Original Research

EAST AFRICAN JOURNAL OF PASTORALISM

Volume 3 Issue 2

Serological Screening of Brucellosis in Small Ruminant Flocks of Somali Regional State

Befekadu Urga Wakayo[a,*], Nasir Dayib[b], Kadar Ma'alin[c] and Tsegalem Abera[d]

[a] *Department of Veterinary Medicines, College of veterinary Medicines, Jigjiga University*
[b] *Department of Veterinary Medicines, College of veterinary Medicines, Jigjiga University*
[c] *Department of Veterinary Medicines, College of veterinary Medicines, Jigjiga University*
[d] *Department of Veterinary Medicines, College of veterinary Medicines, Jigjiga University*

ABSTRACT

Serological survey of small ruminant brucellosis was performed in seven districts of Ethiopian Somali Regional State from November 2016 to April 2018. Sera were extracted from 552 goat and 278 sheep jugular whole blood samples to screen for Brucella antibodies using commercial. Rose Bengal Test (RBT) assay kits. The animals belonged to Jigjiga (261), Gursum (163), Kebri-beyah (102), Awebere (100), Harshin (97), Shinile (57) and Dega-habur (50) district flocks. RBT Brucella sero-prevalence was contrasted in relation to species, sex, age group and source district of small ruminants. Overall, 11 (1.325 %) samples tested RBT Brucella sero-positive. All RBT sero-positive sera belonged to mature small ruminants ($p < 0.05$) and including 10 females ($p < 0.05$). RBT Brucella sero-positivity was relatively higher ($p < 0.1$) in goats 10 (1.8 %) than in sheep 1 (0.4 %). Goat RBT Brucella sero-positivity was higher ($p < 0.05$) in Harshin (7.8 %) followed by Gursum (2.5 %), Awebere (1.7 %) and Jigjiga (0.6 %). Findings suggest elevated natural Brucella infection exposure in Harshin pastoral doe flocks. Considering frequent row milk consumption trends plus lower hygienic practices in such areas, further studies are needed to accurately estimate prevalence, and risk factors of Brucella melitensis infection in all major food animal herds including related zoonotic transmission risks in Somali region.

Key Words: Antibody, B. melitensis, Epidemiology, Goat, Rose Bengal Test, Sheep

1. Introduction

Ethiopia is home to some 30.7 and 30.2 millions heads of sheep and goats, respectively (CSA, 2016 17). Pastoralist and agro-pastoralist areas account for 70.2 % and 41.5 % of national goat and sheep populations, respectively (Shapiro et al., 20117). National small ruminant (SR) flocksplay crucial role for supporting subsistence of millions of poor pastoralists/ agro-pastoralist and small holder farmers as well as significantly contributing to annual livestock export earnings. However, productivity of SR flocks in Ethiopia remains very low in terms of fertility, growth and off-take rates as well as milk and meat yields. This is attributed to constraints related to feed availability and quality; diseases and veterinary service; breeding and genetic improvement; marketing; etc (Legese and Fadiga, 2014; Gizaw, 2010).

Brucellosis is a bacterial zoonotic disease recognized for causing reproductive loss in livestock herds and debilitating illness or localized lesions of different body organs in humans. Sheep and goats are mainly infected by B. melitensis, the principal zoonotic Brucella species. Distribution and impacts of brucellosis vary relative to host (species, breed, sex and age) and environmental (climatic, husbandry, veterinary control programs, etc) factors. Despite having economic and zoonotic impacts, the epidemiology of brucellosis in east African pastoralist SR flocks remains largely unclear (Bonfoh et al., 2012; Radostits et al., 1997). Previously, few studies set in limited geographic areas had estimated low (1.6 to 1.7 %) sero- prevalence of SR brucellosis in Somali region of Ethiopia. However, such figures don't give representative epidemiological picture to diverse regional agro climatic and socio-economic contexts (Megersa et al., 2011). Hence, this study used Rose Bengal Test (RBT) to grossly screen the Brucella antibody sero-positivity in unvaccinated goat and sheep flocks reared under different geographic settings and livelihood zones of Somali regional state (SRS) in eastern Ethiopia.

* Corresponding author:Befekadu Urga Wakayo: fikeurga@gmail.com

Article Information:
Article Received for review: 15 April 2022
Article Reviewed: 15 May 2022
Revised Comments: 25 August 2022
Accepted for publication: 20 September 2022 Available Online: 31 December 2022
How to Cite this Article:
Befekadu U W, Nasir D, Kadar M, Tsegalem A (2022): Serological Screening of Brucellosis in Small Ruminant Flocks of Somali Regional State East African Journal of Pastoralism, 3(2):5-9.

2. Materials and Methods

Study Area and Populations

Serological survey of SR brucellosis was conducted in 7 weredas (districts) from Fafem (Awebere, Gursum, Harshyn,

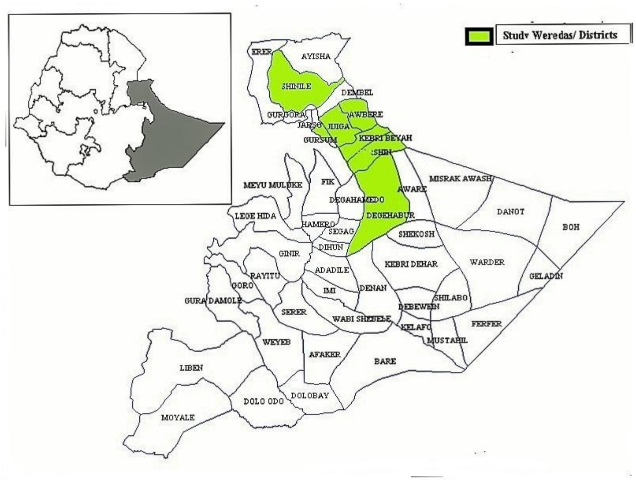

Figure 1: Map of the study area (shaded districts)

Jijiga and Kebribeya), Jerjer (Dega-habur) and Siti (Shinile) zones of SRS (Figure 1). SRS has a total land cover of 350,000 km2 located in eastern and south-eastern Ethiopia which is administratively organized in to 9 zone's, 68 wereda's and 4 city councils. The population of SRS was estimated to be around 5.3 million including 44 % females and 56 % males (CSA, 2016b). Altitudes in the region range from 200 meters above sea level (masl) insouthern/ central parts to 1,800 masl in northern areas. Agro-climatic conditions are mostly hot and arid or semi-arid at lower altitudes but cooler and sub-humid at higher altitudes. Annual rainfall levels in SRS range from 150 - 600 mm and daily ambient temperatures vary from 18 to 45Co. Four generic livelihoods exist in SRS including; pastoralism (60 %), agro-pastoralism (25%), farming and urban (15%) (Wikipedia. https://en.m.wikipedia.org/w; SCUK, 2004). Study wereda's represented pastoralist (Harshin and Shinile), agro-pastoralist (Awbare, Degahabur, Gursum and Kebribeya) and sedentary mixed crop-livestock farming (Awbare and Jigjiga) systems

Livestock are the region's principal source of subsistence by providing milk, meat, and cash income for foods and other expenses. The main livestock populations in SRS comprise of 7.5 million sheep, 6.6 million goats, 1.27 million cattle and 1.27 million camels (CSA, 2016a, 2004) . The study population included black head Somali (BHS) sheep and short eared Somali goat found in 7 study districts (Awgichew and Abegaz, 2008; Abegaz et al., 2008). Total corresponding SR populations varied in Fafem (0.85 and 0.60 million), Jerjer (1.4 and 0.72 million) and Siti (0.67 and 0.85 million) zones (CSA, 2016a, 2004)

Study Design

A cross-section survey of SR brucellosis was carried out in 7 SRS districts between November 2016 and April 2018. Exposure to natural Brucella (B. melitensis) infection was screened by RBT and was evaluated for variability relative to districts (reflects agro-ecology and livelihood system) and host signalment (species, sex and age group).

Sampling Approach and Sample Size Calculation

Guided by local officials and veterinary personnel, 1 or 2 villages having abundant SR populations were selected in each study wereda. Therein, households and study animals were sampled using a semi-randomized systematic approach. Briefly, every 3rd SR household encountered during visit to a village was sampled based on flock availability and informed consent for research participation. In each household, up to 10 sheep and/ or goats older than 6 months were sampled and this was continued until a minimum of 50 study animals were sampled per wereda. Sample size of study animals was calculated using the formula for simple random samples (Thrustfield, 2005) with expected prevalence of 1.6 % in goats and 1 % in sheep , and 0.01 desired absolute precision (99 % confidence level)

$$n = \frac{1.96xPexp(1\lambda Pexp)}{d_2}$$

Where n represents sample size $Pexp$ represents expected prevalence and d represent desired absolute precision Sample sizes of 309 and 194 were calculated for goat and sheep, respectively. However, 212 goats and 55 sheep were additionally sampled to improve accuracy of SR brucellosis sero prevalence estimates

Study methods

Approximately 6 to 10 ml jugular blood was collected from study animals using plain sterile vacutainer tube and needle. Blood samples were properly labeled (date, district, flock , animal , SR species, sex and age group) and transported to Jigjiga University laboratory in cold chain. Blood samples were left tilted overnight at room temperature to allow clotting and serum was harvested the next morning by gentle siphoning using sterile Pasteur pipette. Serum was placed in 2 ml cryovial labeled as the original blood sample and placed at -20 o C until serological screening. Sera screening was performed at Dire-dawa veterinary diagnostic laboratory using commercial RBT reagents for detecting Brucella antibodies (AHVLA) according to OIE (2004) protocol. Briefly, 25 l test and control (standard positive and negative) sera were mixed with 75 l RBT reagent (dye labeled B. abortus antigen) on 12 well white plastic plates. Mixtures were gently agitated for 4 minutes at ambient temperature on 3-directional rocker. Then, RBT reaction was read under bright light. Presence of outer ring shaped agglutination (Figure 2) was considered indicative of Brucella antibody sero-positivity probably reflecting prior natural B. melitensis infection exposure in unvaccinated flocks.

Data Analysis

Data pertaining to study period (year and month), location (district and livelihood system), animal signalment (species, sex and age group), and serum RBT Brucella screening results (sero - positive or negative) were entered on Microsoft Excel spreadsheet and analyzed by SPSS -16 software. Brucella sero-positivity (%) was calculated by dividing number (n) of RBT positive sera to tthat of total test sera (n) x

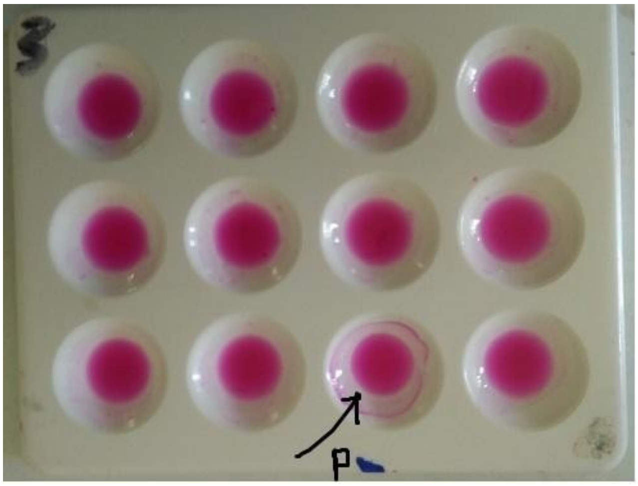

Figure 2: RBT Brucella agglutinin screening result indicating (P) positive reacting test (arrow)

100. Study variables were summarized and contrasted using frequency (n (% column/ row)) distribution tables. Variation of RBT Brucella sero-positivity relative to independent factors was analyzed using Chi square or Fishers exact tests. Statistical significance was established at alpha < 0.05 level.

3. Results and Discussion

3.1. Results

Serological survey of brucellosis was conducted 830 animals including more goats (66.5 %) than sheep (33.5 %). Specifically, frequency of goats and sheep varied with study districts (Chi Square = 20.65, p =0.002). Sex and age distributions of were comparable in respective species wherein majority (p < 0.05) of the animals were adult females (Table 1). Overall, 11 (1.3 %) sera tested RBT positive for Brucella antibodies. Total RBT sero-prevalence of brucellosis was significantly higher in Harshyn district surveyed in April 2018 (Fishers exact = 11.4, p =0.022) and showed relative elevation (Fishers exact = 2.98, p = 0.072) in goats 10 (1.8 %) compared to sheep 1 (0.4 %) as well as in adult 11 (1.6 %) and female 10 (1.5 %) than young and male animals (p < 0.05). Spatial RBT Brucella sero-positivity variations were limited to doe (Fishers exact = 10.6, p =0.030). Single RBT Brucella sero-positive mature ewe was observed in Gursum district surveyed in January 2017 (Table 2).

3.2. Discussion

Spatial-temporal study animals sampling discrepancies were mainly attributed to logistic accessibility of SR flocks (Jigjiga and Gursum > Dega-habur and Shinile districts) and availability of animals on scheduled field research visits. Predominant sampling of adult females and goats closely reflected average composition of SR flocks in pastoralist areas of Ethiopia like SRS. Maintaining larger breeding female stocks mainly indicates producer's interest in higher reproduction rates to maximize herd building (to cope with unexpected losses and/ or allow market/ household meat off-takes) and continuous milk supply particularly from goats (CSA, 2016a; Gizaw, 2010; Fikru and Gebeyew, 2015; Legese and Fadiga, 2014). Variable relative SR species sampling in Jigjiga (goats < sheep) and Shinile (goats > sheep) districts could be attributed to influences of variable biological tolerance to feed/ water scarcity (goat > sheep) as well as agro-climates (rainfall, crop farming intensity and grazing land availability) and market access (Jigjiga < Shinile) on flock composition as previously indicated (Fikru and Gebeyew, 2015; Legese and Fadiga, 2014).

Table 1: Frequency distribution of study setting and animal attributes n (column %)

Factors Categories	Total SR	Goats	Sheep
2016	74 (8.9) c	51 (9.3)	23 (8.3)
Years 2017	313 (37.7) b	221 (40)	92 (33.1)
2018	443 (53.4) a	280 (50.7)	163 (58.6)*
Months January	260 (31.3) a	177 (32.1)	83 (29.9)
February	140 (16.9) b	82 (14.9)	58 (20.9)*
March	145 (17.5) b	91 (16.5)	54 (19.4)
April	154 (18.6) b	114 (20.7)8	40 (14.4)
November	76 (9.2) c	52 (9.4)	24 (8.6)
December	55 (6.6) c	36 (6.5)	19 (6.8)
Awebere	100 (12.0) c	60 (10.9)	40 (14.4)
Dega-habur	50 (6.0)* d	36 (6.5)	14 (5.0)
Districts Gursum	163 (19.6) b	118 (21.4)	45 (16.2)
Harshyn	97 (11.7) c	64 (11.6)	33 (11.9)
Jigjiga	261 (31.4) a	159 (28.8)	102 (36.7) *
Kebri-beyah	102 (12.3) c	65 (11.8)	37 (13.3)
Shinile	57 (6,9) d	50 (9.1)*	7 (2.5)
Female	654 (78.8) a	434 (78.6)	220 (79.1)
Sex Male	176 (21.2) b	118 (21.4)	58 (20.9)
Age Young	97 (11.7) b	58 (10.5)	39 (14.0)
Groups Adult	697 (84.0) a	470 (85.1)	227 (81.7)
Old	36 (4.3) c	24 (4.3)	12 (4.3)

$Superscripts^{a,b\&c}$ reflect row category variation within a column whereas * indicates species variations within a row category at $P < 0.0$

Table 2: Distribution of RBT Brucella sero-positivity relative to study setting and animal signalment n (row %)

Factors	Categories	Total	Goats	Sheep
Years (GC)	2016	-	-	-
	2017	5 (1.6)	4 (1.8)	1 (1.1)
	2018	6 (1.4)	6 (2.1)	-
Months	January	4 (1.5)	3 (1.7)	1 (1.2)
	February	-	-	-
	March	1 (0.7)	1 (1.1)	-
	April	5 (3.2)	5 (4.4)	-
	November	1 (1.3)	1 (1.9)	-
	December	-	-	-
Districts	Awebere	1 (1.0)	1 (1.7)	-
	Dega-habur	-	-	-
	Gursum	4 (2.5)	3 (2.5)	1 (2.2)
	Harshyn	5 (5.2)a	5 (7.8)	-
	Jigjiga	1 (0.4)	1 (0.6)	-
	Kebri-beyah	-	-	-
	Shinile	-	-	-
Sex	Female	10 (1.5)	9 (2.1)	1 (0.5)
	Male	1 (0.6)	1 (0.8)	-
Age Groups	Young	-	-	-
	Adult	11 (1.6)	10 (2.1)	1 (0.4)
	Old	-	-	-

Using RBT, the current study estimated SR brucellosis sero-prevalence of 1.3 %. This wa considered to signify low endemic transmission of natural B. melitensis infection in the study population. RBT is regarded reliable for screening natural Brucella infection in unvaccinated populations. Nevertheless, test specificity could be lowered by immunological cross reactions from any Brucella species having smooth cell wall LPS (B. melitensis, B. abortus and B. suis) and their attenuated vaccine strains (Rev-1, B. melitensis M5 and B. suis S2) as well as unrelated bacteria (F. tularensis, E. coli O:157/ O:116 and Y. enterocolitica O:9) (Spickler/ Rovid, 2018; Quin et al., 2004; Radostitis et al., 2000). However, RBT – CFT tests exhibit average agreement of 80 % in Ethiopia suggest fairly strong specificity of RBT results to Brucella species in general (Abunna et al., 2018) (Tegegn et al., 2016; Megersa et al., 2011). Considerable host preference of Brucella species and absence of brucellosis vaccination program further specify RBT + SR sera interpretation to natural B. melitensis infections (past or ongoing). The current SR brucellosis RBT sero-prevalence was comparable to previous estimates based on RBT (1.72 %) and CFT (1.37 = 1.65 %) in SRS (Megersa et al., 2011; Teshale et al., 2006) as well as RBT (2.34 %) and CFT (1.56 %) in Borana Ethiopia (Addis, 2013). In contrast, higher sero-prevalence rates were reported based on RBT (9.4 - 13 %) and CFT (4.8 - 12.35 %) from Afar region (Gebretsadik and BISHOFTU, 2016; Ashenafi et al., 2007)(Tegegn et al., 2016) as well as 34.3 % in Tigray (Habtamu et al., 2015) and 5.2 % in Hammer and Dasenech Goats from South Omo (Ashagrie et al., 2011) based on RBT alone. Meanwhile, SR brucellosis sero-prevalence was currently higher (p < 0.05) in Harshin district (5.2 %) particularly in goats (7.8 %) but varied weakly relative to sex and age group of SR (p > 0.1). Previous studies in SRS and Borana similarly observed no age and sex related SR brucellosis sero-prevalence variation. Meanwhile, higher brucellosis sero-prevalence was reported in goats, adult SR, late aborting SR, female SR in 3 - 4 parity and in larger flocks of Afar region (Gebretsadik and BISHOFTU, 2016; Ashenafi et al., 2007). Ethiopian SR brucellosis sero-prevalence trends could suggest epidemiological importance of agro-climate modulated host biology, demography and husbandry factors. In hot arid areas, susceptible (breeding females) and infectious (old infected females) goat populations tend to be larger on account of their environmental adaptations and dairy functions. Therein, grater scarcity of feed and water resources could force greater aggregation and mixing of goat flocks elevating risk of infection for susceptible populations by females aborting and silently kidding during transition from long dry to rainy season.

Epidemiology of SR brucellosis can vary from region to region as well as between flocks in a region. This is attributed to interactive influence of host demography and biology; environmental and management conditions; and pathogen biology related factors on natural Brucella infection dynamics. Susceptibility to B. melitensis infection is higher in goats compared to sheep. It is also influenced by age (young < older animals), sex (female > male) and reproductive status (sexually active & pregnant > sexually inactive) of hosts. Susceptible hosts often get infected by ingesting large quantities of Brucella shed to external environment with fetal fluids, fetus, and placenta after abortion or silent carrier parturition. Brucella can survive and remain infective for several months in cool wet environment. Flock size and structure as well as fresh female management practices can affect the maintenance and spread of infection within flocks whereas stock movement and mixing facilitate transmission of infection across flocks. Seasonal or annual climatic changes can alter the demography, management and contact of infected and susceptible host thereby leading to temporal infection transmission trends (Radostits et al., 1997). Extensive mobile use of rangeland pastures and water bodies in arid pastoralist areas can increase risk of contact between livestock and wild life herds carrying geographically or seasonally limited diseases such as brucellosis (Megersa et al., 2011).

4. Conclusion and Recommendations

Potential of RBT Brucella ser-positivity to inflate infection risks but absence of any vaccination to the disease across Ethiopia would reduce such tendencies. On positive note, this work studies larger and more diverse small ruminant populations found in Somali region of Ethiopia. Therein, low endemic SR brucellosis circulation was indicated mainly in mature doe flocks of Harshin pastoralist district. Host, climate and husbandry factors could show relative effect on natural SR Brucella infection exposure in the study. Frequent row milk (goat or other adjacent dairy species) consumption, poor hygienic practices and unprotected handling of aborted materials including fetus and placenta could expose Somali pastoralists/ agro-pastoralist to zoonotic brucellosis. Hence, further research is needed to deepen understanding on level, patterns and determinants of Brucella melitensis transmission in representative regional herds therein addressing zoonotic and economic risks. Raising awareness of communities on exposure risks and hygienic plus dietary zoonotic brucellosis prevention approaches also represents a critical priority.

Conflict of Interest

Authors declare that there is no conflict f interests involve in publishing this research paper.

References

Abegaz, S., Abebe, G., and Awgichew, K. (2008). Sheep and goat production systems in ethiopia. *Sheep and goat production handbook for Ethiopia. Ethiopia sheep and goat productivity improvement program (ESGPIP)*, pages 27–32.

Abunna, F., Merid, B., Goshu, G., Waktole, H., and Mammo, G. (2018). Assessment of major reproductive health problems, their effect on reproductive performances and association with brucellosis in dairy cows in bishoftu town, ethiopia. *Journal of Dairy Veterinary and Animal Research*, 7:14–20.

Addis, M. (2013). Small ruminant brucellosis: serological survey in yabello district, ethiopia. *Asian Journal of Animal Sciences*, 7(1):14–21.

Ashagrie, T., Deneke, Y., and Tolosa, T. (2011). Seroprevalence of caprine brucellosis and associated risk factors in south omo zone of southern ethiopia. *African Journal of Microbiology Research*, 5(13):1682–1476.

Ashenafi, F., Teshale, S., Ejeta, G., Fikru, R., and Laikemariam, Y. (2007). Distribution of brucellosis among small ruminants in the pastoral region of afar, eastern ethiopia. *Revue scientifique et technique*, 26(3):731.

Awgichew, K. and Abegaz, S. (2008). Breeds of sheep and goats. *Sheep and goat production handbook for Ethiopia: Ethiopia Sheep and Goat Productivity Improvement Program (ESGPIP)*, pages 5–26.

Bonfoh, B., Kasymbekov, J., Dürr, S., Toktobaev, N., Doherr, M. G., Schueth, T., Zinsstag, J., and Schelling, E. (2012). Representative seroprevalences of brucellosis in humans and livestock in kyrgyzstan. *EcoHealth*, 9:132–138.

CSA (2004). Ministry of finance and economic development ethiopia (2004). livestock aerial survey in the somali region. ecosystems consultants and save the children uk.

CSA (2016a). Federal democratic republic of ethiopia 2016 17. agricultural sample survey 201617 [2009 e.c.].

CSA (2016b). Population and housing census of ethiopia; statistical report for somali region.

Fikru, S. and Gebeyew, K. (2015). Sheep and goat production systems in degehabur zone, eastern ethiopia: Challenge and opportunities. *J Adv Dairy Res*, 3(134):2.

Gebretsadik, M. T. and BISHOFTU, E. (2016). Seroprevalence of brucellosis and isolation of brucella from small ruminants that had history of recent abortion in selected kebeles of amibara district. *Afar region, Ethiopia*.

Gizaw, S. (2010). *Sheep and goat production and marketing systems in Ethiopia: Characteristics and strategies for improvement*, volume 23. ILRI (aka ILCA and ILRAD).

Habtamu, T., Richard, B., Dana, H., and Kassaw, A. (2015). Camel brucellosis: its public health and economic impact in pastoralists, mehoni district, southeastern tigray, ethiopia. *Journal of Microbiology Research*, 5(5):149–156.

Legese, G. and Fadiga, M. (2014). Small ruminant value chain development in ethiopia: Situation analysis and trends.

Megersa, B., Biffa, D., Abunna, F., Regassa, A., Godfroid, J., and Skjerve, E. (2011). Seroprevalence of brucellosis and its contribution to abortion in cattle, camel, and goat kept under pastoral management in borana, ethiopia. *Tropical animal health and production*, 43:651–656.

Radostits, O. M., Blood, D. C., and Gay, C. C. (1997). Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses.

Tegegn, A. H., Feleke, A., Adugna, W., Melaku, S. K., et al. (2016). Small ruminant brucellosis and public health awareness in two districts of afar region, ethiopia. *J Vet Sci Technol*, 7(335):2.

Teshale, S., Muhie, Y., Dagne, A., and Kidanemariam, A. (2006). Seroprevalence of small ruminant brucellosis in selected districts of afar and somali pastoral areas of eastern ethiopia: the impact of husbandry practice. *Revue de médecine vétérinaire*, 157(11):557.