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Abstract

Background: Q fever is a zoonotic infectious disease caused by a bacterium called *Coxiella burnetii*. This disease often affects small ruminant farms, leading to reproductive disorders and serious economic losses. It is also a serious public health problem. However, no study has been conducted on Q fever in small ruminants in Burkina Faso. The general objective of this study was to estimate the seroprevalence of Q fever in small ruminants in the rural commune of Bama in Burkina Faso, as well as the knowledge about the skills, practices and behaviours that pose a risk of zoonotic transmission of this disease among livestock farmers in the region.

Methods: To do this, blood was collected from 344 small ruminants including 199 sheep and 145 goats. The indirect enzymelinked immunosorbent assay was used. An epidemiological questionnaire was recorded with 43 farmers to better understand the knowledge, skills, practices and frequency of risk behaviors for zoonotic transmission of the disease.

Results: Apparent seroprevalence "animal" and prevalence "herd" were respectively 34.9% and 81.4%. Sheep were more affected with 38.2% (76/199) compared to 30.3% (44/145) in goats. Females had a higher seroprevalence rate of 36.3% compared to 27.3% for males. Only 4.7% of farmers know about Q fever and that it can be transmitted to humans and other animal species, 100% attend births and abortions and consume raw milk.

Conclusion: As Q fever is a major zoonotic disease, human research and an effective control strategy are needed to improve public health. It is a good idea to conduct an awareness campaign among livestock producers on risk behaviours for zoonotic transmission of Q fever and to extend this study to other regions of the country.

Keywords: Q Fever; Small Ruminants; Zoonotic Transmission; Public Health; Rural Commune of Bama; Burkina Faso

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Abbreviations

iELISA: Indirect Enzyme-Linked Immunosorbent Assay; CI: Confidence Interval; OR: Odds Ratio

Introduction

Livestock is widely practised worldwide and is very important in the economies of many countries, making it one of the main economic activities on which the poorest people depend as a source of food and monetary income [1-3]. However, pathogenic germs such as *Coxiella burnetii* can have a negative impact on its development [2,3]. *Coxiella burnetii* is a Gram-negative intracellular bacterium responsible for Q fever [4-6]. This highly infectious bacterium, which causes Q fever, can be excreted in ruminant milk [7-9]. It can also be excreted in urine, feces and especially in large quantities in the placental membranes of animals infected at term or aborted [10]. Q fever affects a large number of animal species including humans, is a widely ubiquitous disease and its incidence is poorly known and probably underestimated [3]. In general, wild and domestic ticks and ruminants such as cattle, sheep and goats are the main sources of disease in humans [11-14]. Ticks are even considered the main vector in the transmission of *Coxiella burnetii* to animals and humans [15]. People are most likely to become infected by inhaling aerosols or consuming unpasteurized raw milk from infected animals [6]. Q fever is a major zoonosis with global distribution.

In Africa, this zoonosis is largely neglected [16]. It can have a major impact on animal health and public health [10]. In animals, it is generally asymptomatic but in pregnant females it manifests itself in infertility, abortions and stillbirths [17,18]. In humans, it is manifested by headaches, prolonged fever, intense headache, myalgia, arthralgia, vertebral osteomyelitis, cough, endocarditis and death [19-21]. Osteo-articular infections have been observed in children [22]. Overall mortality of 38% was observed in patients with proven chronic Q fever [22]. It is considered an occupational hazard [19-21]. The most exposed persons are slaughterhouse workers, farmers, ranchers, farmers, research laboratory personnel, midwives and veterinarians [19,20,23]. The populations at risk are mainly pregnant women, immunocompromised people, people with vascular abnormalities [19,20,23]. In pregnant women, it causes spontaneous abortions in 100% of cases during the first trimester, intrauterine growth retardation, intrauterine fetal death at the end of pregnancy, premature delivery, miscarriage and neonatal hypotrophy [23,24]. The disease has been found everywhere it was researched except in New Zealand [19]. Despite various control measures in many countries, human and animal Q fever is still a problem.

In Burkina Faso, the disease is unknown to the general public and little research has been conducted. In recent years, we have seen an increase in the number of ruminant farms in the intra-urban and peri-urban areas of the country's major cities; this could introduce a new dimension to the epidemiology of fever Q. To our knowledge, so far, only four studies have been conducted on Q fever in this country. The first, by Gidel., *et al.* [25], reported a seroprevalence of 4% in humans and 0% in cattle. The second, carried out by Gidel and Athawet [26], reported a seroprevalence of 0.5% in a nomadic population group from the Sahelian regions of Upper Volta, now Burkina Faso. The third was conducted at Bobo-Dioulasso hospital by Ki-Zerbo., *et al.* [27] who achieved a seroprevalence of 13.1%. The fourth was performed on dairy cows in the peri-urban area of Ouagadougou by Tialla., *et al.* [3] who obtained a seroprevalence of 26.2%. No studies have been conducted in sheep and goats. Thus, there is no data on Q fever in small ruminants in Burkina Faso. The general objective of this study was to estimate the seroprevalence of Q fever in small ruminants in the rural commune of Bama in Burkina Faso, as well as the knowledge about the skills, practices and behaviours that pose a risk of zoonotic transmission of this disease among livestock farmers in the region.

Materials and Methods

Study period and area

The study was conducted between 10 July and 15 December 2021, in the rural municipality of Bama located in the Houet province. This province of the Haut-Bassins region of Burkina Faso is located 25 km from the city of Bobo-Dioulasso with coordinates 11°22 08 North and 4°22 28 West. This municipality has an annual rainfall ranging between 750 mm and 1200 mm, with a Sudano-Guinean climate

that oscillates between a wet season and a dry season with uneven distribution throughout the year. The choice of this area is motivated by the good numerical representation of the small ruminant herd, it is an agropastoral zone. Well before the selection of the study site, contact and exchange with the agropasters' communities based on available information about the size of small ruminant populations, the presence of transhumant breeders, the incidence of reproductive disorders and the accessibility of the site at all seasons.

Population studied, sampling method and data collation

The choice was made on breeders because they are in direct and close contact with small ruminants, and adopt behaviors such as assisting with births and unprotected abortions, handling of the unborn and the placenta without a glove, cleaning of unmasked litter and consumption of unpasteurized raw milk, behaviors recognized by some authors as risk factors for zoonotic transmission of Q fever [6,8,10]. The animal population studied was made up of small ruminant herds in the rural commune of Bama in Burkina Faso. The twodegree random sampling method was used as described above by Toma., et al. [28]. We did not have an exhaustive list of small ruminant farms in our study area, so we conducted a preliminary survey. This survey identified 152 small ruminant farms. Only 53 of them met the inclusion criteria, which was to have a small ruminant farm with at least 25 sheep and 25 goats and to agree to participate in this study. The number of herds and animals required per herd was determined using the spreadsheet in Appendix 09 to the book "Epidemiology for collective control of major communicable animal diseases" [28], with an expected proportion of 30%, relative accuracy of 20% and a class-in coefficient of 0.1. Based on available resources, we selected 8 small ruminants per herd, which corresponded to 344 small ruminants and 43 herds. A unique identification number, from 1 to 43, was assigned to each herd and a draw was made. The first stage involved drawing 43 small ruminant farms out of 53. In each farm, a random draw of 8 small ruminants was made. Each farm was visited twice, first for awareness and written consent of each breeder and second for blood samples from the animals. For each sample, information on the species, race, sex, age, litter rank and number of abortions for females and locality were noted. There was no refusal to participate in this study. To better understand the knowledge, skills, practices and frequency of risk factors for zoonotic transmission of Q fever in farmers in our study area, an epidemiological questionnaire, with 20 closed questions, designed with "KoboCollect" was registered with the breeder of each of the 43 selected farms. The interviews lasted on average 15 minutes per person surveyed and took place in "Dioula", "Mooré" or, in some cases, French.

Diagnostic methods

In animals, blood samples were taken from the jugular vein on a dry tube identified by the breeding code and the animal number. After centrifugation, the sera were collected and placed in micro-tubes for freezing using sterile disposable pipettes. For the serological diagnosis of Q fever, the Indirect Enzyme-Linked Immunosorbent Assay (iELISA) serological test was used. The iELISA test is considered to be very sensitive (\geq 95%) and very specific (\geq 95%) [29]. The Indirect ELISA Kit (ID.vet Innovative Diagnostics) was used to test our serums for anti-Coxiella burnetii antibodies by using a micro-plate method, in accordance with the recommendations of the World Organisation for Animal Health. Plate reading was performed at 450 nm using a plate reader (Thermo SCIENTIFIC Multiskan GO Version 1.00.38). This allowed the detection of recent and old infections by the detection of IgM, IgG and IgA. The results of the analyses were interpreted in accordance with the manufacturer's recommendations.

Statistical processing

The data collected was entered on the software Epidata version 3.1, before being imported into the R software in its version 4.1.2. The statistical analysis was performed to simultaneously consider various factors that could influence the prevalence of Q fever. The variables of interest, coded in presence/absence, were the positivity on the iELISA test. The explanatory variables were individual and collective characteristics. Risk factors and behaviours in humans were identified using a multivariate model. A logistic regression model (proc logistic, SAS 9.3) was used to analyze the positivity of the iELISA test according to the explanatory variables considered as risk factors or risk behaviors. Comparisons of proportions were made using the Pearson χ^2 test. The 95% confidence intervals (Cl_{asse}) were

calculated using the spreadsheet in Appendix 09 of the book "Epidemiology applied to the collective control of major communicable animal diseases" [28]. A logistic regression has allowed to calculate the odd ratio of the different risk factors observed. The significance threshold was set at 5%.

Ethical considerations

The research protocol of this study received a favorable opinion from the Institutional Ethics Committee of the Centre Muraz (CEICM) under number 2016-15/MS/SG/CM/CEI before its implementation. It was stressed in this protocol that the samples collected will not be used for criminal or commercial purposes and that the identity of participants will be kept confidential. Therefore, all participants were included in the study only when informed consent was obtained and test results were provided to the participants.

Results

Individual and collective characteristics of the surveyed breeders

This study involved 43 breeders. Only 27.9% of the breeders surveyed were literate. The majority of breeders surveyed were between 40 and 60 years old, or 67.5% of the respondents. The series included 37 men (86%) against 6 women (14%) with a sex ratio of 6.2. Of the 43 breeders surveyed, only two (4.7%) were aware of Q fever and knew that it could be transmitted to humans and other animal species. The individual and collective characteristics of the livestock breeders surveyed during the period from 10 July to 15 December 2021 in the rural commune of Bama in Burkina Faso are recorded in table 1.

Variables		Breeders surveyed (n = 43)		
Age Class (years)	[20-40]	20.9% (9/43)		
	[40-60]	67.5% (29/43)		
	[60-80]	11.6% (5/43)		
Sex	Man	86.0% (37/43)		
	Woman	14,0% (6/43)		
Level of education	Literate	27.9% (12/43)		
	Illiterate	72.1% (31/43)		
Knowledge of Q fever	Yes	4.7% (2/43)		
	Not	95.3% (41/43)		
Status of the breeder	Sedentary	67.4% (29/43)		
	Transhumant	32.6% (14/43)		
Farming method	Extensive	95.3% (41/43)		
	Semi-intensive	4.7% (2/43)		
Manure management	Fertilizer	83.7% (36/43)		
	Sale	16.3% (7/43)		
Quarantine of animals	Yes	4.7% (2/43)		
	Not	95.3% (41/43)		
Management of abortions and	Throw into the wild	90.7% (39/43)		
placentas	Bury	9.3% (4/43)		

 Table 1: Individual and collective characteristics of the breeders surveyed during the period from 10 July to 15 December 2021 in the

 rural commune of Bama in Burkina Faso.

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Individual and collective characteristics of small ruminants surveyed

This study involved 344 small ruminants including 199 sheep and 145 goats. Only 16% of small ruminants surveyed were males with a sex ratio of 0.2. The majority of small ruminants surveyed were between 4 months and 24 months old, or 61% of the animals surveyed. For females, 88.2% had a history of reproductive disorders and only 38.4% are multiparous. The individual and collective characteristics of small ruminants surveyed during the period from 10 July to 15 December 2021 in the rural commune of Bama in Burkina Faso are reported in table 2.

Variables		Small ruminants surveyed (n = 344)		
Age Class (months)	[4-12]	61.0% (210/344)		
	> 12	39.0% (134/344)		
Sex	Male	16.0% (55/344)		
	Female	84.0% (289/344)		
Species	Sheep	57.8% (199/344)		
	Goats	42.2% (145/344)		
Race	Local	32.6% (112/344)		
	Exotic	67.4% (232/344)		
Rank of carrying	Nulliparous	8.7% (25/289)		
	Primiparous	52.9% (153/289)		
	Multiparous	38.4% (111/289)		
History of reproductive	Abortion	9.7% (28/289)		
disorders	Placental retention	22.5% (65/289)		
	Metritis	41.5% (120/289)		
	Stillbirth	14.5% (42/289)		
	Without any history	11.8% (34/289)		

Table 2: Individual and collective characteristics of small ruminants surveyed during the period from 10 July to 15 December 2021 in the

 rural commune of Bama in Burkina Faso.

Apparent "animal" seroprevalence of Q fever according to species, age, race, sex and reproductive disorders in small ruminants collected in the rural commune of Bama in Burkina Faso in 2021

The apparent "animal" seroprevalence of Q fever in small ruminants in the 43 farms surveyed was estimated at 34.9% (120/344) 95%CI [34.4 - 35.4] with an estimated "herd" prevalence of 81.4% (35/43) 95%CI [80.9 - 81.9]. Table 3 shows the distribution of Q fever seroprevalence by species, age, race, sex and reproductive disorders in small ruminants collected from the rural commune of Bama in Burkina Faso in 2021. Positive iELISA serological test was associated with species (38.2% [37.4 - 39] for sheep, versus 30.3% [29.9 - 30.7] for goats), age (65.7% [64.9 - 66.5] for small ruminants over 12 months of age, versus 15.2% [14.7 - 15.7] for 4 - 12 months of age), breed (44.8% [44.2 - 45.4] for exotic breeds, versus 14.3% [13.9 - 14.7] for local breeds), to sex (36.3% [35.5 - 37.1] for females, versus 27.3% [26.8 - 27.8]) and reproductive disorders (46.3% [45.9 - 46.7] versus 5.9% [5.7 - 6.1]) for females without reproductive disorders) in small ruminants.

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Variables	Small ruminants tested	Positives	Positives Prevalence (%) and 95% Cl	
Species				0.04
Sheep	199	76	38.2 ± 0.8	
Goats	145	44	30.3 ± 0.4	
Total	344	120	34.9 ± 0.5	
Age Class (months)				0.01
[4-12]	210	32	15.2 ± 0.5	
>12	134	88	65.7 ± 0.8	
Total	344	120	34.9 ± 0.5	
Race				0.02
Local	112	16	14.3 ± 0.4	
Exotic	232	104	44.8 ± 0.6	
Total	344	120	34.9 ± 0.5	
Sex				0.04
Male	55	15	27.3 ± 0.5	
Female	289	105	36.3 ± 0.8	
Total	344	120	$34,9 \pm 0,5$	
Reproductive disorders				0.01
Yes	255	118	46.3 ± 0.4	
Not	34	2	5.9 ± 0.2	
Total	289	120	34.9 ± 0.5	

 Table 3: Q fever seroprevalence by species, age, race, sex and reproductive disorders in small ruminants collected from the rural commune

 of Bama in Burkina Faso in 2021.

CI: Confidence Interval.

Behaviours identified as being at risk of zoonotic transmission in livestock producers

The most frequently observed risk behaviours for zoonotic transmission among small ruminant farmers in the rural commune of Bama in Burkina Faso in 2021 were assisted childbirth and abortions, manipulation of the abortion and placenta without a glove, Cleaning of sheds without masks or glasses, permanent and very close contact with animals and consumption of unpasteurized raw milk from infected animals. The results are reported in table 4.

Variables	OR	95% Cl	P-value
Birthing assistance		3.5-4.1	0.02
Abortion assistance		3.3-3.7	0.02
Handling the untreated proton without a glove		3.8-4.6	0.01
Handling the placenta without a glove		4.2-4.8	0.01
Cleaning sheepfolds without mask, or glasses		2.2-2.6	0.04
Close and constant contact with animals		2.4-2.8	0.03
Consumption of unpasteurized raw milk		4.3-5.1	0.01

Table 4: Most frequently observed risk behaviours for zoonotic transmission among small ruminant farmers in the rural commune of Bama

 in Burkina Faso in 2021.

OR: Odd Ratio; CI: Confidence Interval.

Discussion

Apparent "animal" seroprevalence of Q fever according to species, age, race, sex and reproductive disorders in small ruminants collected in the rural commune of Bama in Burkina Faso in 2021

As vaccination against Q fever is not practised in Burkina Faso, the antibodies detected are those from natural contact between the small ruminant and the pathogen. The mean apparent "animal" seroprevalence of 34.9% in the studied farms is significantly higher than that obtained by Gidel., *et al.* [25] who found no anti-antibody-*Coxiella burnetii* in serums collected from blood samples of cattle in Burkina Faso. The Q fever was not reported at population level in Burkina Faso; while our study shows that about 81.4% of the herds in the rural commune of Bama in Burkina Faso are infected, this shows a high exposure of the herds to the disease; Why this issue should be addressed in terms of animal and public health.

Species, age, race, sex and reproductive disorders could certainly influence the seroprevalence of Q-fever in small ruminants, since iELISA positive testing was associated with the species, age, race, sex and reproductive disorders. We found a higher seroprevalence in sheep (38.2%) compared to goats (30.3%). This observation is consistent with that observed by Turcotte [30] in sheep (70.8%) compared to goats (66.7%). Khalili and Sakhaee [31] also made the same remark with 12% in sheep versus 9% in goats. Ruiz-Fons., et al. [32] also found that sheep were the most infected with 74% in sheep versus 45% in goats. Psaroulaki., et al. [33] found that the most affected goats were 48.2% compared to 18.9% for sheep. The same is true for Sidibé., et al. [34] who observed 24.9% in goats versus 16.9% in sheep. This may explain the size of the non-equivalent sample. These differences could be justified by the presence of grassland that favours a gathering of these two species thus sharing the same grazing areas and watering points. But we still need to do more research on these two species in order to detect which is the most sensitive to Coxiella burnetii. The association with age seems logical because the older the animal, the more likely it is to have been infected, remain infected and be dangerous for other animals [35]. Small ruminants of exotic breeds were more infected than local ones. This may be explained by their low resistance to harsh weather conditions in our study area. Extrinsic factors, such as climate and environment, can also have an impact on disease transmission. The high prevalence of the disease in females who have had at least one abortion is most likely related to Q fever. However, the fact that some females who have never aborted have tested positive could be either a result of an infection which did not cause an abortion or a bias in information, as breeders do not keep track of the history of these females' abortions. Females were more infected than males. This is likely due to the low impact of males in the epidemiology of this disease. The circulation of Coxiella burnetii in small ruminant farms in the municipality of Bama caused reproductive disorders and a statistically significant association was established between seroprevalence and previous reproductive disorders. This situation is similar to that described by Agag., et al. [36] but different from that observed by Yahyaoui., et al. [37]. It should be noted that the role of Coxiella burnetii in the development of reproductive disorders is not entirely clear. In addition to Q fever, other abortifacient diseases such as brucellosis, toxoplasmosis and Rift Valley fever, to name a few, would likely remain in the same study area. Agerholm [38] reported that there is no solid evidence to support the hypothesis that *Coxiella burnetii* is responsible for all cases of metritis, abortions or placental retention, nor to confirm or deny the direct involvement of Coxiella burnetii. Thus, the reasons for an abortion, metritis or cases of placental retention in a breeding are diverse. The attribution of one of these cases to Coxiella burnetii should be based on the detection of the bacterium or its DNA by direct methods such as PCR.

Knowledge, skills, practices and behaviours that pose a risk of zoonotic transmission of this disease in livestock producers

The survey found that the majority of farmers surveyed (95.3%) have no knowledge of Q fever either by name or symptoms and do not know it can be transmitted to humans and other animal species. Doumalin [39] reports in a survey that 57% of farmers are not aware of the disease, and 56% of veterinary agents. Given the zoonotic nature of Q fever, it would be necessary to develop a synergy between veterinarians and physicians around this zoonosis that could affect the general public. The results also show the presence of migrant transhumants (32.6%) in search of pasture and water for their livestock. Kiema., *et al.* [40] report that the main reasons for transhumance

in Burkina Faso are grazing (51.2%), water for drinking of herds (39%) and salt cures (4.3%). When transhumance is to be cited as an effective means of adaptation to climatic hazards and sustainable management of natural resources. It is a source of conflict between farmer and breeder, and also a source of pathogen spread through the many contacts it imposes between infected and healthy animals from different herds or with other animal species.

Some skills and practices such as throwing abortions and placentas into the wild. The sale of organic manure from infested animals and the introduction of new animals into its holding without quarantine could promote the spread of the disease and maintain the survival of the pathogen. Meadows., *et al.* [41] observed that the most affected sheep herds were those where animals were loaned to other farms before they returned to their home farm. Schimmer, *et al.* [42] argue that especially when it comes to female excreta. Beaudeau., *et al.* [43] report that the placentas and abortions of infected females have a very high bacterial load. The release of these products into the wild would be a potential source of contamination.

The lack of knowledge about the dangerousness and epidemiology of Q fever and the low level of literacy among farmers would encourage these breeders to adopt behaviours that are at risk of zoonotic transmission. The most common risk behaviours observed among farmers were assisted childbirth and abortions, handling of untreated calves without a glove, and consumption of unpasteurized raw milk. These behaviours result in potentially very dangerous contacts with the pathogen [6,8,10], which may be responsible for the transmission of this zoonosis [7-9]. Porten., *et al.* [44] reported that human contamination is mainly caused by inhalation of aerosols or products (wool, manure, spreading) from animal excrements or during assisted delivery (placenta, amniotic fluid).

Conclusion

The general objective of this study was to estimate the seroprevalence of Q fever in small ruminants in the rural commune of Bama in Burkina Faso, as well as the knowledge about the skills, practices and behaviours that pose a risk of zoonotic transmission of this disease among livestock farmers in the region. It is clear that Q fever is present in small ruminant farms in the rural municipality of Bama, with an apparent "herd" prevalence of 81.4% and an average "animal" apparent seroprevalence of 34.9% in the herds studied. The most common risk behaviours for zoonotic transmission among farmers in this area were assisted birthing and abortions, handling of the baby and placenta without gloves, cleaning of sheds without masks, the use of unpasteurised raw milk. *Coxiella burnetii* is highly pathogenic for humans, and therefore adequate measures must be taken to better protect the population against this zoonosis. Farmers and consumers need to be made aware of biosafety, the risks of zoonotic transmission of Q fever and the benefits of pasteurizing raw milk before consumption. The implementation of an integrated approach, which takes into account the complex relationships between humans, animals and the environment within different production systems; and the establishment of a multisectoral framework involving physicians, Veterinarians and all public health stakeholders in the context of a "One Health" approach should be considered.

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Conflict of Interest

No conflict of interests is declared.

Author's Contribution

Dieudonné Tialla, Mama Agnès Tea, Apollinaire Lanfo Tialla, Jean-Baptiste Sebou Dah, Aboubacar Kiendrébéogo, Justin Wendwoumgna Kaboré, Amado Ouédraogo, Michel Kéré, Martin Bienvenu Somda, Salimata Pousga, Valérie Marie Christiane Bougma/Yaméogo have contributed equally to the work of this article.

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