73 years). One hundred and fifty-nine (58.2%) participants attended the primary education, 79 (28.9%) the secondary education and 35 (12.8%) did not have any formal education (Table 1). Two hundred and nine (79.6%) of these participants has expended over 5 years in their occupation (Table 1).

3.2. Prevalence of *Brucella* Antibodies According to Villages

Figure 2 illustrates results of RBPT where some plasma samples generated agglutinations and other no agglutination. The overall seroprevalence of *Brucella* antibodies revealed with RBPT and i-ELISA tests was 12.5% (95% CI: 8.6 - 17.4) and 10.3% (95% CI: 6.8 - 14.8) respectively. For subsequent analyses, only participants that were positive for both RBPT and i-ELISA were considered as have been in contact with *Brucella* or have *Brucella* antibodies. On this basis, the

Table 1. Prevalence of *Brucella* antibodies and bivariate analysis of factors associated with human brucellosis amongst the study participants.

			BRUCELLA ANTIBODIES						BIVARIATE ANALYSIS	
VARIABLE	DESCRIPTION	N (%)	**RBPT+ (%)**	**95% CI**	**i-ELISA+ (%)**	**95% CI**	**Number of samples positive for both RBPT and i-ELISA (%)**	**95% CI**	**OR (95% CI)**	**P-Value**
LOCALITY	Foumban	186 (68.1)	23 (12.7)	0.6 - 24.6	18 (9.7)	4.1 - 21.5	17 (9.1)	0.05 - 14.6	1.1 (0.1 - 9.1)	0.92
	Magba	21 (7.7)	3 (14.3)	2.9 - 41.7	3 (14.3)	2.9 - 41.7	3 (14.3)	2.95 - 41.7	1.9 (0.1 - 21.1)	0.54
	Koutaba	54 (19.8)	7 (12.9)	5.2 - 26.7	6 (11.1)	4.1 - 24.2	5 (9.3)	0.03 - 21.6	1.1(0.1 - 10.6)	0.91
	Massangam	12 (4.4)	1 (8.3)	0.2 - 46.4	1 (8.3)	0.2 - 46.4	1 (8.3)	0.21 - 46.4	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				0.91		0.60			
	P-Value				0.92		0.89			
SEX	Female	16 (5.8)	0 (0.0)	NA	0 (0.00)	NA	0 (0.0)	NA	-	
	Male	257 (94.1)	34 (13.2)	9.3 - 18.5	28 (10.9)	7.2 - 15.7	26 (10.1)	6.6 - 14.8	3.8 (0.2 - 64.8)	0.35
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				1.94		9.73			
	P-Value				0.16		0.04*			
AGE	<20	27 (9.89)	2 (7.4)	0.8 - 26.7	2 (7.4)	0.9 - 26.7	2 (7.4)	0.9 - 26.7	-	
	21 - 30	73 (26.7)	9 (12.3)	5.6 - 2	7 (9.6)	3.8 - 19.7	6 (8.2)	3.0 - 17.9	1.1 (0.2 - 5.9)	0.9
	31 - 40	65 (23.8)	6 (9.2)	3.4 - 20.1	5 (7.7)	2.5 - 17.9	5 (7.7)	2.5 - 17.9	1.0 (0.1 - 5.7)	0.96
	>40	108 (39.5)	17 (15.7)	9.2 - 25.2	14 (12.9)	7.1 - 21.7	13 (12.0)	6.4 - 20.6	1.7 (0.4 - 8.1)	0.49
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				1.33		1.33			
	P-Value				0.66		0.18			
PROFESSIONAL ACTIVITIES	slaughterhouse workers	61 (22.3)	8 (13.1)	5.6 - 24.8	6 (9.8)	6 (9.8)	3.6 - 21.4	3.6 - 21.4	3.3 (0.2 - 62.3)	0.42
	livestock keepers/ herdsmen	101 (36.9)	19 (18.8)	11.3 - 29.4	17 (16.8)	16 (15.8)	9.0 - 25.7	9.0 - 25.7	4.4 (0.2 - 79.0)	0.31
	butchers	84 (30.7)	7 (8.3)	3.3 - 17.2	5 (5.9)	4 (4.8)	1.3 - 12.2	1.3 - 12.2	1.3 (0.1 - 25.5)	0.87
	veterinarians	11 (4.02)	0 (0.0)	NA	0 (0.0)	0 (0.0)	NA	NA	-	
	meat or milk sellers	16 (5.8)	0 (0.0)	NA	0 (0.0)	0 (0.0)	NA	NA	0.69 (0.0 - 37.7)	0.85
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				9.53		1.33			
	P-Value				0.04*		0.72			

Continued

LEVEL OF EDUCATION	None	35 (12.8)	8 (22.9)	9.9 - 45.0	5 (14.2)	4.6 - 33.3	5 (14.3)	4.6 - 33.3	6.4 (1.2 - 34.9)	0.03*
	Primary	159 (58.2)	22 (13.8)	8.7 - 20.9	21 (13.2)	8.2 - 20.2	19 (11.9)	7.2 - 18.7	5.8 (1.33 - 25.31)	0.02*
	Secondary	79 (28.9)	4 (5.1)	1.4 - 12.9	2 (2.5)	0.3 - 9.1	2 (2.5)	0.3 - 9.1	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				6.85		6.48			
	P-Value				0.03*		0.04*			
DURATION IN THE PROFESSIONAL ACTIVITY (YEARS)	1 - 5	64 (23.44)	4 (6.2)	1.7 - 16.0	3 (4.7)	0.9 - 13.7	3 (4.7)	0.9 - 13.7	-	
	5 - 10	89 (32.6)	9 (10.1)	4.6 - 19.2	6 (6.7)	2.5 - 14.7	5 (5.6)	1.8 - 13.1	1.2 (0.2 - 5)	0.8
	>10	120 (43.9)	21 (17.5)	10.8 - 26.7	19 (15.8)	9.5 - 24.7	18 (15)	8.9 - 23.7	7.3 (2.0 - 25.9)	0.002*
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)		26 (9.5)	6.2 - 13.9		
	X²				5.99		7.48			
	P-Value				0.02*		0.02*			
KNOWLEDGE ON ZOONOTIC DISEASES	Yes	79 (28.9)	5 (6.3)	2.0 - 14.8	2 (2.5)	0.3 - 9.1	2 (2.5)	0.3 - 9.1	0.2 (0.04 - 0.7)	0.02*
	No	194 (71.06)	29 (14.9)	10.1 - 21.4	26 (13.4)	8.8 - 19.6	24 (12.4)	7.9 - 18.4	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				19.9		6.32			
	P-Value				<0.0001*		0.02*			
AWARENESS ON ZOONOTIC BRUCELLOSIS	Yes	34 (12.45)	3 (8.8)	1.8 - 25.8	2 (5.9)	0.7 - 21.2	1 (2.9)	0.0 - 16.4	-	
	No	239 (87.54)	31 (12.9)	8.8 - 18.4	26 (10.8)	7.1 - 15.9	25 (10.5)	6.8 - 15.4	3.8 (0.5 - 29.4)	0.19
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				0.77		1.9			
	P-Value				0.3		0.15			
USE OF PERSONAL PROTECTIVE EQUIPMENT	Yes	84 (30.76)	6 (7.1)	2.6 - 15.5	4 (4.8)	1.3 - 12.1	3 (3.8)	0.7 - 10.4	0.26 (0.1 - 0.9)	0.03*
	No	189 (69.23)	28 (14.8)	9.8 - 21.4	24 (12.7)	8.1 - 18.9	23 (12.2)	7.7 - 18.3	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				3.9		4.7			
	P-Value				0.04*		0.03*			
OBSERVATION OF HYGIENE MEASURES	Yes	227 (83.15)	20 (8.8)	5.9 - 13.6	17 (7.5)	4.4 - 11.9	16 (7.0)	4.0 - 11.4	0.4 (0.2 - 1.0)	0.004*
	No	46 (16.84)	14 (30.4)	16.6 - 51.0	11 (23.9)	11.9 - 42.8	10 (21.7)	10.4 - 39.9	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				11.12		9.59			
	P-Value				0.008*		0.001*			
CONSUMPTION OF RAW MILK	Yes	121 (44.32)	26 (21.5)	14.0 - 31.5	22 (18.18)	11.4 - 27.5	20 (16.5)	10.1 - 25.5	4.8 (1.9 - 12.1)	0.001*
	No	152 (55.67)	8 (5.3)	22.7 - 10.3	6 (3.9)	1.5 - 8.6	6 (3.9)	1.5 - 8.6	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X²				14.9		12.4			
	P-Value				0.0001		0.004*			

Continued

ASSISTANCE DURING PARTURITION	Yes	112 (41.02)	24 (21.43)	13.7 - 31.9	21 (18.7)	11.6 - 28.7	21 (18.7)	11.6 - 28.7	7.2 (2.6-19.7)	0.0001*
	No	161 (58.97)	10 (6.2)	2.9 - 11.4	7 (4.34)	1.7 - 8.9	5 (1.9)	1.0 - 7.2	-	
	Total	273	34 (12.5)	8.6 - 17.4	28 (10.3)	6.8 - 14.8	26 (9.5)	6.2 - 13.9		
	X^2				14.77		23.16			
	P-Value				0.0001		< 0.0001*			

N: Number of human samples tested; OR: odds ratio; CI: confident interval; i-ELISA: indirect Enzyme-Linked Immunosorbent Assay; *significant *P-value*.

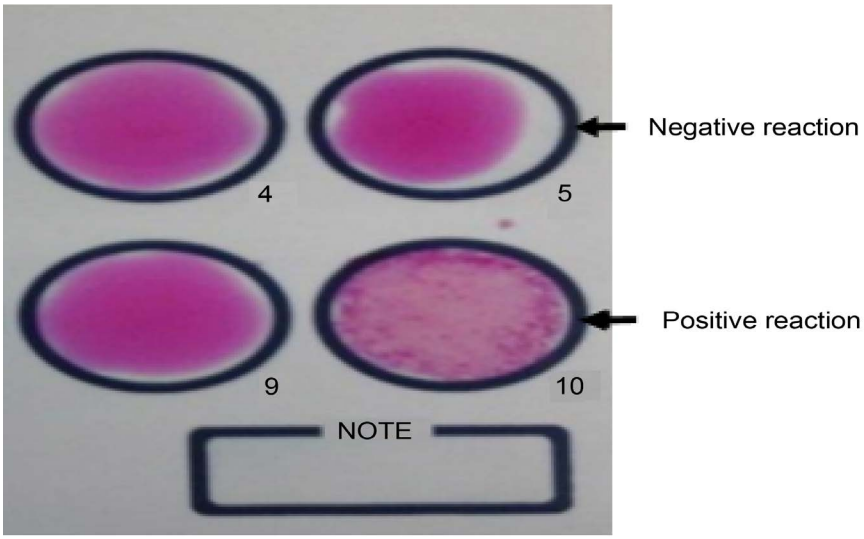

Figure 2. Example of Rose Bengal plate Test presenting result obtained after assay of plasma samples: 4, 5 and 9 are spots showing no agglutination reaction (plasma samples that were negative or do not contain *Brucella* antibodies); 10: spot showing an agglutination reaction (plasma sample that was positive or contains *Brucella* antibodies).

overall seroprevalence of *Brucella* antibodies was 9.5%. The highest seroprevalence of *Brucella* antibodies of 14.3% was recorded in Magba following by Koutaba with 9.5% and Foumban with 9.1%. Massangam had the lowest prevalence of 8.3% (Table 1). No significant difference ($P = 0.60$; $X^2 = 0.89$) was observed when comparing the seroprevalence of *Brucella* antibodies between villages (Table 1). Moreover, the OR ranges from 1.1 (95% CI: 0.1 - 10.6) at Koutaba to 1.9 (95% CI: 0.1 - 21.1) at Magba. Whatever the village, no *P value* was statistically significant (Table 1). This indicates that belonging to any village cannot be considered as risk factors of having *Brucella* infections.

3.3. Prevalence of *Brucella* Antibodies According to Participants' Activities

The seroprevalence of *Brucella* antibodies in slaughterhouse workers, livestock keepers/herdsmen and butchers were respectively 9.8%, 15.8% and 4.8%. No participant belonging to veterinary staff, meat and milk sellers was found with *Brucella* antibodies (Table 1). Comparing the seroprevalence of *Brucella* antibodies according to activities performed by participants, significant difference ($P =$

0.04; X^2 = 9.73) was observed. The ORs were high for livestock keepers/herdsmen (OR = 4.4; 95% CI: 0.2 - 79.0) and slaughterhouse workers (OR = 3.3; 95% CI: 00.2 - 62.3). For butchers and meat or milk sellers, the values of ORs were respectively 1.3 (95% CI: 0.1 - 25.5) and 0.69 (0.0 - 37.7). Whatever the activity, the *P-value* was not significant and consequently, no association can be inferred regarding the activities performed by the participants (Table 1).

3.4. Prevalence of *Brucella* Antibodies According to Other Demographic Factors and Risk Factors Associated with *Brucella* Infections

The factors considered here include the sex of the participants, age, educational level, longevity in the activity and risky behaviors of the participants. No female out of 16 was found with *Brucella* antibodies while 26 males were reported with *Brucella* antibodies. This gives a seroprevalence of 10.1% in males (Table 1). Between males and females, no significant difference (P = 0.18; X^2 = 1.78) was observed in the seroprevalence of *Brucella* antibodies. The OR of 3.8 [95% CI: 0.2 - 64.8] with a *P-value* of 0.35 (Table 1) indicates no significant association between sex and the presence of *Brucella* antibodies.

Among the 273 participants, 27 (9.9%) had less than 20 years, 73 (26.7%) were between 21 to 30 years, 65 (23.8%) between 31 to 40 years and 108 (39.6%) above 40 years (Table 1). The highest seroprevalence (12%) of *Brucella* antibodies was observed in participants above 40 years while the lowest seroprevalence (7.2%) was observed in those of less than 20 years (Table 1). Comparing the seroprevalence of *Brucella* antibodies between age groups, no significant difference (P = 0.72; X^2 = 1.33) was observed (Table 1). The ORs vary from 1.0 (95% CI: 0.1 - 5.7) in participants of 31 - 40 years to 1.7 (95% CI: 0.4 - 8.1) in those above 40 years. However, no significant association was found between age groups and the risk to be in contact *Brucella* infections (Table 1).

Looking at the educational level, 12.8% of participants did not attend any formal education while 58.2% attended the primary education and 28.9% the secondary education. The seroprevalence of *Brucella* antibodies was significantly higher (P = 0.04; X^2 = 6.48) in participants without any formal education (14.3%) and those with a primary education (11.9%) compared to those with secondary education (2.5%) (Table 1). The risk to be in contact with *Brucella* seems to significantly increase in participants with no formal education [OR = 6.4 (95% CI: 1.2 - 34.9); P = 0.03] and those with primary education [OR = 5.8; (95% CI: 1.3 - 25.3); P = 0.02].

Of the 273 participants, 64 (12.4%), 89 (32.6%) and 120 (43.9%) had respectively less than 5 years, 5 to 10 years and above 10 years of experience in their professional activities. The highest seroprevalence of *Brucella* antibodies of 15% was recorded in participants with more than 10 years of experience and the lowest seroprevalence of 4.7% in those with less than 5 years of experience. Comparing the seroprevalence of *Brucella* antibodies between groups of participants with different duration in their professional activities, significantly difference (P

= 0.02; X^2 = 7.48) was observed (Table 1). Performing risky activities for more than 10 years was found to be significantly associated [OR = 7.3; (95% CI: 2.0 - 25.9); P = 0.002] with the risk to be in contact with *Brucella* (Table 1).

One hundred and twenty-one (44.3%) participants consume raw milk. For this group of participants, the overall seroprevalence of *Brucella* antibodies of 16.5% was significantly higher (P = 0.004, X^2 = 12.40) compared to 3.9% reported in people who did not consume raw milk (Table 1). The OR of 4.8 (95% CI: 1.9 - 12.1) with a significant P-value of 0.001 indicates that consumption of raw milk may increase the risk to be in contact with *Brucella* infections.

For participants with and without awareness of zoonotic diseases, the seroprevalence of *Brucella* antibodies were respectively 2.5% and 12.4%. Comparing the seroprevalence of *Brucella* antibodies between the two groups of participants, significant difference (P = 0.01; X^2 = 6.32) was observed (Table 1). An OR of 0.2 (95% CI: 0.04 - 0.7) with a significant *P value* of 0.02 were obtained in participants having knowledge on zoonotic diseases (Table 1). Having such knowledge seems to reduce the risk of contracting *Brucella* infections.

In participants adopting or not the preventive measures by using protective equipment, the seroprevalence of *Brucella* antibodies were respectively 3.8% and 12.2% (Table 1). Between these two groups of participants, the difference in their seroprevalence was statistically significantly (P = 0.03; X^2 = 4.70). Moreover, for participants observing or not the hygiene measures, the seroprevalence of *Brucella* antibodies were respectively 7% and 21.7%. Significant difference (P = 0.001; X^2 = 9.59) was observed between these two groups of participants (Table 1). For participants who observe hygiene measures and wearing protective equipment, the ORs respectively of 0.4 (95% CI: 0.2 - 1.0) and 0.2 (95% CI: 0.1 - 0.9) with significant P-values of 0.03 and 0.004 indicates that observing such measures may reduce the risk of contracting *Brucella* infections (Table 1).

Amongst groups of participants usually assisting or not animals during parturition, the seroprevalence of 18.7% obtained in those assisting was significantly higher (P = 0.004, X^2 = 12.40) compared to 1.9% recorded in other groups (Table 1). The OR of 7.2 (95% CI: 2.6 - 19.7) with a significant P-value of 0.0001 indicates that wearing protective equipment may decrease the risk to be in contact with *Brucella* infections (Table 1).

4. Discussion

For this study, the high number of participants enrolled at Foumban could be explained not only by the presence of the main slaughterhouse and several farms around this village, but also the largest cattle market of the West region. Compared to other villages, several animals are sold and killed at Foumban because it is the capital with the largest population of the Noun division. In such context, several inhabitants of Foumban practice activities related to livestock and their derived products. In other villages, the population is of small size and consequently, few animals are killed per week. In addition to that, the number of in-

habitants practicing risky activities for human brucellosis is limited.

The overall seroprevalence of *Brucella* antibodies of 9.5% obtained in this study is consistent with 10.0% and 12.96% reported respectively in Uganda [38] and in the Adamawa region of Cameroon [24]. Although this seroprevalence is lower than 48.8% and 24.1% reported respectively in Nigeria [32] and Tanzania [34], it is higher than 4.7% reported in Ethiopia [33]. The observed variations could be due to the levels of exposure of each study population, the presence or absence of control program for brucellosis, the differences in the study design, the studied populations or the groups at risk that were enrolled in different studies and the participant's behavior (eating habits and cultural practices) [24] [39]. Indeed, previous studies targeted the abattoir workers and some febrile patients [25] [32] [33] [34] while our study included more groups of people practicing activities that exposed them to *Brucella* infections. Known as zoonotic disease, it is important to point out that the situation of brucellosis in humans may reflect what happens in animals. In this light, our recent investigations on animal brucellosis revealed the circulation of *Brucella* antibodies in cattle and small ruminants of the same locality [14]. Data of the present study with those generated on animals suggest a probable transmission of *Brucella* between animals and humans. This hypothesis is plausible because cattle and small ruminant are recognized as the main sources for human brucellosis [6] [10] [40]. Therefore, for a better understanding of the transmission dynamics of *Brucella* and the current epidemiological situation of brucellosis in the affected areas, further investigations aiming to isolate and molecularly characterize *Brucella* strains are needed to identify *Brucella* species circulating in humans and animals of this locality.

The present study revealed a significant higher seroprevalence of *Brucella* antibodies for some socio-demographic factors like professional activities, level education and the longevity in some risky activities. These results are in agreement with previous ones [32] [34] [41]. The high seroprevalence of *Brucella*antibodies in livestock keepers/herdsmen could be explained by the fact that they have several levels of expositions. For instance, they routinely consume unpasteurized milk when rearing animals. During calving and ticks picking, they are in close contact with infected animals and can easily become infected from these animals. During slaughter process, slaughterhouse workers are permanently in contact with animal discharges such as fesses, urines, fetuses. Such contacts exposed them to the risk of acquiring *Brucella* infections [41] [42]. Although veterinarians as well as meat and milk sellers are regularly in contact with animals and derived products, none of them were found with *Brucella* antibodies. This could be explained by the fact that they have been sensitized against brucellosis and other zoonotic diseases. They have therefore enough knowledge about the transmission and prevention of this disease [33]. These hypotheses are strengthened by our results of association studies reporting reduced risks for brucellosis in people having knowledge on brucellosis. The high seroprevalence reported

in people practicing risky activities for more than 5 years could be explained by their permanent and longtime exposition to animals [32] [33].

Although no woman was found with *Brucella* antibodies, no significant difference was observed in the seroprevalence of *Brucella* antibodies between males and females. Our results are in agreement with those of Tsegay *et al.* (2017) [33]. They could be explained by the fact that the low sample size of women. In addition, the risky activities (selling milk or cooked meat) performed by women have been reported in our study to be of lower risk for brucellosis. The higher prevalence of *Brucella* antibodies in participants of more than 40 years could be explained by the fact that most of them have been performing the risky activities for several years. This hypothesis is in line with results of association studies reporting high risk of *Brucella* infections in participants practicing risky activities for long time.

Our results of bivariate analyses revealed that the consumption of unpasteurized milk and animal assistance during parturition seem to increase the risk of getting *Brucella* infections. These results are in agreement with those of Rubach *et al.* (2013) [43]. They could be explained by the fact that *Brucella* are mainly releasing from animal body through the milk, fetal fluids and semen. Moreover, milk and fetal fluid contain growth factors for *Brucella* spp [44]. As semen, milk and fetal fluids constitute the main sources for *Brucella* contamination, individuals who are regularly in close contact with these fluids by consuming raw milk or assisting animals during the delivery have been reported to have higher risk of contracting brucellosis [8] [45] [46]. In addition, manipulating infected materials such as carcasses, viscera, organs, blood and urine have been considered as practices enhancing the risk for the transmission of brucellosis [47]. Although the pasteurization of milk is well known to reduce the infectivity of *Brucella* and to limit the transmission of *Brucella* to humans [43], this treatment cannot be done in rural settings where equipment for pasteurization is inexistent. In such settings, boiling or heating the milk for several minutes could reduce the infectivity of *Brucella* [48] [49] [50].

Our results of bivariable logistic regressions analysis showed that observing personal hygiene measures and adopting safety practices such as wearing protective equipment and disinfection of premises during parturition may reduce the risk to be infected by *Brucella*. These results are in agreement with those of previous studies [50] [51] [52].

Although results of this study cannot be generalized to the entire population, they highlight the need of designing appropriate control measures at least for these restricted groups of people. Since the vaccination is not generally recommended for the management of human brucellosis [53] [54], the control strategies to address *Brucella* infections must include education and sensitization of the population, especially those for whom their daily activities are directly linked to livestock and their derivatives. For preventive measures, it will be important to avoid the consumption of raw milk or products made from raw milk. Putting together results of this study with those generated on animals of the same locali-

ty, the designing and the implementation of efficient control program for human and animal brucellosis requires the "One Health approach". In such approach, all actors involved in livestock and human health must join their efforts for sustainable control of brucellosis. Implementing the One health concept could reduce the impact of brucellosis in order to improve human and animal health.

5. Conclusion

This study revealed *Brucella* antibodies in different groups of people practicing activities exposing them to brucellosis. The seroprevalence of *Brucella* antibodies was higher in participants with low educational level and those practicing risky activities for above 5 years. Consuming unpasteurized milk, participating to parturition and lacking knowledge on brucellosis were associated with the risk of getting human brucellosis in the Noun division of Cameroon. Results of this study suggest the need to develop and implement control measures for both human and animal brucellosis.

Acknowledgements

We wish to acknowledge all the participants of this study for their cooperation. We also thank the anonymous reviewers for helpful comments on the manuscript.

Competing Interests

The authors declare no competing interests.

References

[1] Chikeka, I. and Dumler, J.S. (2015) Neglected Bacterial Zoonoses. *Clinical Microbiology and Infection*, **21**, 404-415. https://doi.org/10.1016/j.cmi.2015.04.022

[2] Boukary, A.R., Saegerman, C., Abatih, E., Fretin, D., Alambédji Bada, R., De Deken, R., Harouna, H.A., Yenikoye, A. and Thys, E. (2013) Seroprevalence and Potential Risk Factors for *Brucella* Spp. Infection in Traditional Cattle, Sheep and Goats Reared in Urban, Periurban and Rural Areas of Niger. *PLoS ONE*, **8**, e83175. https://doi.org/10.1371/journal.pone.0083175

[3] Hull, N.C. and Schumaker, B.A. (2018) Comparisons of Brucellosis between Human and Veterinary Medicine. *Infection Ecology & Epidemiology*, **8**, Article ID: 1500846. https://doi.org/10.1080/20008686.2018.1500846

[4] Lado, D., Maina, N., Lado, M., Abade, A., Amwayi, S., Omolo, J. and Oundo, J. (2012) Brucellosis in Terekeka County, Central Equatoria State, Southern Sudan. *East African Medical Journal*, **89**, 28-33.

[5] Ayayi, J.A., Têko-Agbo, A. and Koné, P. (2009) The Impact of Brucellosis on the Economy and Public Health in Africa. *Conference OIE*, Paris, 12-14 October 2009, 85-98.

[6] Seleem, M.N., Boyle, S.M. and Sriranganathan, N. (2010) Brucellosis: A Re-Emerging Zoonosis. *Veterinary Microbiology*, **140**, 392-398. https://doi.org/10.1016/j.vetmic.2009.06.021

[7] Mangen, M.J., Otte, J., Pfeiffer, D. and Chilonda, P. (2002) Bovine Brucellosis in

Sub-Saharan Africa: Estimation of Seroprevalence and Impact on Meat and Milk Off-Take Potential. Food and Agriculture Organisation of the United Nations, Rome.

[8] Madut, N.A., Muwonge, A., Nasinyama, G.W., Muma, J.B., Godfroid, J., Jubara, A.S., Muleme, J. and Kankya, C. (2018) The Sero-Prevalence of Brucellosis in Cattle and Their Herders in Bahr el Ghazal Region, South Sudan. *PLOS Neglected Tropical Diseases*, **12**, e0006456. https://doi.org/10.1371/journal.pntd.0006456

[9] Awah-Ndukum, J., Mouiche, M.M., Bayang, H.N., Ngu Ngwa, V., Assana, E., Feussom, K.J.M., Manchang, T.K. and Zoli, P.A. (2018) Factors of Brucellosis among Indigenous Cattle in the Adamawa and North Regions of Cameroon. *Veterinary Medicine International*, **2018**, Article ID: 3468596. https://doi.org/10.1155/2018/3468596

[10] Ducrotoy, M., Bertu, W.J., Matope, G., Cadmus, S., Conde-Álvarez, R., Gusi, A.M., Welburn, S., Ocholi, R., Blasco, J.M. and Moriyón, I. (2017) Brucellosis in Sub-Saharan Africa: Current Challenges for Management, Diagnosis and Control. *Acta Tropica*, **165**, 179-193. https://doi.org/10.1016/j.actatropica.2015.10.023

[11] Assenga, J.A., Matemba, L.E., Muller, S.K., Malakalinga, J.J. and Kazwala, R.R. (2015) Epidemiology of Brucella Infection in the Human, Livestock and Wildlife Interface in the Katavi-Rukwa Ecosystem, Tanzania. *BMC Veterinary Research*, **11**, 189. https://doi.org/10.1186/s12917-015-0504-8

[12] Mazeri, S., Scolamacchia, F., Handel, I.G., Morgan, K.L., Tanya, V.N. and Bronsvoort, B.M.D.C. (2013) Risk Factor Analysis for Antibodies to Brucella, Leptospira and *C. burnetii* among Cattle in the Adamawa Region of Cameroon: A Cross-Sectional Study. *Tropical Animal Health Production*, **45**, 617-623. https://doi.org/10.1007/s11250-012-0268-0

[13] Nakoune, E., Debaere, O., Koumanda-Kotogne, F., Selekon, B., Samory, F. and Talarmin, A. (2004) Serological Surveillance of Brucellosis and Q Fever in Cattle in the Central African Republic. *Acta Tropical*, **92**, 147-151. https://doi.org/10.1016/j.actatropica.2004.06.007

[14] Kamga, R., Silatsa, B.A., Farikou, O., Kuiate, J.R. and Simo, G. (2020) Detection of *Brucella* Antibodies in Domestic Animals of Southern Cameroon: Implications for the Control of Brucellosis. *Veterinary Medicine and Science*, **6**, 410-420. https://doi.org/10.1002/vms3.264

[15] Mantur, B.G., Amarnath, S.K. and Shinde, R.S. (2007) Review of Clinical and Laboratory Features of Human Brucellosis. *Indian Journal of Medical Microbiology*, **25**, 188-202. https://doi.org/10.4103/0255-0857.34758

[16] Maichomo, M.W., McDermott, J.J., Arimi, S.M. and Gathura, P.B. (1998) Assessment of the Rose-Bengal Plate Test for the Diagnosis of Human Brucellosis in Health Facilities in Narok District, Kenya. *East African Medical Journal*, **75**, 219-222.

[17] Madut, N.A., Ocan, M., Muwonge, A., Muma, J.B., Nasinyama, G.W., Godfroid, J., Jubara, A.S. and Kankya, C. (2019) Sero-Prevalence of Brucellosis among Slaughterhouse Workers in Bahr el Ghazal Region, South Sudan. *BMC Infectious Diseases*, **19**, 450. https://doi.org/10.1186/s12879-019-4066-4

[18] Pappas, G., Papadimitriou, P., Akritidis, N., Christou, L. and Tsianos, E.V. (2006) The New Global Map of Human Brucellosis. *The Lancet Infectious Diseases*, **6**, 91-99. https://doi.org/10.1016/S1473-3099(06)70382-6

[19] Bayemi, P.H., Webb, E.C., Nsongka, M.V., Unger, H. and Njakoi, H. (2009) Prevalence of *Brucella abortus* Antibodies in Serum of Holstein Cattle in Cameroon.

Tropical Animal Health and Production, **41**, 141-144.
https://doi.org/10.1007/s11250-008-9184-8

[20] Bayemi, P.H., Mah, G.D., Ndamukong, K., Nsongka, V.M., Leinyuy, I., Unger, H. and Luogbou, N.D. (2015) Bovine Brucellosis in Cattle Production Systems in the Western Highlands of Cameroon. *International Journal of Animal Biology*, **1**, 38-44.

[21] Scolamacchia, F., Handel, I.G., Fèvre, E.M., Morgan, K.L., Tanya, V.N., de, C. and Bronsvoort, B.M. (2010) Serological Patterns of Brucellosis, Leptospirosis and Q Fever in *Bos indicus* Cattle in Cameroon. *PLoS ONE*, **5**, e8623.
https://doi.org/10.1371/journal.pone.0008623

[22] Ladbury, G., Allan, K.J., Cleaveland, S., Davis, A., De Glanville, W.A., Forde, T.L. and Zadoks, R.N. (2017) One Health Research in Northern Tanzania—Challenges and Progress. *East African Health Research Journal*, **1**, 8-18.
https://doi.org/10.24248/EAHRJ-D-16-00379

[23] Laine, C.G., Wade, A., Scott, H.M., Krecek, R.C. and Arenas-Gamboa, A.M. (2020) Scoping Review of Brucellosis in Cameroon: Where Do We Stand, and Where Are We Going? *PLoS ONE*, **15**, e0239854. https://doi.org/10.1371/journal.pone.0239854

[24] Awah-Ndukum, J., Mouiche, M., Kouonmo-Ngnoyum, L., Bayang, H.N., Manchang, T.K., Poueme, R., Kouamo, J., Ngu-Ngwa, V., Assana, E., Feussom, K. and Zoli, A.P. (2018) Seroprevalence and Risk Factors of Brucellosis among Slaughtered Indigenous Cattle, Abattoir Personnel and Pregnant Women in Ngaoundéré, Cameroon. *BMC Infectious Diseases*, **18**, 611.
https://doi.org/10.1186/s12879-018-3522-x

[25] Ndoe, M.G., Damdam, F.B.H., Marat, M.K.I., Djoulde, I., Octavie, F.M.D. and Ntungwen, C.F. (2016) Assessment of Safety and Hygiene Measures about the Slaughtering System of Slaughterhouses with Regard to the Human Brucellosis. *Journal of Analytical & Pharmaceutical Research*, **4**, 437-444.

[26] Baba, M.M., Sarkindared, S.E. and Brisibe, F. (2001) Serological Evidence of Brucellosis among Predisposed Patients with Pyrexia of Unknown Origin in the North Eastern Nigeria. *Central European Journal of Public Health*, **9**, 158-161.

[27] Kubuafor, D.K., Awumbila, B. and Akanmori, B.D. (2000) Seroprevalence of Brucellosis in Cattle and Humans in the Akwapim-South District of Ghana: Public Health Implications. *Acta Tropica*, **76**, 45-48.
https://doi.org/10.1016/S0001-706X(00)00088-7

[28] Silatsa, B.A., Kuiate, J.R., Njiokou, F., Simo, G., Feussom, J.K., Tunrayo, A., Amzati, G.S., Bett, B., Bishop, R., Githaka, N., Opiyo, S.O., Djikeng, A. and Pelle, R. (2019) A Countrywide Molecular Survey Leads to a Seminal Identification of the Invasive Cattle Tick *Rhipicephalus* (Boophilus) *microplus* in Cameroon, a Decade after It Was Reported in Cote d'Ivoire. *Ticks and Tick-Borne Diseases*, **10**, 585-593.
https://doi.org/10.1016/j.ttbdis.2019.02.002

[29] Lebel Anne (2011) (et Emmanuelle Pontié), Foumban. In: *Le Cameroun aujourd'hui*, Éditions du Jaguar, Paris, 140-151.

[30] Thursfield, M. (1995) Diagnostic Testing. Veterinary Epidemiology. 2nd Edition, Blackwell Science Ltd., Cambridge, 483.

[31] Mukhtar, F. and Kokab, F. (2008) Brucella Serology in Abattoir Workers. *Journal of Ayub Medical College*, **20**, 57-61.

[32] Aworh, M.K., Okolocha, E., Kwaga, J., Fasina, F., Lazarus, D., Suleman, I., Poggensee, G., Nguku, P. and Nsubuga, P. (2013) Human Brucellosis: Seroprevalence and Associated Exposure Factors among Abattoir Workers in Abuja, Nigeria. *The Pan*

African Medical Journal, **16**, 103.
https://doi.org/10.11604/pamj.2013.16.103.2143

[33] Tsegay, A., Tuli, G., Kassa, T. and Kebede, N. (2017) Seroprevalence and Risk Factors of Brucellosis in Abattoir Workers at Debre Zeit and Modjo Export Abattoir, Central Ethiopia. *BMC Infectious Diseases*, **17**, 101.
https://doi.org/10.1186/s12879-017-2208-0

[34] Mirambo, M.M., Mgode, G.F., Malima, Z.O., John, M., Mngumi, E.B., Mhamphi, G.G. and Mshana, S.E. (2018) Seropositivity of *Brucella* spp. and Leptospira spp. Antibodies among Abattoir Workers and Meat Vendors in the City of Mwanza, Tanzania: A Call for One Health Approach Control Strategies. *PLoS Neglected Tropical Diseases*, **12**, e0006600. https://doi.org/10.1371/journal.pntd.0006600

[35] Alton, G.G., Jones, L.M., Angus, R. and Verger, J. (1988) Techniques for the Brucellosis Laboratory. Institut National de la recherche Agronomique (INRA), 2738000428.

[36] Nielsen, K. and Yu, W.L. (2010) Serological Diagnosis of Brucellosis. *Prilozi*, **31**, 65-89.

[37] ID-VET (2008) ID Screen Brucellosis Serum Indirect Multi-Species Fact Sheet. France: ID-VET, 1-6.

[38] Nabukenya, I., Kaddu-Mulindwa, D. and Nasinyama, G.W. (2013) Survey of *Brucella* Infection and Malaria among Abattoir Workers in Kampala and Mbarara Districts, Uganda. *BMC Public Health*, **13**, 901.
https://doi.org/10.1186/1471-2458-13-901

[39] Madut, N.A., Nasinyama, G.W., Muma, J.B., Sube, K.L.L., Ocan, M., Muwonge, A., *et al*. (2018) Prevalence of Brucellosis among Patients Attending Wau Hospital, South Sudan. *PLoS ONE*, **13**, e0199315.
https://doi.org/10.1371/journal.pone.0199315

[40] Díaz Aparicio, E. (2013) Epidemiology of Brucellosis in Domestic Animals Caused by *Brucella melitensis*, *Brucella suis* and *Brucella abortus*. *Revue Scientifique et Technique* (*International Office of Epizootics*), **32**, 43-60.
https://doi.org/10.20506/rst.32.1.2187

[41] Karimi, A., Alborzi, A., Rasooli, M., Kadivar, M.R. and Nateghian, A.R. (2003) Prevalence of Antibody to *Brucella* Species in Butchers, Slaughterers and Others. *Eastern Mediterranean Health Journal La revue de sante de la Mediterranee orientale*, **9**, 178-184.

[42] Desalew, M., Lawrence, Y., Hiwot, T., Teshome, G., Asfawesen, G., Abraham, A., Theresia Abdoel, H. and Smits, H.L. (2009) Human Brucellosis in Traditional Communities in Ethiopia. *International Journal of Tropical Medicine*, **4**, 59-64.

[43] Rubach, M.P., Halliday, J.E., Cleaveland, S. and Crump, J.A. (2013) Brucellosis in Low-Income and Middle-Income Countries. *Current Opinion in Infectious Diseases*, **26**, 404-412. https://doi.org/10.1097/QCO.0b013e3283638104

[44] Corbel, M.J. (1997) Brucellosis: An Overview. *Emerging Infectious Diseases*, **3**, 213-221. https://doi.org/10.3201/eid0302.970219

[45] Radostits, O.M., Gay, C.C., Blood, D.C. and Hinchcliff, K.W. (2007) Disease Caused by *Brucella* spp. A Textbook of the Disease of Cattle, Sheep, Pigs, Goats and Horses. 10th Edition, ELBS Bailliere Tindall, London, 870-871.

[46] Doganay, M. and Aygen, B. (2003) Human Brucellosis: An Overview. *International Journal Infectious Diseases*, **7**, 173-182.
https://doi.org/10.1016/S1201-9712(03)90049-X

[47] Migisha, R., Dan Nyehangane, Boum, Y., Page, A.L., Zúñiga-Ripa, A., Conde-Álvarez, R., Bagenda, F. and Bonnet, M. (2018) Prevalence and Risk Factors of Brucellosis among Febrile Patients Attending a Community Hospital in South Western Uganda. *Scientific Reports*, **8**, Article No. 15465. https://doi.org/10.1038/s41598-018-33915-9

[48] Franc, K.A., Krecek, R.C. and Häsler, B.N. (2018) Brucellosis Remains a Neglected Disease in the Developing World: A Call for Interdisciplinary Action. *BMC Public Health*, **18**, 125. https://doi.org/10.1186/s12889-017-5016-y

[49] John, K., Fitzpatrick, J., French, N., Kazwala, R., Kambarage, D., Mfinanga, G.S., MacMillan, A. and Cleaveland, S. (2010) Quantifying Risk Factors for Human Brucellosis in Rural Northern Tanzania. *PLoS ONE*, **5**, e9968. https://doi.org/10.1371/journal.pone.0009968

[50] Corbel, M.J. (2006) Brucellosis in Humans and Animals. World Health Organization, Geneva.

[51] Swai, E.S. and Schoonman, L. (2009) Human Brucellosis: Seroprevalence and Risk Factors Related to High Risk Occupational Groups in Tanga Municipality, Tanzania. *Zoonoses and Public Health*, **56**, 183-187. https://doi.org/10.1111/j.1863-2378.2008.01175.x

[52] Sofian, M., Aghakhani, A., Velayati, A.A., Banifazl, M., Eslamifar, A. and Ramezani, A. (2008) Risk Factors for Human Brucellosis in Iran: A Case-Control Study. *International Journal of Infectious Diseases*, **12**, 157-161. https://doi.org/10.1016/j.ijid.2007.04.019

[53] McDermott, J., Grace, D. and Zinsstag, J. (2013) Economics of Brucellosis Impact and Control in Low-Income Countries. *Revue scientifique et technique* (*International Office of Epizootics*), **32**, 249-261. https://doi.org/10.20506/rst.32.1.2197

[54] Briones, G., Iñón de Iannino, N., Roset, M., Vigliocco, A., Paulo, P.S. and Ugalde, R.A. (2001) *Brucella abortus* Cyclic Beta-1,2-glucan Mutants Have Reduced Virulence in Mice and Are Defective in Intracellular Replication in HeLa Cells. *Infection and Immunity*, **69**, 4528-4535. https://doi.org/10.1128/IAI.69.7.4528-4535.2001