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Seroprevalance of Brucellosis in Small Ruminants and Associated Risk Factors in Ziway Dugda Woreda, Arsi Zone Southeast Ethiopia

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Research Article

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ABSTRACT

A cross sectional sero-prevalence study was conducted on 384 small ruminants in Ziway Dudga district, Ethiopia to investigate the status of small ruminants' brucellosis and identify potential risk factors. Serum samples collected from 202 sheep and 182 goats were analyzed using Rose Bengal Plate Test (RBPT), and those who were RBPT positive was subjected to Complement Fixation Test (CFT). The overall sero-prevalence was 1.56% (6/384). The prevalence of brucellosis among sheep and goats was 4 (1.04%) and 2 (0.52%), respectively. Small ruminants brucellosis in female and male animals was 5 (1.30%) and 1 (0.26%), respectively. Sero-prevalence of brucellosis in adult animals (1.30%) was higher than that of young animals (0.26%). The prevalence with a history of retained fetal membrane (6.7%) was higher than animals with no history of retained fetal membrane (1.6%). The result of study showed that there is no statistical association between species, sex, age and history of retained fetal membrane ($P>0.05$). The prevalence among females with a history of abortion (16.7%) was significantly higher ($P<0.05$) than those without history of abortion (1.2%). Generally, the sero-prevalence of small ruminants brucellosis found in Ziway Dudga area was low and the sero-prevalence was closely associated with history of abortion. Training should be provided for farmers and livestock attendants regarding effective sanitary and hygienic management practices during handling of animals and animal products such as aborted fetal materials and other body secretions.

INTRODUCTION

Ethiopia is believed to have the largest livestock population in Africa. Ethiopia has a total 59.5 million heads of cattle, 30.70 million of heads sheep, 30.20 million heads of goat, 56.53 million of chicken, 2.16 million horse, 8.44 million of donkey, 0.41 million heads of mules and about 1.21 million camels which have a significance contribution to national economy. Small ruminants (sheep and goats) have a unique role in smallholder agriculture as they require small investments; faster growth rates, have shorter production cycles and greater environmental adaptability as compared to large ruminants. They are important protein sources in the diets of the poor and help to provide extra income and support survival for many farmers in the tropics and sub-tropics [1].

Although sheep and goat plays a significant role in national economy of the country to date the benefit obtained from these livestock are hampered by different constrains. Livestock diseases are among the important technical constraints that have hindered the development of the sector by decreasing production and hampering trade in animal and animal products. Among many constraints infectious disease that are caused by bacteria and virus are the most common problem of livestock production [2].

Of many bacterial diseases of animals, brucellosis is a disease that brings reproductive failure to livestock and serious health problems to humans. The disease poses a barrier to trade of animals and an animal product represents a public health hazard and is an impediment to free animal movement. The economic and public health impact of brucellosis remains of particular concern in developing countries. Even though brucellosis has been eradicated from developed countries, it remains endemic in many parts of the world, including Latin America, the Middle East, western Asia, some Mediterranean regions and Africa.

Brucellosis is a disease caused by infection with bacteria of the genus *Brucella* and there is a general host preference within the genus. *Brucella melitensis (B. melitensis)* (biovars 1, 2 or 3) is the main causative agent of caprine and ovine brucellosis. *B.melitensis* and *B. abortus* are responsible for late abortions and birth of weak and sickly new born in cattle, sheep and goats. *B. melitensis* is the main cause of disease in adult male and female sheep and goats. The principal manifestation of brucellosis is reproductive failure, including abortion, birth of unthrifty neonates and infertility in female animals. Orchitis, epididymitis and rarely arthritis are the clinical manifestation of brucellosis male animals. Abortions in the mid third of gestation that are associated with retained placenta and fetal membranes as well as reduced milk production may be indicative [3]. Diagnosis depends on the isolation of *Brucella* from abortion material, udder secretions or from tissues removed at post-mortem. Presumptive diagnosis of *brucella* infection can be made by assessing specific cell-mediated or serological responses to *brucella* antigens.

Various prevalence rates have been reported in Small ruminant brucellosis in Ethiopia so that the disease is endemic to the country. Reported prevalence include, 1.5% in sheep and 1.3% in goats in central highlands of Ethiopia, 4.8% in Afar, 17 (1.99%) and 15 (1.76%) in small ruminants slaughtered at Debre Ziet and Modjo export abattoirs using Rose Bengal plate test and complement fixation test, respectively, 1.37% (4/291) in three selected districts of Jijiga zone, Somali region.

Despite the disease is endemic in the country there is scarcity in research, particularly in Ziway Dugda woreda, thus the objective of this study was to assess sero-prevalance and associated risk factors of brucellosis in small ruminants in Ziway Dugda district.

MATERIALS AND METHODS

Study area

The study was conduct in Ziway Dugda district, Arsi zone, Oromia regional state, South East Ethiopia. Ziway Dugda is one of the worada in Oromia region of Ethiopia, a part of Arsi zone located in the great reft valley. The woreda situated at 222 km from capital city of Addis Ababa and 47km from Arsi Zone Assella town. The Woreda is located between 7°27'00'N-8°00'34''N Latitude and 38°45'00''E-39°03'13''E Longitude in the Great Rift Valley at an altitude range 1500 m-2300 m. With annual rainfall ranges from 200 mm to 1200 mm. Due to its altitudinal location, the climatic conditions of the woreda is dominantly moderately warm which has a temperature ranging from 20°C to 25°C. Ziway dugda is bordered on the south by Munesa, on the West and North by shewa zone on the East by Hetosa and on the Southeast by Tiyo also on its edge lake zuway whose area is woreda share with east shewa zone.

Study design and population

The study design was cross sectional and conducted from December, 2017 to March, 2018 to study the prevalence and associated risk factors of brucellosis in small ruminants. Randomly all animals above six month age was included in the study. The age of animals was determined by dentition as young (<1½ years) and adult (>1½ years).

Sampling and sample size

The sample was collected from animals above six years using method called simple random sampling. The sample size for study animal was calculated. In study area no study was conducted in small ruminant brucellosis thus 50% expected prevalence was considered in sample size determination [4]. 95% confidence interval and 5% desired absolute precision were also considered as other determinants in sample size determination. According to the relevant formula for sample size at 95% confidence interval is:

$$n = \frac{(1.96)^2 p_{exp}(1 - p_{exp})}{d^2}$$

Where: n=required sample size

P_{exp}=expected prevalence

d=desired absolute precision.

Then, substituting 50% of expected prevalence and 5% desired absolute precision about 384 samples was collected.

Sample collection and processing

About 6 ml-7 ml of blood was collected from jugular venipuncture using sterile plain vacutainer tubes (non heparinized vacutainer tube), needle holder and needle. Then collected sample from each animals was labeled by asking the owners of animals with sex, age, history of abortion, Retained Fetal Membrane (RFM) and management to asses associated risk factors for disease occurrence and left for 24 hour at room temperature to allow serum to separate from collected blood and the separated serum was gently decanted it to sterile cryovials [5]. The serum samples were then transported using an ice box to Asella Regional Veterinary Laboratory and later stored at -20°C until testing for *Brucella* antibodies.

Rose Bengal Plate Test (RBPT)

In this study two types of serological tests were employed as a screening and confirmatory test for the detection of *Brucella* antibody. Rose Bengal Plate Test (RBPT) was done at Asella regional veterinary laboratotory in serology

room. This test was used as a screening test for the presence of *Brucella* agglutinins. Rose Bengal stained antigen, negative and positive sera were required in doing RBPT. About 30 µl of the sera samples were dispensed onto the plate and 30 µl of RBPT antigen was dropped alongside the sera. By using the tip of the automatic micropipette tips, the sera were mixed and examined for agglutination. For interpretation of results both positive and negative controls were employed. The test positive sample was sent to National Veterinary Association (NVI), Bishoftu Ethiopia for further confirmatory test [6].

Complement Fixation Test (CFT)

Complement Fixation Test (CFT) was conducted in NVI which was found in Bishoftu, Ethiopia. All sera which were tested positive by the RBPT were further retested using the CFT for confirmation.

Data management and statistical analysis

All raw data generated from this study were coded and recorded in a Microsoft Excel 2010 spread sheet and statistical analysis was conducted using SPSS statistical software version 20.0. Descriptive statistics were used to compute percentages, proportions and frequency distributions of the data. Logistic regression was used to measure the association between prevalence of brucellosis and the different risk factors such as age, species, and sex, history of Retained Fetal Membrane (RFM) and history of abortion. Confidence level was held at 95% and statistical analysis for the difference in prevalence of brucellosis among risk factors were considered significant when $P<0.05$.

RESULTS

Overall results

Of the total 384 animal screened using RBPT about 6 (1.56%) was found positive test serum for Brucellosis and all positive tested sera 6 (1.56%) was also found positive in CFT test, thus the overall seroprevalance of Brucellosis in small ruminants of Ziway dugda woreda was found 6 (1.56%) [7].

Seroprevalance of small ruminants Brucellosis based on species

Of the total 202 (52.6%) sheep and 182 (47.4%) goat, 4 (1.04%) and 2 (0.52%) was sero positive for brucellosis in sheep and goat respectively with no statistical significant ($P>0.05$). The study showed a higher sero-prevalence of Brucellosis in goats than in sheep (Table 1).

Table 1. Seroprevalance of Brucellosis in small ruminants based on species.

Factor	Group	Number of examined	Prevalence (%)	OR	95% CI for OR	p-value
Species	Sheep	202(52.6%)	4(1.04%)	0.55	0.1- 3.03	0.487
	Goat	182(47.4%)	2(0.52%)			
Total		384(100%)	6(1.56%)			

Seroprevalance of Brucellosis in small ruminants based on age

Age wise sero-prevalence of the brucellosis was 1 (0.26%) and 5 (1.30%) in young and adult animals respectively with no statistical significant ($P>0.05$) difference

Seroprevalance of Brucellosis in small ruminants based on sex

Out of total 120 (31.3%) male and 264 (68.7%) female animals, 1 (0.26%) and 5 (1.30%) were sero-positive for brucellosis respectively with no statistically significant (P>0.05) variation. Female animals were 0.435 (CI 0.050-3.76) times more at risk than male animals in this study.

Seroprevalance of Brucellosis in small ruminants based on history of RFM

The total seroprevalance of 249 (94.32%) female animals with no history of Retained Fetal Membrane (RFM) and 15 (5.68%) female animal with history of RFM was found to be 4 (1.6%) and 1 (6.67%) in animals with absence and presence of RFM respectively with no statistical significant difference (P>0.05).

Seroprevalance of Brucellosis in small ruminants based on history of abortion

History of abortion was also considered as one of the major outcomes of brucellosis in this study and about 12 (4.5%) animal with history of abortion and 252 (95.5%) animals with no history of abortion was examined and found with the prevalence of 2 (16.7%) and 3 (1.2%) respectively with highly statistical significant variation (p=0.000).

DISCUSSION

The overall seroprevalance of brucellosis of small ruminants in this study was (1.56%). In this study we found that, result of screening test (RBPT) is equal with the overall prevalence of the test (CFT). This may be due to the animal truly infected with brucellosis and no cross reaction with other microorganism, screening test is highly sensitive and CFT has highly specific. This finding is similar with the previous finding who reported a prevalence of 2.34% and 1.56% by RBPT and CFT respectively in Yabello district. However, higher than the sero-prevalence report of 1.37% and 0.4% in three selected districts of Somali region and Bahir Dar respectively [8].

The higher prevalence was reported than this study with prevalence rate of 5.8% in goats and 3.2% sheep, 4.2% in goats and 1.2% in sheep and 1.9% in goats in pastoral regions of Afar, South Omo zone and in Jijiga area respectively. The finding of current study was reported 13.7% in Tellalak district of Afar region using equal sample (384 samples) and laboratory technique with this study (RBPT and CFT test). The difference may be attributed due to the animal husbandry system, environmental conditions, age and sex of animal involved in study and reports higher prevalance than this study (4.6%) from 840 sampled animals using modified RBPT (MRBPT) and I-ELISA [9]. The difference in sample size and the variation in degree of sensitivity between CFT and I-ELISA could also be another source of difference in the prevalence reports.

In the current study the infection rate was compared among species and the prevalence rate was higher in sheep (1.04%) than goat (0.026%). Although not statistically significant, sheep were 0.550 (CI 0.1- 3.03) times more at risk of being sero-positive as compared to goat. The recent finding of brucellosis in sheep showed fair agreement On the contrary, it had reported higher (15.4% and 4.9%) prevalance of brucellosis in goat in selected district of Arsi and shewa zone, and Tellalak district of Afar region respectively mainly due to differences in husbandry system, susceptibility of the sheep and goat breeds in the particular area and number of sample in species [10].

When the sero-prevalence of small ruminant Brucellosis was compared between young and adult higher sero-positivity was observed in adult 5 (1.04%) than young 1 (0.26%) age groups even though, prevalence was found to be statistically not significant (P>0.05). This fact may be explained by the fact that brucellosis is a disease of sexual matured and pregnant animals, younger animals are resistant to infection and frequently clear an established infection, although latent infection can occur.

In the present study prevalence of 5 (1.30%) and 1 (0.26%) were recorded in female and male animals respectively. The prevalence was found to be statistically not significant (P>0.05), despite female animals was 0.435 (CI 0.050-3.76) times more at risk than male animals in this study. However in support of present fining and also reported the absence of statistical significant differance between two sexes. This could be due to the small sample size of males then females. Males are also kept in the herd for shorter period which decrease their exposure to the disease [11]. Similarly, it is an established fact that male animals are less susceptible to *Brucella* infection, due to the absence of erythritol, which stimulate the growth and multiplication of *Brucella* organism. But it difficult to draw strong decision in this study because the number female animals involved in this study was twice than that of male animals.

History of retained placenta was not found as a major risk factors (P>0.05) in the prevalence of brucellosis infection in this study. A prevalence of 6.7% and 1.6% were recorded in animals with and without history of retained fetal membrane respectively. In this study the occurrence of brucellosis have strong association with animal with history of abortion with statistical significant (p<0.05). The current study revealed the prevalance of brucellosis in animals with history of abortion was found to be (16.7%) and that animal without history abortion was found (1.2%), this show that animals with history of abortion have more risk than animal without history of abortion (OR=0.23 at CI, 0.024-2.2). This indicates that abortions or stillbirths and retained placenta are typical outcomes of brucellosis. Generally female animals contain four carbon sugar alcohols called erythritol which enhances the growth and multiplication of brucellosis and the amount this erythritol sugar increases in pregnant animals [12]. It is produced naturally by the developing foetus may favour multiplication of *brucella* where it causes degeneration and necrosis of the cotyledons leading to abortion from about the last months of gestation.

CONCLUSION

The present study in Ziway Dugda district shows that, there was existence of brucellosis in small ruminants and the overall result also reveals that sero prevalence of small ruminant brucellosis in the study area was low. In the current study, Sero-prevalence of brucellosis was higher in sheep than goats as well as female, adult aged and animal with history of abortion. Generally, existence of the disease in the study area has possible risk of spread in the area unless management system is improved and can pose great public health and economic threat since the livelihood of community is depends on these species of animals, providing milk, meat and cash income to cover family expenses for food and other essential consumers goods.

RECOMMENDATIONS

Based on above conclusion the following points should be considered to minimize the risk of this disease.

- Regular surveillance, screening and immunization should be implemented to control brucellosis in animals.
- Detailed epidemiological investigation of the three pillars of the disease should be conducted in the study to identify the risk factors with the occurrence and endemicity of the disease.
- Efforts should be made by the government and other concerned parties to raise awareness of the disease and its impact on public health.
- Moreover, Training should be provided for farmers and livestock attendants regarding effective sanitary and hygienic management practices during handling of animals and animal products such as aborted fetal materials and other body secretions.

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