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Prevalence and risk factors associated with Brucellosis A critical literature review

By

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Abstract

Purpose: Brucellosis is an infectious disease primarily of domestic and wild animals caused by bacteria of the genus Brucella. It is transmissible to humans through direct contact with infected animal products. This paper aims to determine the prevalence and the risk factors associated with Brucellosis.

Methodology: The paper used a desk study review methodology where relevant empirical literature was reviewed to identify main themes

Findings: The study concludes that factors such as methods of food preparation and consumption of milk, pasteurization of dairy products, and direct contact with infected animals, lack of awareness and sensitization increase the risk of Brucellosis in communities.

Recommendations: Creating awareness and promoting sensitization programs run in the community is necessary so as to impact knowledge on the methods of transmission, identification of the infection, prevention and types of treatment. This will eventually report decreased cases of Brucellosis in animals and humans.

Keywords: *Brucellosis, risk factors, prevalence*

1.0 INTRODUCTION

1.1 Background of the Study

Brucellosis is an infectious disease primarily of domestic and wild animals caused by bacteria of the genus Brucella. It is transmissible to humans through direct contact with infected animal products. It constitutes a major health problem in many parts of the world. Brucellosis is a multisystem disease with a broad spectrum of manifestation (Jiang, W. et al., 2019). The true incidence of human brucellosis is unknown. Reported incidences in endemic-disease areas vary widely, from 200 per a hundred thousand people. While some areas, such as Peru, Kuwait, and parts of Saudi Arabia, have a high incidence of acute infections, the low incidence reported in other known brucellosis endemic areas may reflect low levels of surveillance and reporting. Other factors such as methods of food preparation, pasteurization of dairy products, and direct contact with infected animals also influence risk to the population.

Brucellosis is prevalent in all major livestock production systems in Sub-Saharan Africa. Yet its presence is often unrecognized through lack of awareness by both the health care staff and veterinarians and absence of accessible laboratory diagnostic facilities (Pathak, A. D et al 2016). As a consequence, brucellosis remains a largely neglected disease with little attention to control

and prevent except in South Africa where a successful control policy of vaccination combined with test and slaughter has been initiated. The incidence of brucellosis is highest in pastoral production systems where large numbers of animals mix. Bovine brucellosis seems to be more common than ovine brucellosis. However, this may be an artifact reflecting the serological testing of livestock species. Much less is known on the prevalence in humans and of the effect on health in this region of the world. Provision of improved diagnostics is essential to enable such investigation to be undertaken.

Human brucellosis is rarely caused by consumption of infected meat but humans may acquire the disease readily by percutaneous infection. Even imperceptible lesions of the skin may serve as portals of entry and there is a marked predisposition to brucellosis among the occupational handlers of animals and meat in areas where the disease is enzootic.

Temperature, humidity and pH of the environment influence the survival of Brucella melitensis as well as Babortus (Ahmed et al, 2016). Bruccellae are sensitive to direct disinfections and pasteurization. In optimal conditions Brucellae survives in tap water, damp aborted fetuses, and uterine exudates and in frozen tissues.

Ideally, effective control of brucellosis should be through a combination of improved diagnosis, vaccination and treatment, together with measures to increase awareness, and improved farm sanitation and food hygiene (Dorneles et al, 2015). Collectively, these will increase the effect of control measures and lessen the burden of disease. An integrated disease education and community participation program may assist the achievement of this goal. Traditional beliefs and habits may interfere with disease prevention and prohibit its acceptance due to lacunae in disease and health knowledge. Awareness of the cause of this disease and knowledge of measures for prevention and resulting benefits of this can be provided through such a program, creating a positive attitude towards disease prevention. A disease education and community participation program will promote involvement, encourage acceptance thereby increasing the efficacy of control measures.

1.2 Statement of the problem

Human brucellosis causes physical and psychological suffering due to infection, hospitalization, the cost of drugs and loss of income due to illness. The country incurs huge 4 costs generated by prophylactic measures taken to control brucellosis and provision of testing and treatment facilities. There is no doubt that outbreak of brucellosis especially Brucella melitensis causes significant loss of income to the farmers. Human brucellosis has been poorly studied in Africa. A prevalence of 3.8% has been reported in nomadic pastoralists from Chad. Slaughter house workers in Djibouti had a prevalence of 6.5% and the high risk groups in Eritrea showed a prevalence of 3.0% to 7.1% while in Eastern Nigeria 5.2% were seropositive.

The consumption of unpasteurized milk by many people in the community is a major risk factor. There is little awareness in this community about this risk factor. The large livestock population is a potential source of infection (Pelini et al, 2019). Many people have large flocks of goats and sheep, which is a potential source of Brucella melitensis, a highly virulent form of Brucella infection. Lack of appropriate medical care makes it difficult for cases to be diagnosed and treated. Health facilities especially those in rural areas need to be equipped for tackling this problem. Poor reporting system on cases of the disease and unavailability of necessary information to the

stakeholders such as the health workers, veterinarians, community's own resource persons and the community indicate that there is a long way to go in curbing this problem.

1.3 Objectives of the Study

The general objective of the study was to determine the prevalence and risk factors associated with Brucellosis.

1.4 Justification and Significance of the Study

This study is designed to determine the prevalence and risk factors associated with Brucellosis. This will in the long run help in controlling and preventing the spread of the diseases ultimately improving the health status of the community. Cases often remain unrecognized because of the inaccurate diagnosis, and are thus treated as other diseases or as "fever of unknown origin". This therefore justifies the need for this study which intends to determine the disease prevalence in the study area.

Although this disease is becoming an increasing community health problem, there is lack of knowledge especially on modes of disease transmission. Therefore, the finding will be useful to many stakeholders, especially, Ministries of Health and Livestock Development in understanding the various socio-medical underlying factors and in doing disease surveillance. These findings will improve the disease surveillance and encourage collaboration and participation of health workers, veterinarians, the local administration and the community in reducing the disease burden.

2.0 LITERATURE REVIEW

2.1 Occurrence and transmission of the disease

The disease "Malta fever" or "undulant fever" was prevalent in the island of Malta and various other places along the Mediterranean coast, but at first with only a local significance. However, as transport and communication became available between different parts of the world, the disease spread from its original home. David Bruce first discovered the causative organism in 1889 from a fatal case of the disease. It was first called Micrococcus melitensis and subsequently named Brucella melitensis after its discoverer. Brucellosis is predominantly an occupational disease of those working with infected animals or their tissues, especially farm workers, veterinarians and abattoir workers; hence it is more frequent among males. Sporadic cases and outbreaks occur among customers of raw milk and milk products especially unpasteurized soft cheese from cows, sheep and goats. Brucellosis imposes a serious public health and socio-economic problem globally.

A study on the prevalence of human brucellosis was conducted in Saudi Arabia where a sample of 4900 subjects was randomly selected from house to house survey. Investigation included interview, clinical examination and blood sampling for antibody titre determination. A total of 4794 completed the study. The result of laboratory test indicated that a significant proportion of the population (19.2%) had serological evidence of exposure to Brucella antigen and 2.3% had active disease. Direct contact with animals and consumption of raw products of animal origin were identified as the main risk factors (Alballa., 1995).

Similarly, a study on knowledge, attitude and practice in Saudi Arabian community towards problem of brucellosis revealed that out of 337 patients examined for knowledge on methods 10

and means of transmission of the disease, 309 (92%) were ignorant while 28 (8%) appeared to have some knowledge about the source, type of animal contact and prevention of illness. None of the 337 subjects was able to link the disease with microbial infections. The common practice associated with the occurrence includes; consumption of raw milk, unpasteurized animal products like butter and cheese, close animal contact and slaughter and disposal of animal wastes (Bilal et al., 1991).

Humans working in the meat industry may contract the disease conjunctively or by nasal mucous membrane infection. Many became infected with brucellosis when handling aborted fetuses or healthy calves born to infected cows, performing gynecological operations. Human beings can be infected with all species of Brucella. When milk is contaminated with Brucella abortus, cases are usually sporadic but when it is infected with Brucella melitensis or Brucella, outbreaks are often of epidemic proportions. When Brucella melitensis occurs, human disease is always recognized and the same applies to Brucella suis.

2.2 Clinical manifestations of Brucellosis in humans

Brucellosis is a complex system of infection that varies considerably and may last three days to six months and occasionally for longer than a year (Hugh, 2000). Patients may present with an acute systemic, febrile insidious chronic infection or a localized inflammatory process. There are no pathognomonic signs of brucellosis and patients present with non-specific signs as fever, malaise, sweats, fatigue, anorexia, muscle or joint aches. Infection may be severe and may be followed by chronic intermittent relapses.

Other forms of presentations of the disease include respiratory system involvement (Georgios et al.,2003), ocular complications (Isaias et al., 2008), epididymoorchitis in males (Amalia, 2001, Stamatiou et al.,2009) and spontaneous abortion, in-utero infection of the fetus, cardiovascular complications, and gastrointestinal system complications have also occur.

2.3 Clinical manifestations of Brucellosis in animals

Brucellosis is a herd disease and abortion is the principle clinical manifestation in an infected herd with abortions occurring at about 5-7 months of pregnancy. Full-term calves may be born but die soon after birth. In fully susceptible herds, abortion rates may vary from 30% to 80% (Corbel, 2006). Retained placenta and secondary metritis are common in infected females and may lead to permanent sterility. Subsequent gestations are normal, after a period of temporary sterility and only 5% of infected females have residual sterility. Most cows will shed the bacteria in the milk and this is the main source of infection to humans. In bulls acute or chronic infections of the reproductive tract; orchitis, epididymitis and seminal vesiculitis may occur and this contributes to continued infection to the females especially if the bulls are used for breeding or producing semen for artificial insemination. Hygromas, particularly of the carpal joints, occur in some animals in chronically affected herds (FAO, 2006).

2.4 Treatment and prevention of Brucellosis

The essential element in the treatment of all forms of human brucellosis is the administration of effective antibiotics for an adequate length of time (Corbel, 2006). Generally, the treatment recommended by the WHO for acute brucellosis in adults is rifampicin 600mg to 900 mg and

doxycycline 200mg daily for a minimum of six weeks. Treatment of uncomplicated brucellosis in persons above eight years of age with intake of 100mg of doxycycline twice a day for six weeks combined with 1gm streptomycin daily for two to three weeks is also adequate. It has also been suggested that the combination of doxycyline and an aminoglycoside in addition of rifampicin may be a better option.

There are no effective human vaccines that exist and experiments have been proven less effective and cause allergies. Prevention of brucellosis in humans still depends on the eradication or control of the disease in animal hosts, the exercise of hygienic precautions to limit exposure to infection through occupational activities, and the effective heating of dairy products and other potentially contaminated foods (Gul et al., 2007).

2.5 Empirical view

Abdalla (2016) conducted a study to determine the prevalence and factors associated with brucellosis among community members in Mandera East Sub-County, Mandera County. The study was descriptive cross sectional study which collected both qualitative and quantitative data from where a sample of 420 respondents was systematically selected from heads of 2,617 households form Mandera East Sub-county. The study instruments included questionnaire, Focus Group Discussion guide and Interview Guide. Blood samples were screened for brucellosis using Rose Bengal Plate Test (RBPT) and the positive sera were subjected through Serum Slow Agglutination Test (SSAT) which acted as a confirmatory test. Data was analyzed using SPSS Version 20 and results of the study presented in frequencies and percentages in Tables and Figures. Rose Bengal Plate Test (RBPT) indicated a prevalence of 24.8% (95% CI: 20.0–29.6) and Serum Slow Agglutination Test (SSAT) indicated that the prevalence was at 14.3% (95% CI: 8.7– 19.9) among the respondents. The study showed that the prevalence was higher among the male respondents (98%; n=103) as detected through RBPT and (98%; n=57) confirmed through SSAT. The results of the study showed that there was significant relationship between the gender and prevalence as tested through RBPT ($P<0.001$) and through SSAT ($P<0.001$). It further showed that the majority of the respondents (69%) was aware of the disease and that 31% (n=130) took fermented milk without boiling, while only a few (6%; n=25) of respondents pasteurized it. There was a significant relationship between the respondents' milk preparation practices before consuming and brucellosis status using RBPT ($\chi2 = 17.115$; $df=4$; $p=0.002$) but not when tests were done through SSAT ($\chi2=8.737$; $df=4$; $p=0.068$).Factors associated with the spread of brucellosis among the community members in Mandera East Sub-county included directly getting into contact with animals such as goats, cows, wild animals dogs, camels, and sheep and taking poorly prepared milk; consuming raw blood from livestock; taking raw or poorly cooked meat and getting involved in various activities touching on livestock.

Mark O (2018) conducted a cross-sectional study to determine the prevalence of Brucellosis and identify herd-level factors associated prevalence in small ruminants in Garissa County of North Eastern Kenya. The study also assessed the pastoralists' knowledge, attitude and practices towards Brucellosis. A total of 2,400 sera from 120 flocks were collected from sheep and goats which were randomly selected using a multi-stage sampling technique and data on potential herd-level factors were collected from the pastoralists' ≥15 years using a pre-tested structured questionnaire. The sera were analyzed using Rose Bengal Plate Test (RBPT) and positive reactors confirmed by

Complement Fixation Test (CFT) using serial interpretation. A sample was considered to be positive when both tests results were positive and a herd was considered positive when a single animal within the herd tested positive on both tests. Multivariable logistic regression was used to investigate for independent factors associated with flock Brucellosis positivity in small ruminants. The overall prevalence of Brucellosis at individual animal-level was 20.0% (95% CI: 18.2% to 22.0%); in goats 24.3% (95% CI: 21.8% to 27.1%) and sheep 12.5% (95% CI: 10.2% to 15.2). Overall true herd-level prevalence was 65.8% (95% CI: 54.3% to 77.2%). Seeking veterinary services [aOR=0.30 (95% CI: 0.12 to 0.76], introduction of new animals into the flock [aOR=8.0 (95% CI: 3.09 to 20.70] and experiencing abortions in the flock [aOR=3.43 (95% CI: 1.33 to 8.88] were independently associated with Brucellosis herd prevalence in small ruminants. A total of 120 pastoralists were interviewed of which 95 (79%) had heard of Brucellosis and 17(18%) mentioned bacteria/germ as cause. Forty-four (46%) would do nothing if they had aborting animal in their herd, 91 (96%) consumed raw milk in the past year and 72 (76%) assisted an animal during parturition process and none used glove. The results of the study highlight that there is a considerable high prevalence of Brucellosis and factors that contributes for its transmission in small ruminants in Garissa County. This poses potential public health threat associated with zoonotic transmission. The study also highlights that though the community has some knowledge on Brucellosis, attitudes and practices are poor.

Mamo (2017) conducted a study to determine the socio-medical factors underlying the disease prevalence among the nomads in Bubisa sub-location of Maikona Division of Marsabit District. Descriptive cross sectional study design was used and 400 respondents were selected following systematic random sampling method. Interview schedule for households, key informants and focus group discussions were used for data collection. Odds ratio and chi-square test was used for testing relationship between variables. A majority of the subjects (75.8%) had no formal education, with the main occupation being livestock rearing (78.2%) and the average household size being 5-6 persons. Statistical test showed that women are more susceptible to infection than men in this community (Cross products odds ratio=1.62, x2=4.02, dal and (p) <0.05). Most informants (73.3%) had knowledge on the prevalence of brucellosis in the area, locally referred to as "dukub annani"(the disease of the milk). A substantial number of households interviewed (31.8%) had experienced at least a case of human brucellosis in the last one year. The results of the study showed that consumption of raw milk without boiling (38.5%) is among the risk factors reported to contribute to disease prevalence in the area. Statistically significant relationship was established between consumption of raw milk and the household prevalence of brucellosis. Similarly, a significant relationship was observed between household prevalence of brucellosis and drinking of animal blood (Cross product odds ratio=1.64 and 4.30,df--1(p)<0.05). Households with large livestock population reported more cases of the disease. Majority of reported cases (62.2%) were from households with livestock population of more than 100 as compared to 19.7% and 17.3% reported by households with a livestock population of 50-100 and 0-49, respectively. A large majority 347 (86.8%) reported that one container is used for milking and milk storage, a statistically significant relationship was found to exist between the household prevalence of brucellosis and milk harvesting. Thorough health education especially on the mode of transmission of brucellosis and the main risk factors such as consumption of raw milk, consumption of animal blood and failure to seek treatment from health institutions needs to be emphasized.

Philemon (2016) conducted a study to determine the prevalence and factors associated with Brucellosis in livestock in Baringo County, Kenya. This cross sectional study was carried out in four sub-counties of Baringo County, in which farms were randomly selected from each sub-county. Blood (10ml) from selected cattle (n= 250), sheep (n= 142) and goats (n= 166) was collected in sterile plain vacutainers. Bulk raw cattle milk (n=83) was also collected. All serum samples were screened for Brucella antibodies using Rose Bengal Plate test (RBPT) and by competitive Enzyme Linked Immunosorbent Assay (cELISA). Brucella antibodies in milk were assayed using Milk Ring Test (MRT). Polymerase chain reaction (PCR) was carried out on blood clots from all RBPT-positive serum samples as well as on blood clots of 7% of the serum samples that turned negative on RBPT to determine presence of brucella antigens in those samples. Twenty three (9.2%) of the 250 cattle serum samples reacted positive to RBPT while 17 (6.8%) reacted positive to cELISA with cumulative reactors of 25 (10%). The 166 caprine serum samples had 17 (10.2%) positive reactors to RBPT and 11 (6.6%) by cELISA. Cumulative caprine reactors were 18 (10.8%). Positive ovine serum samples were 10 (7%) and 7 (4.9%) on RBPT and cELISA respectively, yielding positive cumulative reactors of 11 (7.7%). The sensitivity and specificity of RBPT was 88.6% and 96.4% respectively with a predictive value positive of 62% and predictive value negative of 99%. From the 83 milk samples collected, 9 (10.7%) tested positive to Milk Ring Test. xv Brucella abortus DNA was extracted from 11 of cattle blood clots and from two goat and one sheep blood clots respectively. Brucella melitensis DNA was extracted from one goat blood clot. Mixed farming was reported by 57% of the respondents, communal grazing reported by 32% of the respondents, use of communal watering points reported by 38% of the interviewees and allowing of animals to calve down on pasture reported by 91% of the respondents. The results of the study showed that the introduction of a new animal reported by 42% was found not to be a risk factor associate with Brucellosis. From the serological results, it is evident that brucellosis occurs in livestock in Baringo, predominantly caused by B. abortus. This study also established that there is a huge knowledge gap on its risk factors in the region. It is therefore important to establish an educational campaign in the region on the significance of the disease, and establish possible control measures.

2.6 Research Gaps

Methodological gap is the gap that is presented as a result in limitations in the methods and techniques used in the research (explains the situation as it is, avoids bias, positivism, etc.). This study aimed at determining the prevalence and factors associated with Brucellosis adopted a desktop literature review unlike the previous studies done by Philemon, Mamo and the others that used a descriptive cross- sectional methodology.

Conceptual gap arises because of some difference between the user's mental model of the application and how the application actually works. Mamo (2017) conducted a study to determine the socio-medical factors underlying the disease prevalence among the nomads in Bubisa sub-location of Maikona Division of Marsabit District. Descriptive cross sectional study design was used and 400 respondents were selected following systematic random sampling method. Interview schedule for households, key informants and focus group discussions were used for data collection. Odds ratio and chi-square test was used for testing relationship between variables. A majority of the subjects (75.8%) had no formal education, with the main occupation being livestock rearing

(78.2%) and the average household size being 5-6 persons. Statistical test showed that women are more susceptible to infection than men in this community. The study presented a conceptual gap as it focused on to determine the socio-medical factors underlying the disease prevalence among the nomads in Bubisa sub-location of Maikona Division of Marsabit District while the current study focused on determining the prevalence and risk factors associated with Brucellosis.

3.0 METHODOLOGY

3.1 Introduction

The study adopted a desktop literature review method (desk study). This involved an in-depth review of studies related to the prevalence and risk factors associated with Brucellosis. Three sorting stages were implemented on the subject under study in order to determine the viability of the subject for research. This is the first stage that comprised the initial identification of all articles that were based on the prevalence and risk factors associated with Brucellosis. The search was done generally by searching the articles in the article title, abstract, keywords. A second search involved fully available publications on the subject on the prevalence and risk factors associated with Brucellosis. The third step involved the selection of fully accessible publications. Reduction of the literature to only fully accessible publications yielded specificity and allowed the researcher to focus on the articles that related to prevalence and risk factors associated with Brucellosis which was split into top key words. After an in-depth search into the top key words (risk factors, Brucellosis, prevalence), the researcher arrived at 5 articles that were suitable for analysis.

The four articles were findings by Abdalla (2016) conducted a study to determine the prevalence and factors associated with brucellosis among community members in Mandera East Sub-County, Mandera County. The study was descriptive cross sectional study which collected both qualitative and quantitative data from where a sample of 420 respondents was systematically selected from heads of 2,617 households form Mandera East Sub-county. The study instruments included questionnaire, Focus Group Discussion guide and Interview Guide. Blood samples were screened for brucellosis using Rose Bengal Plate Test (RBPT) and the positive sera were subjected through Serum Slow Agglutination Test (SSAT) which acted as a confirmatory test. The results of the study showed that there was significant relationship between the gender and prevalence as tested through RBPT ($P<0.001$) and through SSAT ($P<0.001$). It further showed that the majority of the respondents (69%) was aware of the disease and that 31% (n=130) took fermented milk without boiling, while only a few (6%; n=25) of respondents pasteurized it. There was a significant relationship between the respondents' milk preparation practices before consuming and brucellosis status using RBPT ($\chi 2 = 17.115$; df=4; p=0.002) but not when tests were done through SSAT ($\chi 2 = 8.737$; df=4; p=0.068).

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transmission in small ruminants in Garissa County. This poses potential public health threat associated with zoonotic transmission. The study also highlights that though the community has some knowledge on Brucellosis, attitudes and practices are poor.

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Philemon (2016) conducted a study to determine the prevalence and factors associated with Brucellosis in livestock in Baringo County, Kenya. This cross sectional study was carried out in four sub-counties of Baringo County, in which farms were randomly selected from each sub-county. The results of the study showed that the introduction of a new animal reported by 42% was found not to be a risk factor associate with Brucellosis. From the serological results, it is evident that brucellosis occurs in livestock in Baringo, predominantly caused by B. abortus. This study also established that there is a huge knowledge gap on its risk factors in the region. It is therefore important to establish an educational campaign in the region on the significance of the disease, and establish possible control measures.

4.0 SUMMARY, CONCLUSIONS AND RECOMMENDATIONS

4.1 Introduction

This section presents results based on the objectives of the study. The results are on the prevalence and risk factors associated with Brucellosis.

4.2 Conclusion

The high prevalence of Brucellosis in communities is associated by the large number of animals these pastoralists and are in constant contact with, that is through livestock herding and birthing and handing aborted materials. The findings of this study are consistent with Racloz et al., (2013) who studied the persistency of brucellosis in pastoral systems. Getting into direct contact with animals was found to be one of the risk factors of Brucellosis transmission. This is consistent with observations made by Jones et al., (2008) that nearly two-thirds of human pathogens are zoonotic and, of greater concern, nearly three-quarters of emerging and re-emerging diseases of human beings are zoonoses. Every case of human brucellosis has an animal origin and endemic nature of the disease in animals poses a continuous risk for human infection. A study by Kenneth et al., (2009) indicated high brucellosis prevalence among the study participants who had handled animals or their products in one way or the other.

Another risk factor associated with Brucellosis found among these pastoralist communities was the consumption of milk that is not properly prepared. They fermented and consumed milk without boiling or pasteurizing it. These results were consistent with previous studies Geoffrey et al., 2002; Kenneth et al., 2009; Mutanda et al., 1998) who noted that unprocessed milk from the market and

consuming it raw were independently associated with brucellosis. Mode of milk preparation before consumption contributed to the prevalence where majority of the infected respondents consumed un-boiled and fermented milk. The prevention of brucellosis infection in humans is a major reason for the advocacy of milk pasteurization worldwide (Staal, 2000).

A great majority of the pastoralist communities are not aware of the risk factors associated with infections and spread of Brucellosis. The level of awareness was found to be significantly associated with the level of education. This was consistent with studies done by Adesiji et al., 2005andMubyaziet al., 2013.

4.3 Recommendations

It is important to scale up the level of awareness of the identification, prevention and treatment of Brucellosis among community members. Communities need to be sensitized on the preventive measures from Brucellosis infections. Most human brucellosis cases have mainly two different origins: food borne (milk and milk products) or occupational (farmer, butcher, veterinarian,). If human cases are predominantly found in certain professional categories, it suggests that sanitary measures related to milk and milk products are well implemented, whereas control should be enhanced in the reservoir animal species. In addition to that, A study is required on the appropriate and sustainable methods of dealing with high prevalence of brucellosis among pastoralists and a need to establish the existence and prevalence of other zoonotic diseases.

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