

Sero-prevalence, Risk Factors and Community Perceptions of Small Ruminant Brucellosis in Wachile District of Borena Zone, Ethiopia

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Abstract

This study was conducted to estimate the seroprevalence of brucellosis in small ruminants of Wachile district Borena. Three hundred twenty-four samples (188 goats and 136 sheep) were proportionally selected from five kebeles. Five animals were sampled systematically from each flock at a Kth interval until the sample size of kebeles was attained. All animals were sampled in a flock with less than five animals, whereas from flock size above five animals, a maximum of five were sampled at an interval of $n/N = k^{\text{th}}$. The assumed risk factors like physiological status, age, sex, flock size, and other factors recorded. Serum prepared from blood samples collected for serological tests (Rose Bengal Plate Test and indirect ELISA). Data was analyzed using Stata-14 software. Seroprevalence association with assumed risk factors were compared by Chi-square using Fisher's exact test. The study revealed a 5.2% seroprevalence by a screening test, of which four positives were confirmed by iELISA (1.23%) with seroprevalence of 1.6% and 0.74% in goats and sheep, respectively. There was no significant ($p > 0.05$) association observed in age, sex, flock size, BCS, and introduction of new animals. Reproductive parameters had a significant association with seroprevalence ($p < 0.05$). Perception of the community shows they have no experience of proper disposal. Protection during parturition shows 26% use soap and 22% use water only. Poor community knowledge about the disease and its zoonotic significance was recorded. Low seroprevalence in small ruminant observed which was more associated with the introduction of new animals. A significant association between reproductive parameters and seroprevalence indicates the disease related to reproduction. Therefore, attention should be given even though the prevalence is low.

Introduction

Ethiopia has a large number of small ruminants, estimated at 31.30 million sheep and 32.74 million

goats (1). They are managed in all agroecological zones, mainly in arid and semiarid environments. Most pastoral communities, including the Borana

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community, rear a large goat population in addition to camels and cattle because of their adaptive nature in harsh environments and use for immediate cash income, apart from a source of food (2-4).

However, despite its significant importance of small ruminants', reproductive diseases have prevented their productivity. Reproductive problems are usually manifested by; infertility, abortion, stillbirth and weak offspring (5). Among others, brucellosis has been recognized as one of the neglected tropical zoonotic diseases responsible for reproductive problems due to the causative agents affinity to reproductive organs. The causative agent is fastidious bacteria under the genus *Brucella* (6,7). Different species of the genus *Brucella* are responsible for the disease among those, *B. melitensis* and *B. ovis* are the most common cause of brucellosis in sheep and goats (8,9), while *B. melitensis* is the primary cause of goat brucellosis (10).

Brucellosis is an overlooked disease by animal owners and policymakers, which affects the economy and human health in various ranges. Among health problems reported, retarded growth of the newborn, decreased kidding rate, culling due to infertility, decreased milk yield, abortions, stillbirths, weak neonate, absenteeism at working hours (in humans) and cost of treatment (11-13). Generally, the risk of infection is influenced by the species of *Brucella* involved such as *B. melitensis* more serious public health hazard than *B. abortus* and it is responsible for 70% of all infections (6, 14).

In Ethiopia, the disease was reported in 1970 (15). There was no vaccination practice in Ethiopia before and after the first report. Any positive case reported is designated as caused by natural infection. Following the first report, various studies have demonstrated seropositivity associated with abortion, retained placenta or stillbirth in small ruminants (16, 17). Observation showed that 58.68 % of goats and 17.73 % of sheep flocks had abortions in a 12-month survey, with a prevalence

of 16.1 % and 12.6 % in doe and ewes at the individual level (18). Furthermore, the overall seroprevalence of brucellosis in small ruminants was higher in goats than in sheep (19). The overall seroprevalence ranges from 0.24% in Chiro West Haraghe zone (20) to the highest seroprevalence of 17.36% reported in Elwaya, Borena zone (21). In the Borena pastoral community, few districts were studied, despite several districts and large livestock populations in the area (22). Most reports are related to serological surveys and isolation of the organism is minimal due to facility shortage (23).

Wachile district is among the neglected districts in the Borana zone due to road inaccessibility. Although there are frequent abortion cases reported in small ruminants of the district, no survey was conducted. In line with this information gap, this study was designed to address it. Therefore, the objective of the current study was to assess the seroprevalence of brucellosis in small ruminants, associate it with assumed risk factors, and assess the perception of the community.

Materials and methods

Description of the study areas

The study was carried out in the Wachile district of the Borena zone, Oromia region. Wachile district covers an area of approximately 13,000 km² and is divided into 13 kebele (the smallest government administrative units). It is located at a distance of 695 km south of Addis Ababa and geographical coordinates of 04.55° N latitude and 39.06° E longitude with an altitude of 1095.0 m.a.s.l. The agroecology of the district is characterized by lowland. The mean annual rainfall of the study area ranges from 250 to 700 mm, and the mean annual temperature varies from 19 to over 42°C. The estimated livestock population of Wachile district is 101,436 Camels, 130,772 Goats, 90,618 Sheep, 1,383,616 Cattle, 3,350 Mules and 5,411 Donkeys (24, 25).

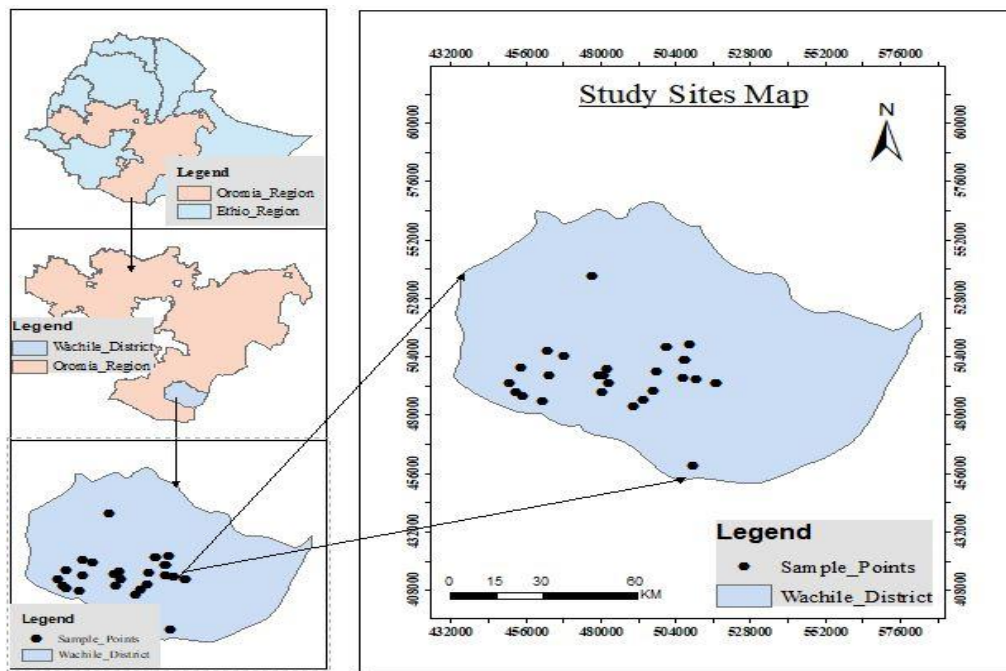


Fig. 1. Geographical location of the Wachile district within Oromia region Ethiopia.

Study population

The study population is small ruminants kept under the extensive management system in study districts. Small ruminants found in thirteen kebeles were considered in the study population. Among these, five kebeles were used as the target population, and the determined sample size was selected proportionally based on the population of small ruminants that existed in selected kebeles. Sheep and goats regardless of sex and age above six months were included in the selection. Kebeles used for sampling were Wachile, Harajarte, Webi, Walensu and Kakallo. All these kebeles were managed under the extensive pastoral management system. There was no *Brucella* vaccination history.

Study design and sample size determination

A cross-sectional study was conducted from August 2023 to January 2024 in the Wachile district of Borena Zone. The total sample size was estimated by the formula given for a simple random sampling procedure (26), and calculated using a 95 % confidence interval level at 5% desired absolute precision. Expected prevalence used from nearby

district data since there was no previous report of the prevalence of sheep and goat brucellosis in Wachile district. Therefore, the prevalence of 9.2% in goats and 6.1% in sheep from the Yabello district of the Borena zone (near the same agroecological zone) was reported (27). So, we used an expected prevalence of 9.2 % and 6.1% for goats and sheep, respectively; to get a minimum sample size. $n = (1.96)^2 P_{exp} (1 - P_{exp}) / d^2$ formula is used.

Where n = required sample size, d = desired absolute precision and P = expected prevalence by substituting the value; the calculated value of 216 sample sizes was determined. However, the sample size increased to 324 small ruminants by adding around (50%) to increase precision. Out of these sample sizes, 188 goats and 136 sheep were sampled.

Sampling Strategy

A multistage sampling method was used to select sampling units from different flocks. Wachile district was selected purposively among Borena zone districts based on the information on the study gap. A total of 5 kebeles were selected out of 13

kebele because of high small ruminant population compared to other kebeles. To select individual animals from selected kebele, the proportion of total goat and sheep population in each kebele was calculated and then, based on the size of goat and sheep, the determined sample size was allocated to five selected kebeles. To get a proportional sample size in each kebele the total sample size was divided by the total size of the population in five selected kebeles and calculated into percentages (26). Accordingly, for sheep the sample size was 132 and the total population were 43362 which was calculated as $(136/43,362)$ gotten 0.00314 and for goats, the sample size was 188 and the total population in five kebele were 52,990, which was again calculated as $(188/52,990)$ obtained 0.0036 and these two outputs used as multiplying factor in each kebele population. Then the allocated sample size in each kebele was further again reallocated to flocks within each selected kebele; if the total flock size was less than five animals, then all animals were sampled and if the flock size was above five then a maximum of five animals per flock has been sampled in each kebeles until the required sample size in that kebele attained. Each animals were selected using a systematic sampling technique from each flock by putting animals in a fence or yard.

Sample collection techniques

Information was collected about individual animals at the time of blood collection. Sex, species, body condition scores and flock size, pregnancy stage, abortion history, reproduction status, history of retained placenta, kind of kids at birth and introduction of new to the flock were recorded. The flocks were categorized as small (when less than 15 sheep/goats), medium (between 15 to 30 sheep/goats) and large (when greater than 30 sheep/goats) flock sizes. Parity categorization, body condition scoring and age classification are recorded based on the guidelines established by research institutions and scholars (28-34). Abortion was defined as the loss of a fetus before 140 days of pregnancy (28). The gestation stage of aborted

fetuses was categorized as first trimester (less than 50 days pregnancy), second trimester (between 51-100 days) and third trimester (101-154 days) (32). The consent of the animal owner was obtained before blood sample collection. If an animal owner refused to accept the consent, then proceeded to the next owner. Sample collection was managed early in the morning while small ruminants were in their collecting pen otherwise animals would be released for browsing and grazing. It would be difficult to collect samples once they were scattered in the bush. Five to eight millilitres of blood were taken from the jugular vein of each selected animal using a venoject needle and non-heparinized vacutainer tube. After collection, each test tube was coded to coincide with the record sheet. Then the sampled blood was placed in a slant position to allow clotting at room temperature and then serum was decanted into cryovial test tubes. Then Serum samples were kept at -20°C in the Wachile district veterinary clinic until transported using cold chain to the Yabello regional veterinary laboratory to conduct a serology test.

Serological test

Two types of serological tests were employed for the detection of *Brucella* antibody: Rose Bengal Plate Test (RBPT) (as a screening test) and i-ELISA (indirect Enzyme-Linked Immunosorbent Assays) as a confirmatory test (DEXX Montpellier and ID vet (IDScreen®) in Louis Pasteur institute France, respectively). All serum samples prepared were subjected to screening tests using Rose Bengal Plate Test (RBPT), after screening all positive samples were further confirmed by iELISA (indirect Enzyme-Linked Immunosorbent Assays) at Yabello regional veterinary laboratory, according to the standard given by (9). Following this protocol, it was done by adding 25 μl of antigen and 75 μl of serum onto a plate. The antigen and test serum were then completely mixed with the plastic applicator, shaken for 4 min, and the degree of agglutination was visually inspected and recorded as positive or negative for the presence or absence of agglutination (8, 34, 35). RBPT-positive blood

sample should be confirmed by a definitive test (36) because it is highly sensitive which results in a false negative. Therefore, Sera tested positive by RBPT were subjected to ELISA (Enzyme-Linked Immunosorbent Assays) for confirmatory test popular and more specific test as a standard assay for the diagnosis of brucellosis, it measures IgG, IgA and IgM antibodies (37).

Questionnaire survey

Open and closed semi-structured and structured questionnaires were designed to evaluate community perception of small ruminant brucellosis in the study area. Information about each flock was recorded by interviewing owners or herders using the local language (Afaan Oromo). The consent from each interviewee was obtained before the beginning of the questionnaire. For this survey, respondents were selected purposely, from flock owners whose sheep and goats were sampled were selected and interviewed. For the questionnaire survey, heads of household or any individual from the family member whose age is greater than 18 years were considered for interview. Accordingly, a total of 73 pastoral households were interviewed, of these 50 male and 23 female respondents were involved.

Data management and analysis

Data were recorded and coded in Microsoft Excel spreadsheets before being transferred to statistical software for analysis (Stata TM 14.0, Stata

Corporation, and College Station, Texas, USA). The data generated were analyzed for seroprevalence using descriptive statistics and the association of seroprevalence with kebele, species, sex, flock size, age group, BCS and entry of new animals to the flock were analyzed with chi-square (χ^2) test using Fisher exact test; Individual seroprevalence was calculated the number of individual positive animals divided by the total samples size and multiplied by 100. Whereas, flock level prevalence was also calculated by dividing the number of flocks having at least one positive animal by the total number of examined flocks and multiplied by 100. For all analyses, a $P < 0.05$ is taken as statistically significant.

Results

Seroprevalence of brucellosis in small ruminant

The study conducted in small ruminants at Wachile district revealed seventeen positive samples using Rose Bengal Plate Test, of these positive samples only four of them were confirmed positive using I-ELISA ($4/324 * 100 = 1.23\%$). Among these positives goats and sheep share different seroprevalence of 1.6% and 0.74%, respectively. The seroprevalence of brucellosis at the kebele level shows that only two kebele had positive results as shown in Table 1. The difference between species showed statistical significance ($p < 0.05$), where goats had high seroprevalence compared to sheep.

Table 1. Seroprevalence of brucellosis in selected pastoral associations and species of small ruminant

Factor	Number	RBPT positive	I-ELISA positive	Prevalence (%) using I-ELISA	χ^2	<i>p-value</i>
Kebele						
Wachile	38	2	0	0		
Walensu	64	6	2	3.12 [2.68-3.55]		
Kakallo	68	3	0	0		
Webi	87	3	2	2.3 [1.98-2.62]	4.84	0.303
Harjarte	67	3	0	0		
Species						
Goats	188	10	3	1.6 [1.4-1.9]		
Sheep	136	7	1	0.74 [0.67-0.81]	0.48	0.49
Total	324	17	4	1.23 [1.11-1.35]		

Sero-prevalence of brucellosis in different assumed biotic risk factors

Seroprevalence results using i-ELISA showed different prevalences as indicated in Table 4. The sex of small ruminants showed females were relatively higher than males. Out of 73 flocks, only four flocks showed at least one positive recorded (4/73) *100 = 5.5% overall flock seroprevalence obtained. Seroprevalence at the flock level showed large flock size had the only positive flock with seroprevalence of 10.8% [9.7-11.9] compared to

zero prevalence in small and medium flock sizes. The adult age group had relatively higher than the young age group with a seroprevalence of 1.4 and 95% CI of 1.24 – 1.56, whereas the seroprevalence of the young was 0.93 [0.75-1.11]. Small ruminants with poor body condition scores showed higher than medium and good body condition scores. Animals contact with a new group of animals either at the market or introduced in the flock showed all positive for I-ELISA (Table 2).

Table 2. Seroprevalence of brucellosis in different assumed risk factors using Fisher exact

Risk factor	Number	RBPT +ve	IELISA +ve	Prevalence of ELISA [95%CI]	χ^2	p-value
Sex						
Female	216	11	3	1.4 [1.24-1.56]	0.127	0.72
Male	108	6	1	0.93 [0.75-1.11]		
Flock size						
Small	14	3	0	10.8 [9.7 -11.9]	2.96	0.23
Medium	22	2	0			
Large	37	12	4			
Age group						
Young	108	4	1	0.93 [0.75-1.11]	0.127	0.72
Adult	216	13	3	1.4 [1.24-1.56]		
BCS						
Good	214	11	1	0.48 [0.38-0.57]	3.0789	0.215
Medium	77	3	2	2.6 [2.24-2.96]		
Poor	33	3	1	3.03 [2.97-3.09]		
Introduction of new animals						
Yes	129	12	4	3.1[2.8-3.4]	6.1221	0.24
No	195	5	0			

Reproductive parameters and brucellosis seroprevalence in small ruminant

Reproductive parameters were measured to learn about any difference in seroprevalence of brucellosis. Among sampled animals' brucellosis sero-prevalence difference were not significantly ($p>0.05$) different between monoparity and multiparity. Whereas, only lactating animals had seropositive under the physiological status of animals, all others (pregnant, dry and young) were negative. Animals with abortion history and abortion at third-trimester pregnancy and with a history of the retained placenta had seropositive

results as shown in Table 3. Seroprevalence was a significant association ($p<0.05$) with abortion history, trimester period and retained placenta. Weak lamb and kidding were not associated with seropositive results. Among females' animals with the history of abortion is 20% (43/216) which is quite large, but still brucellosis seropositive is lower compared to the percentage history record. There are several pathogens responsible for second and third-trimester pregnancy abortion. Among animals with a history of abortion, 46.5% (20/43) were aborted in the trimester gestation period.

Table 3. Physiological parameters of sampled animals in relation to seroprevalence

Parameters	Number	RBPT +ve	I-ELISA +ve	Seroprevalence of I-ELISA	χ^2	p-value
Parity	No parity	68	1	0	2.04 [1.64-2.43]	1.40
	Mono parity	49	4	1		
	Multiparity	99	6	2		
Physiology status	Lactating	53	5	3	5.66 [5.02-6.30]	10.34
	Pregnant	25	1	0		
	Dry	70	4	0		
	Young	68	1	0		
Abortion history	Yes	43	4	3	6.97[6.18-7.75]	20.71
	no	173	7	0		
Stage of Abortion	First trimester	5	0	0	5.56 [4.47-6.65]	17.20
	Second trimester	18	2	1		
	Third trimester	20	2	2		
Retained placenta	Yes	48	4	3	6.25[5.54-6.96]	11.62
	No	168	7	0		
Weak lamb/kid delivery	Yes	31	1	0	1.6[1.42-1.78]	0.43
	No	185	10	3		

Questionnaire results

Demographic characteristics of respondents

The study comprised a total of 73 participants, selected from 5 Pastoral associations located within Wachile, district of Borena zone. All of interviewed participants were engaged in rearing small ruminant, 68.5% of them were male and 41% of them were in the age group greater than 41 years old. While a substantial proportion of the interviewed participants 79.5% (n=58) were owners of the flock and 78% (n=57) were illiterate as shown in Table 4. Community awareness about the disease is limited. The result of this study shows that most of the respondents, 89 % (n=65) and 93% (n=68) did not hear about brucellosis and have no awareness about its zoonotic potential, respectively. A large proportion of participants 49.3 % (n= 36), normally assist delivery. However, all of them that

means 100 % (n=73) were used their bare hands (without using protective glove) while helping their animals during parturition and only 19 % (n= 26) of them was properly wash their hands after contact with abortion materials. Furthermore, 85 % (n=62) of the respondents said that they consume raw animal milk regularly. In addition to raw milk; 2 % (n=3) of the respondents were consumed raw meat. Despite, around 49.3 % (n=36) of respondents had experienced abortion in their flocks; the majority of households in this study were unable to implement any control measures to prevent the spread of disease. None of the respondents indicated to neither bury nor burn aborted fetuses and fetal membrane; rather 17.8 % (n=13) and 31.5 % (n=23) of them have experience of throw it on the field and feeding them to dogs, respectively as shown in (Table 5 and 6).

Table 4. Socio-demographic characteristics of respondents

Characteristics of respondents	Categories	No (%)
Sex	male	50 [68.5]
	female	23 [31.5]
Age groups	<25	15 [20.5]
	26-40	28 [38.5]
	>41	30 [41]
Responsibility	owner	58 [79.5]
	attendants	15 [20.5]
Kebele	Wachile	9
	Walensu	15
	Kakallo	16
	Webi	18
	Harjarte	15
level of education	illiterate	57 [78]
	Primary school	12 [16.5]

Table 5. Knowledge, attitude and practice of community about brucellosis

Knowledge, attitude and practice of respondents	Response Category	Frequency (%)
Abortion in flock	yes	36 [49.3]
	no	37 [50.7]
Management of aborted fetus and fetal membrane	burying	0 [0]
	Given to dog	23 [31.5]
	Disposed at open field	13 [17.8]
	not encountered abortion	37 [50.7]
Assisting delivery	yes	36 [49.3]
	no	37 [50.7]
safety after assisting delivery	washed hands properly using soap	19 [26]
	washed hands only using water	16 [22]
	clean hand by soil and plant leaf	1 [1.3]
	have not encountered abortion	37 [50.7]
Using glove during assist	yes	0 [0]
	no	36 [49.3]
	have not encountered	37 [50.7]

Table 6. Community practice towards prevention and control of the disease

Factors	Response	Frequency (%)
Introducing new animal to flocks	yes	25 [34.3]
	no	48 [65.7]
Consume raw milk	yes	62 [85]
	no	11 [15]
Consume raw meat	yes	2 [3]
	no	71 [97]
Know/heard brucellosis	yes	8 [11]
	no	65 [89]
Know the way of transmission from animal to animal and human	yes	4 [5]
	no	69 [95]
know zoonosis of brucellosis and species affected	yes	5 [7]
	no	68 [93]
know prevention and control	yes	4 [5]
	no	69[95]

Discussion

This study shows that the overall small ruminant seroprevalence of brucellosis in selected pastoral associations of Wachile district was 1.23% with 95% CI [1.11-1.35], whereas seroprevalence in goats and sheep was 1.6% and 0.74%, respectively; seroprevalence difference between species was not statistically significant ($p > 0.05$). The current finding was lower than with seroprevalence of 3.2% and at species level seroprevalence of 3.7% and 1.4% in goat and sheep, respectively; from the same agroecological zone of Borena pastoralist area (38). In addition, other studies in the Borena zone also showed high seroprevalence, where the overall seroprevalence of 8.1% and at species level seroprevalence of 9.2% and 6.1% in goat and sheep, respectively (39); the overall seroprevalence of 4.8%, and species level seroprevalence of 5.8% in goats and 3.2% in sheep reported from Afar region (35). The current seroprevalence of brucellosis in small ruminants is relatively similar with overall seroprevalence of 1.6% as reported by (40, 41). A seroprevalence of 1.6% was reported from Konso (42). Seroprevalence of 1.23% from Korahey Zone, Somalia region (43) and with seroprevalence of 3.5% from Southern Tigray reported by (44). Moreover, seroprevalence findings of 1.7% in goats

and 1.6% in sheep from Afar and Somali pastoral regions reported by (45) and seroprevalence of 1.9% in goats and 1.2% in sheep from the Somali region were almost the same seroprevalence with the current study findings (16). Asmare *et al* (2013) (46) also reported at individual animal level analysis, seroprevalence of 32.5% (95% CI 21.9, 43.0) was recorded in pastoral production system, followed by agro-pastoral, 13.0% (95% CI 7.0, 19.0) and sedentary production system, 3.6% (95% CI 1.3, 6.0), complement fixation test was used for confirmatory diagnosis, which was higher than the current study findings (46). The observed difference in seroprevalence could be due to the variation in the use of confirmatory tests and even the skill of the individual to conduct the test, sensitivity and specificity of the various test, agro-ecological location and sample size and sampling strategy, management and production system could play a major role.

It was observed that goats were relatively had higher seroprevalence than sheep, though it was not statistically significant. This finding agrees with reports from Ethiopia and other African countries. 5.8% and 3.2% in goats and sheep, respectively reported from Afar region, North Eastern Ethiopia (35). This finding agrees also with the results of

(47) who reported a seroprevalence of 19.6% and 9.4% in caprine and ovine respectively from animals slaughtered at abattoirs in Abuja, Nigeria, which indicated seroprevalence of brucellosis in caprine dominates compared to ovine. This may be associated with high susceptibility of goats than in sheep and higher excretion of the pathogens and transmission might be more than in sheep.

Seroprevalence association between sex, flock size, age group and body condition score showed a non-significant association ($p > 0.05$). Prevalence is also very low compared to other studies, this might be associated with management or other unforeseen factors. However, the history of animals in contact with newly introduced ones showed a significant seroprevalence association ($p < 0.05$), which might indicate that animal movement has a great contribution to the spread of disease. Other studies indicated that large flock size has high seroprevalence compared to small flock size. As reported by (44) who stated larger flock sizes were found to be significantly ($p < 0.05$) associated with *Brucella* seropositivity in small ruminants. This finding indicates that herd size and animal density are directly contributing to seroprevalence of the disease (5). There is a tendency of increasing prevalence among adults kept in larger flock (46) and reproductive inefficiency has been associated with *Brucella* exposure (44). There was significant difference observed between age group and sex in other studies contrary to the current study. This study might be associated with low seroprevalence in the area. Sexual maturity in female animals plays a role in *Brucella* multiplication (5). The high seroprevalence of brucellosis in females might be due to high concentration of erythritol, which is scarcely produced in males' reproductive organs. The extended period of stay of female animals on the farm exposes them to *Brucella* organisms and hence the chances of acquiring infection are more than the males (5). Statistically significant ($p < 0.05$) seroprevalence of brucellosis was observed in animals with a history of abortions, those at their third-trimester gestation stage, and animals with a

history of previous retained fetal membranes. This finding is in agreement with the study reported by (48) in central Ethiopia and (49) in northern Ethiopia who reported 44.3% and 40.3%, respectively.

Pregnant animals are more susceptible to infection by the organism than sexually immature animals. Susceptibility also increases with pregnancy, as stage of gestation increases (50). This indicates that abortions or stillbirths and retained placenta are typical outcomes of brucellosis (5). This might be associated with the tropism/preference of *Brucella* species to the key target cells called trophoblast. Growth of *Brucella* inside trophoblast is apparently enhanced synergistically in the presence of high concentrations of steroid hormones and erythritol during the final gestation of ruminants. The capacity to replicate readily and extensively in trophoblasts can compromise the integrity of the placenta and infection of fetus, resulting in abortion or birth of weak offspring (8,51).

In this study serological test (I-ELISA) is used for confirmatory diagnosis whereas most other studies, especially those with high seroprevalence used, complement fixation test (CFT) confirmatory diagnosis used. The difference in seroprevalence might also be associated with the types of sensitivity and specificity of diagnostic kits. According to comparative study conducted among RBPT, CFT and I-ELISA diagnostic tests showed that the highest specificity and sensitivity was observed in I-ELISA (52). In these comparative studies sensitivity of I-ELISA was 100%, which was higher than RBPT and CFT but similar specificity with CFT (100%). The skill of the laboratory technician and unexpected inflation of the result might be the reason for the difference. The low seroprevalence of the current study despite more sensitivity of I-ELISA in the area dictates brucellosis in small ruminants is very low. Problems associated with abortion and other reproductive problems might be associated with other overlooked etiological agents. Further investigation is important to rule out the major

reproductive problems in the area. As it was reported in other districts of Borena zone reproductive problems, especially in abortion cases could be due to chlamydiosis, coxiellosis in addition to brucellosis (53). Community awareness towards brucellosis infection and its zoonotic importance in this study is very low. Their practice knowledge on managing aborted fetuses and fetal membranes is very risky, so they either throw aborted materials into the open field or give it to a nearby dog. Such conditions significantly contribute to the dissemination of agents in the area.

Conclusion

A seroprevalence study conducted at Wachile district showed low seroprevalence. However, the finding is associated with the introduction of new animals to the flock, and seroprevalence is also associated with reproductive problems like abortion at the last gestation period, and retained placenta. According to the information obtained from the owners every small ruminants at their first kidding experienced abortion. This may draw attention to further study. Community educational status is very low which accompanied by low awareness about disease zoonotic importance and poor management of aborted waste is very pronounced. In line with this, the following recommendation is forwarded:

1. The community needs to be trained to prevent the entry of new animals into their flock as this can contaminate the healthy flock
2. Communities also need to be trained about zoonosis and ways of protecting mechanisms for their own and their families
3. The community has to be trained as to how to dispose of wastes associated with dead animals and aborted materials.

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Laboratory is acknowledged for being collaborative in conducting the serological test.

Ethical approval

The study was approved by the research ethics review committee (RERC) of the College of Natural and Computational Science under the reference number CNCS-REC031/23 on date 01 November 2023. To obtain consent from the animal owner and keep the welfare of animals while animal handling and blood collection.

Conflict of interest

Both authors declare there is no conflict of interest.

References

1. Central Statistics Agency (CSA): Agricultural sample survey. Livestock and livestock characteristic report (Private Peasant Holdings. 2018. Addis Ababa, Ethiopia
2. FARM AFRICA. Goat types of Ethiopia and Eritrea: physical description and management systems. International Livestock Research Institute. 1996. Addis Ababa, Ethiopia.
3. CARE-Ethiopia: Value Chain Analysis of Milk and Milk Products in Borana Pastoralist Area. Regional Resilience Enhancement against Drought Project. 2009. pp. 12-25. Yonad Business Promotion and Consultancy P LC, Addis Ababa, Ethiopia.
4. Edea Z, Bikila N, Debela G. Morphometric and Physical Characterization of Borana Indigenous Goats in Southern Oromia, Ethiopia. *Univers J Agric Res.* 2019;7:25-31. <http://doi.org/10.13189/ujar.2019.070104>
5. Radostits OM, Gay CC, Blood DC, Hinchliff KW, Constable PD. *Veterinary medicine: A text book of the disease of cattle, sheep, pigs, goat and horses*, 10th ed. London, Saunders, 2007. pp. 963-1110.
6. World Health Organization (WHO): *Brucellosis in humans and animals*. Geneva, Switzerland: WHO Press: 2006: 1-51.
7. World Organization of Animal Health (WOAH): *Terrestrial manual: Reference Laboratories for brucellosis (Brucella*

- abortus*, *B. melitensis* and *B. suis*). 2022. pp. 1-47. <https://www.woah.org/en/what-we-offer/expertise-network/reference-laboratories/#ui-id-3>.
8. OIE (Office International des Epizooties): Manual of standards for diagnostic tests and vaccines. Paris: Office international des epizooties; 2009.
 9. OIE (Office International des Epizooties): Bovine brucellosis. In: Manual of Diagnostic Tests and Vaccines for terrestrial animals (mammals, birds and bees). 6th. ed. Office International des Epizootics, OIE, Paris, France. 2008. 2: 624-659.
 10. Constable PD, Hinchcliff KW, Done SH, Grünberg W. Veterinary Medicine: A Textbook of the Diseases of Cattle, Horses, Sheep, Pigs, and Goats. 11th ed. vol. 1. Elsevier, 3251 River port Lane, St. Louis, Missouri 63043: 2017. pp.1781–1791.
 11. Seifert SH. Tropical Animal Health. 2nd ed. Dordrecht, the Netherlands: Kluwer Academic. 1996; 356-367.
 12. Bamaiyi PH, Hassan L, Khairani-Bejo S, Zainal AM. Updates on brucellosis in Malaysia and Southeast Asia. Malaysian J Vet Res. 2014;5(1):71-82.
 13. Gul ST, Khan A. Epidemiology and epizootiology of brucellosis. Pakistan Vet J, 2007. 27(3): 145-151.
 14. Anna Rovid. Brucellosis: *Brucella melitensis*. [Internet]. Available from: <http://www.cfsph.iastate.edu/>. [Accessed 5th December 2022].
 15. Domenech J, Lefevre PC. Serological survey on contagious bovine pleuropneumonia and bovine brucellosis in Ethiopia. Rev Elev Med Vet Pays Trop. 1974. 27 (1): 397–402.
 16. Megersa B, Biffa D, Abunna F, Regassa A, Godfroid J, Skjerve E. Seroprevalence of brucellosis and its contribution to abortion in cattle, camel, and goat kept under pastoral management in Borana, Ethiopia. Trop Anim Health Prod. 2011;43(3):651-6. <https://doi.org/10.1007/s11250-010-9748-2>
 17. 1. Yilma M, Mamo G, Mammo B. Review on Brucellosis Sero-prevalence and Ecology in Livestock and Human Population of Ethiopia. Achiev Life Sci. 2016;10(1):80-6. <http://doi.org/10.1016/j.als.2016.05.008>
 18. Alemayehu G, Mamo G, Alemu B, Desta H, Tadesse B, Benti T, et al. Causes and Flock Level Risk Factors of Sheep and Goat Abortion in Three Agroecology Zones in Ethiopia. Front Vet Sci. 2021;8:615310. <https://doi.org/10.3389/fvets.2021.615310>.
 19. Aloto D, Abebe R, Megersa B. Seroepidemiology of brucellosis in small ruminants, and herder's knowledge and risky practices in southern Ethiopia. Small Rumin Res. 2022;216:106785. <http://doi.org/10.1016/j.smallrumres.2022.106785>
 20. Geletu US, Usmael MA, Mummed YY. Seroprevalence and Risk Factors of Small Ruminant Brucellosis in West Hararghe Zone of Oromia Regional State, Eastern Ethiopia. Vet Med Int. 2021;2021:6671554. <https://doi.org/10.1155/2021/6671554>.
 21. Teshome D, Sori T, Banti T, Kinfe G, Wieland B, Alemayehu G. Prevalence and risk factors of *Brucella* spp. in goats in Borana pastoral area, Southern Oromia, Ethiopia. Small Rumin Res. 2022;206:106594. <https://doi.org/10.1016/j.smallrumres.2021.106594>
 22. Megersa B, Biffa D, Abunna F, Regassa A, Godfroid J, Skjerve E. Seroepidemiological study of livestock brucellosis in a pastoral region. Epidemiol Infect. 2012;140(5):887-96. <https://doi.org/10.1017/s0950268811001178>
 23. Gumi B, Firdessa R, Yamuah L, Sori T, Tolosa T, Aseffa A, Zinsstag J, Schelling E. Seroprevalence of Brucellosis and Q-Fever in Southeast Ethiopian Pastoral Livestock. J Vet Sci Med Diagn. 2013;2(1):10.4172/2325-9590.1000109. <https://doi.org/10.4172/2325-9590.1000109>
 24. Mihertu Y, Fentahun T. Socio-economic profile of arid and semi-arid agropastoral region of Borana rangeland Southern, Ethiopia. MOJ Ecol Environ Sci. 2020;

- 5:113-22.<http://doi.org/10.15406/mojes.2020.05.00183>
25. CSA 2022. Central statistics Authority of federal government of Ethiopia. Sampling survey
26. Thrusfield M. Veterinary epidemiology. 4th ed. New York: John Wiley & Sons. 2018;270-290.
27. Zewdie W. Sero-Prevalence Of Brucellosis In Goats And Community Awareness In Liban District Of Guji Zone, Oromia Regional State, Southern Ethiopia. *Juniper Online J Public Health*. 2017;2:(3). <http://doi.org/10.19080/JOJPH.2017.02.555587>
28. Margatho G, Rodríguez-Estévez V, Quintas H, Simões J. The Effects of Reproductive Disorders, Parity, and Litter Size on Milk Yield of Serrana Goats. *Animals (Basel)*. 2019;9(11):968. <http://doi.org/10.3390/ani9110968>
29. Villaquiran M., Gipson T, Merkel RC, Goetsch A, Sahlu T. Body condition scores in goats in Meat Goat Production Handbook, Langston University, Langston, OK. 2008.
30. Kenyon P, Maloney S, Blache D. Review of sheep body condition score in relation to production characteristics. *New Zealand J Agric Res*. 2014;57. <http://doi.org/10.1080/00288233.2013.857698>
31. Solomon A, Kassahun A. Estimation of weight and age of sheep and goats. Ethiopia sheep and goat productivity improvement program (ESGPIP), Technical Bulletin. 2009;23.
32. Bokko PB. Pregnancy wastage in sheep and goats in the Sahel region of Nigeria. *Nig Vet J*. 2011;32(2):120-126.
33. Vatta AF, Abbot MA, Villiers JD, Gumede SA, Harrison LJ, Krecek RC et al. Goat keepers' animal health care manual. Agricultural Research Council. Onderstepoort Veterinary Institute with KwaZulu-Natal Department of Agriculture and Environment, South Africa. 2006;19:60.
34. McGiven J, Taylor A, Duncombe L, Sayers R, Albert D, Banai M, et al. The first International Standard anti-*Brucella melitensis* Serum. *Rev Sci Tech*. 2011;30(3):809-19.
35. Ashenafi F, Teshale S, Ejeta G, Fikru R, Laikemariam Y. Distribution of brucellosis among small ruminants in the pastoral region of Afar, eastern Ethiopia. *Rev Sci Tech*. 2007;26(3):731-9. <https://doi.org/10.20506/rst.26.3.1781>
36. Saavedra M, Fernandes C, Queiroga C. Laboratory Diagnosis of Brucellosis. 2019;151-80.
37. Khan MZ, Zahoor M. An Overview of Brucellosis in Cattle and Humans, and its Serological and Molecular Diagnosis in Control Strategies. *Trop Med Infect Dis*. 2018;3(2):65. <https://doi.org/10.3390/tropicalmed3020065>
38. Edao BM, Ameni G, Assefa Z, Berg S, Whatmore AM, Wood JLN. Brucellosis in ruminants and pastoralists in Borena, Southern Ethiopia. *PLoS Negl Trop Dis*. 2020;14(7):e0008461. <https://doi.org/10.1371/journal.pntd.0008461>
39. Wubishet Z, Sadik K, Abdala B, Mokonin B, Getachew T, Getachew K. Small ruminant brucellosis and awareness of pastoralists community about zoonotic importance of the disease in Yabello districts of Borena Zone Oromia Regional State, Southern Ethiopia. *Current Trends in Biomedical Engineering & Biosciences*. 2018;12(1):5-10.
40. Dabasa G, Tefera M, Addis M. Small Ruminant Brucellosis: Serological Survey in Yabello District, Ethiopia. *Asian J Animal Sci*. 2013;7:14-21.
41. Sintayehu G, Melesse B, Abayneh D, Sintayehu A, Melaku S, Alehegne W, et al. Epidemiological survey of brucellosis in sheep and goats in selected pastoral and agro-pastoral lowlands of Ethiopia. *Rev Sci Tech*. 2015;34(3):881-93. <https://doi.org/10.20506/rst.34.3.2403>
42. Mengistu M. Seroepidemiology of brucellosis in small ruminants in Southern Ethiopia. Abstract of Thesis of the 2006/07 Graduates of Master s programs. 2007;22.
43. Hussen AM, Alemu F, Hasan Hussen A, Mohamed AH, Gebremeskel HF. Herd and

- animal level seroprevalence and associated risk factors of small ruminant brucellosis in the Korahey zone, Somali regional state, eastern Ethiopia. *Front Vet Sci.* 2023;10:1236494. <https://doi.org/10.3389/fvets.2023.1236494>
44. Teklue T, Tolosa T, Tuli G, Beyene B, Hailu B. Sero-prevalence and risk factors study of brucellosis in small ruminants in Southern Zone of Tigray Region, Northern Ethiopia. *Trop Anim Health Prod.* 2013;45(8):1809-15. <https://doi.org/10.1007/s11250-013-0439-7>
45. Teshale S, Muhie Y, Dagne A, Kidanemariam A. Seroprevalence of small ruminant brucellosis in selected districts of Afar and Somali pastoral areas of Eastern Ethiopia: the impact of husbandry practice. *Rev Med Vet.* 2006;157(11):557.
46. Asmare K, Megersa B, Denbarga Y, Abebe G, Taye A, Bekele J, et al. A study on seroprevalence of caprine brucellosis under three livestock production systems in southern and central Ethiopia. *Trop Anim Health Prod.* 2013;45(2):555-60. <https://doi.org/10.1007/s11250-012-0258-2>
47. Aworh MK, Okolocha EC, Awosanya EJ, Fasina FO. Sero-prevalence and intrinsic factors associated with *Brucella* infection in food animals slaughtered at abattoirs in Abuja, Nigeria. *BMC Res Notes.* 2017;10(1):499. <https://doi.org/10.1186/s13104-017-2827-y>
48. Hadush A, Abdella A, Gari F. Major prepartum and postpartum reproductive problems of dairy cattle in Central Ethiopia. *J Vet Med Anim Health.* 2013;5:118-23. <http://doi.org/10.5897/JVMAH2013.0205>
49. Dawite T, Ahmed S. Reproductive health problem of ruminants under different management system in kombolcha, Northeast Ethiopia. *Adv Biol Res.* 2013;7(3): 104- 108.
50. González-Espinoza, G.; Arce-Gorvel, V.; Mémet, S.; Gorvel, J.-P. *Brucella*: Reservoirs and Niches in Animals and Humans. *Pathogens* 2021, 10, 186. <https://doi.org/10.3390/pathogens10020186>
51. Saxena HM, Raj S. A novel immunotherapy of Brucellosis in cows monitored non invasively through a specific biomarker. *PLoS Negl Trop Dis.* 2018;12(4):e0006393. <https://doi.org/10.1371/journal.pntd.0006393>
52. Legesse A, Mekuriaw A, Gelaye E, Abayneh T, Getachew B, Weldemedhin W, et al. Comparative evaluation of RBPT, I-ELISA, and CFT for the diagnosis of brucellosis and PCR detection of *Brucella* species from Ethiopian sheep, goats, and cattle sera. *BMC Microbiol.* 2023;23(1):216. <https://doi.org/10.1186/s12866-023-02962-2>
53. Tesfaye A, Sahele M, Sori T, Guyassa C, Garoma A. Seroprevalence and associated risk factors for chlamydiosis, coxiellosis and brucellosis in sheep and goats in Borana pastoral area, southern Ethiopia. *BMC Vet Res.* 2020;16(1):145. <https://doi.org/10.1186/s12917-020-02360-0>