

## Sero-epidemiological Investigations of Camel Brucellosis and Community Perception in Selected Districts of Borana Zone, Southern Oromia, Ethiopia

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### Abstract

Understanding the epidemiology and natural history of camel brucellosis is crucial for control. A cross-sectional study was conducted in two selected districts of Borana Pastoral setting, Southern Ethiopia from November 2020 to April 2021 to estimate sero-prevalence and assess associated risk factors of camel brucellosis. A total of 315 blood samples were collected from camels older than 6 months in Arero and Elwoye districts. The collected serum samples were screened using Rose Bengal plate test and positive samples were confirmed using indirect enzyme-linked immunosorbent assay. The association between potential risk factors and sero-prevalence was computed using multivariable logistic regression and chi-square tests. Out of the total of 315 serum samples screened by Rose Bengal plate test 29 (9.21 %; 95 % CI: 6.25 – 12.95) were positive for brucellosis, of which 9 (2.86 %; 95 % CI: 13.15 – 53.54) were confirmed to be positive using indirect enzyme-linked immunosorbent assay. The statistical analysis showed that female camels which had history of abortion were more likely to be seropositive than those which did not have abortion history ( $\chi^2=5.51$ ;  $p=0.014$  and  $OR=6.2$ ; 95 % CI=1.08 -35.86). Camels tested from large herd size were more at risk of acquiring brucellosis than those from small herd size ( $\chi^2=9.02$ ;  $p=0.0027$  and  $OR=17.04$ ; 95% CI=1.77-164.04). The prevalence was higher (3.17 %; 95 % CI: 0.87 – 7.93) in Elwoye district than in Arero (2.65 %; 95 % CI: 0.86 – 6.07) although the difference was not statistically significant. The results of questionnaires revealed that 33 (73.33 %) of the respondents knew the brucellosis. Most of the animal herders had greater knowledge about the brucellosis than other participants of the study, which was statistically significant ( $P<0.05$ ). The major signs of brucellosis recognized by the pastoralists include abortion, retain placenta and stillbirth with 100%, 81.82% and 66.67%, respectively. The majority of the pastoralists interviewed (27; 81.82 %) were not aware about brucellosis being transmitted from domestic animals to humans. Although the prevalence of brucellosis observed in this in camels is low, the lack of control and prevention programs could make it a public health threat for the pastoral community.

**Key Words:** borana; brucellosis; camel; community perception; ethiopia; seroprevalence

### 1. Introduction:

Camels (Dromedaries) are important livestock species adapted to hot and arid environments prominently due to its unique anatomical, physiological and behavioral characteristics. It highly contributing to food security and social stability in the pastoral areas of Africa and The Middle East. Camels not only serve the community by providing food and darft power but also they are used to fetch water and other resources used for other livestock species during harsh conditions. The optimal utilization and the development of camel production is, however, hampered by different technical and non-technical constraints including infectious diseases [1]. Brucellosis is one of infectious Camel disease caused by *Brucella abortu* (B. abortus), *Brucella melitensis* (B. melitensis), *Brucella ovis* (B. ovis) and *Brucella suis* (B. suis) with considerable public health and economic importance. Geographical distribution of brucellosis occurs more frequently in countries with poorly standardized animal and public health services [2]. Camel brucellosis is endemic in countries of the Mediterranean basin, Middle East, Central Asia, horn of African countries such as Ethiopia, Eritrea, Somalia and Sudan [3] Where extensive traditional production with minimal veterinary services. In these



areas brucellosis has been reported in many domestic animal species including human beings [4].

The occurrence of brucellosis can be in any season of a year but the epidemic peak is mostly associated with delivery and abortion in animals [2]. Poor management and large herd size contribute to high prevalence of brucellosis. Increase in herd size increase the chances of contact between animals leading to infection prevalence in Borana zone.

particularly during calving or abortion [5]. This is often the practice adopted in pastoral areas where large number of animals of various age and species are reared together. Mixing of camels with other domestic animals during the time of migration, at watering time, in communal rangeland or at night enclosure can play role in the transmission of the disease from infected species to camels [6]. Transmission of brucellosis in animals occurs mainly through ingestion of food or water contaminated by

infected uterine discharges, aborted fetuses or fetal membranes and even through licking the genital of diseased animals. In addition, infected males can also spread the infection among females through natural mating and artificial insemination [7]. The most

common clinical manifestation of brucellosis in Camel is Abortion in pregnant Camels infected with *Brucella* organism's and non-Pregnant developed only mild, transient clinical symptoms including reduced appetite, slight lameness and bilateral Rock of Gibraltar fever and Bang's disease. In 1884, Dr. Bruce lacrimation [8], stillbirth or a weak, non-viable calf, retain was able to differentiate between brucellosis (Malta fever) and placenta, placentitis, uterine infections, fetal mummification and death, delayed maturity and infertility. Other conditions caused by the disease in male camel were Orchitis, epididymitis, arthritis and hygroma have also been associated with brucellosis [4].

Bacterial isolation is the gold standard in diagnosis of brucellosis, long cultivation periods and great care during handling any material containing *Brucella* organisms [9]. A serological test is closely related to Bruces's organism. In the year 1938, it was another test which frequently used to diagnose camel brucellosis, possible to differentiate among the caprine, bovine and swine which include RBPT (Rose Bengal Plate Test), CFT (Complement fixation test), ELISA (enzyme-linked immunosorbent assay), FPA suis, respectively [7]. Camel brucellosis has not received proper (Fluorescence Polarization Assay) and SAT (Serum agglutination test) [10]. Different serological tests combination can increase in camels as early as in 1931 by Solonitsiun in Russia then the diagnostic efficacy of tests [3]. Generally brucellosis cause disease has been reported from all camel-keeping countries. Camel significant loss of productivity through low herd fertility as a result of abortions, sterility, late first calving age, long calving interval world such as middle East and the Arabian Gulf, parts of Africa, time and comparatively low milk production [9]. The costs associated with medical care of *Brucella* infected humans and the duration of time the infected people are out of work account for financial losses [11]. The disease can also have an impact on export and import of animals constraining livestock trade and is an impediment to free animal movement [12]. In Ethiopia, brucellosis is endemic and highly prevalent in cattle, camels and small ruminants in pastoral and agro-pastoral areas [5]. Camel *Brucella* consists of at least six species, designated on the basis of brucellosis has been reported from pastoral areas, with prevalence ranging between 0.73 to 11.9% when RBPT was used for *B. melitensis* (goats, sheep and camel), *B. abortus* (cattle and screening and 0.53 to 9.6% using CFT [13]. The differences in camel prevalence is hypothesized to be associated with different environmental and management conditions [14].

and the ability of the camel to survive in harsh areas of the world, its endurance in prolonged drought, and above all its high potential to convert the scanty resources of the desert into milk and meat makes them more important to the pastoralists. Camels are versatile animal species in ensuring food security and fulfilling the

liveliness of pastoral households [15]. Its production would only be effective in understood and improved factor affect productivity and health burden. Since camels are becoming important livestock species in the pastoral areas where millions of people inhabit, understanding epidemiology and natural history of brucellosis is crucial. Therefore, this study was conducted to assess the community knowledge about camel brucellosis and estimate its prevalence in Borana zone.

#### The specific objective is:

- To assess the knowledge and perception of the community on brucellosis
- To identify the associated risk factors of camel brucellosis in study area

## 2. Literature Review:

### 2.1. *Brucella* Organisms:

#### 2.1.1. Historical prospective of brucella organisms:

*Brucella* is an organism's that has very old history of detection in carbonized cheese from the Roman era [7]. Several synonyms of *Brucella* have been known like Malta fever, undulant fever, including reduced appetite, slight lameness and bilateral Rock of Gibraltar fever and Bang's disease. In 1884, Dr. Bruce was able to differentiate between brucellosis (Malta fever) and typhoid outbreaks affected in Malta. Three years later, he isolated the causative agent of Malta fever and named the bacterium *Micrococcus melitensis* [2]. In 1897, Dr. Bang studied the disease in Denmark and could isolate *B. abortus* strains from aborted cattle. He noticed that the pathogen can also infect sheep, goat and

horses; the disease became known as Bang's disease. Later in 1918, Evans detect connection between animal and human cases which relevant under epidemiological point of view. It requires long cultivation periods and great care during handling any material containing *Brucella* organisms [9]. A serological test is closely related to Bruces's organism. In the year 1938, it was another test which frequently used to diagnose camel brucellosis, possible to differentiate among the caprine, bovine and swine which include RBPT (Rose Bengal Plate Test), CFT (Complement fixation test), ELISA (enzyme-linked immunosorbent assay), FPA suis, respectively [7]. Camel brucellosis has not received proper (Fluorescence Polarization Assay) and SAT (Serum agglutination test) [10]. Different serological tests combination can increase in camels as early as in 1931 by Solonitsiun in Russia then the diagnostic efficacy of tests [3]. Generally brucellosis cause disease has been reported from all camel-keeping countries. Camel significant loss of productivity through low herd fertility as a result of abortions, sterility, late first calving age, long calving interval world such as middle East and the Arabian Gulf, parts of Africa, time and comparatively low milk production [9]. The costs associated with medical care of *Brucella* infected humans and the duration of time the infected people are out of work account for financial losses [11]. The disease can also have an impact on export and import of animals constraining livestock trade and is an impediment to free animal movement [12]. In Ethiopia, brucellosis is endemic and highly prevalent in cattle, camels and small ruminants in pastoral and agro-pastoral areas [5]. Camel *Brucella* consists of at least six species, designated on the basis of brucellosis has been reported from pastoral areas, with prevalence ranging between 0.73 to 11.9% when RBPT was used for *B. melitensis* (goats, sheep and camel), *B. abortus* (cattle and screening and 0.53 to 9.6% using CFT [13]. The differences in camel prevalence is hypothesized to be associated with different environmental and management conditions [14].

#### 2.1.2. Etiology of brucellosis:

*Brucella* is a disease affecting a wide range of animal species including human beings, and caused by non-motile, aerobic, gram negative belonging to the cocobacilli genus of *Brucella*. The genus consists of at least six species, designated on the basis of brucellosis has been reported from pastoral areas, with prevalence ranging between 0.73 to 11.9% when RBPT was used for *B. melitensis* (goats, sheep and camel), *B. abortus* (cattle and screening and 0.53 to 9.6% using CFT [13]. The differences in camel prevalence is hypothesized to be associated with different environmental and management conditions [14].

and vaginal swabs of diseased of camels [16]. Even though camels



are not known to be the primary hosts of Brucella, they are susceptible to both B. abortus and B. melitensis consequently, the infection depends upon the infection rate in primary hosts being in contact with them [3].

Country	Brucella species	Specimen	References
Jordan	B. melitensis biotype 3	Aborted foetus, milk	[19]
Iran	B. melitensis biotype 1 B. abortus biotype 1	Lymph nodes Blood	[16]
Yemen	B melitensis	Vaginal swabs & blood	[20]
Libya	B.melitensis biotype 1	Milk, aborted foetus, vaginal swab	[21]
Egypt	B. melitensis biotype 3 B. abortus biotype 1 B. suis biotype 1	Milk	[22]

**Table 1:** Brucella species infecting camels reported from different countries of the world.

**2.2. Epidemiology of Camel Brucellosis:**

**2.2.1. Geographical distribution:**

Brucellosis is a worldwide bacterial disease affecting both animals and humans which subsequently causes serious human health hazards and economic loss. The geographical distribution of brucellosis shows that it is common in countries with poorly standardized animal and public health programed [2]. Though it has been eradicated from many developed countries like Australia, Canada, Israel, Japan, New Zealand and Europe), it remains an uncontrolled problem in regions of high endemicity such as Africa, Mediterranean, Middle East, and parts of Asia and Latin America [23]. Camel brucellosis is a wide area distributed disease were camel raring are being practiced. It is endemic in countries of the Mediterranean basin, Middle East, Central Asia, horn of African countries such as Ethiopia, Eritrea, Somalia and Sudan [3]. The prevalence of camel brucellosis reported from different countries is presented in Table 2.

Country	Prevalence %	Lab Test	Reference
Pakistan	21% 21% 13%	RBPT SAT c-ELISA	[24]
Libya	5.7%	CFT	[25]
Oman	1.5%	c-ELISA	[14]
Kenya	2% 10.5%	RBPT SAT	[26]
Egypt	4.17% 3.73%	m-RBPT c-ELISA	[22]
Eritrea	3.1%	CFT	[27]
Iran	13 %	PCR	[16]
Iraq	3.03%	RBPT/ 2ME	[28]
Sudan	5.8% 5%	RBPT c-ELISA	[9]
India	8.9% 4.9%	RBPT ELISA	[29]
Nigeria	11.2% 10.5%	RBPT SAT	[30]
Somalia	1.7% 3.9%	RBPT c-ELISA	[31]
Yemen	5.1%	MRT	[20]

**Table 2:** Summary of occurrence of camel brucellosis in the world

**2.2.2. Risk factors of brucellosis:**

Brucellosis can affect almost all animal species including human beings, and cross transmission can occur between cattle, sheep, goats, camels and other species. It causes significant reproductive losses in sexually mature animals [4]. Susceptibility to infection depends on pregnancy status, age, sex, and breed of the animals. Sexually matured animals are more prone to Brucella infection than sexually immature animals of either sex. On the other hand, it is also true that younger animals tend to be more resistant to infection and frequently clear an established infection, although latent infections can occur [11]. This may be due to the fact that sex hormones and erythritol, which stimulate the growth and multiplication of Brucella organisms, tend to increase in concentration with age and sexual maturity [32]. Occurrence of brucellosis is not seasonal but the epidemic peak occurs season is associated with delivery and abortion in animals [2]. After reaching sexual maturity, the state of pregnancy has a greater influence on the degree of susceptibility. In pregnant camels, the bacteria localize in the placenta and are most abundant in abortion material (up to 10<sup>13</sup> bacteria) including the fetal stomach, vaginal discharge and colostrum [33]. parturition in camels is occurred in a laying or standing position without extra help, they may deliver or abort on the pasture and the aborted material may spread over a wide area of the pasture by stray dogs and foxes [3].

Poor management and large herd size contribute to high prevalence rate of brucellosis. Increases in herd size increase the chances of contact between animals which leading to infection particularly during calving or abortion [5]. Placentophagy with camels as a noted exception, which may contribute to the transmit of Brucella organism [18]. Camel herd kept in close contact with other domestic animals during the time of migration, at watering time or at night enclosure can also play the transmission of the disease from infected animals to healthy ones [6]. Close contact between infected and susceptible camels, and sharing the same watering points and pastures with other livestock promotes the spread of diseases [8]. Survival of the organisms in the environment is enhanced by cool temperatures and humidity however it can also survive in a hot desert environment [34]. Under appropriate conditions, Brucella organisms can survive in the environment for prolonged periods. Their ability to withstand inactivation under natural conditions is relatively high compared with most other groups of non sporing pathogenic bacteria. B. abortus survival outside the host is largely dependent on environmental conditions. The pathogen may survive in aborted fetus in the shade for up to eight months, for two to three months in wet soil, one to two months in dry soil, three to four months in faeces, and eight months in liquid manure [35].

Brucella has ability to adapt to the environmental conditions in intracellular replication including low levels of nutrients and oxygen, acidic pH and reactive oxygen intermediates. Inside the cells, Brucella has the ability to interfere with intracellular trafficking, preventing fusion of the Brucella containing microphages with lysosome markers, and directing the vacuole toward a compartment that has rough endoplasmic reticulum, which is highly permissive to intracellular replication of Brucella [3]. These endoplasmic reticulum-associated compartments are the niche for intracellular replication of Brucella in macrophages, epithelial cell lines and placental trophoblasts. Once inside this



compartment, the bacteria can establish chronic infection [36].

### 2.2.3. Transmission and sources of infection:

Brucellosis is transmitted horizontally under normal conditions. Domestic and wild animals can contract brucellosis through direct lameness and bilateral lacrimation [8], retain placenta, placentitis, contact with infected animals and their excreta. The primary uterine infections, fetal mummification and death, delayed shedding routes of organisms is uterine fluids and placenta maturity and infertility. Other conditions caused by the disease in expelled from infected animals [6]. Natural infection in animals male camel were Orchitis, epididymitis, arthritis and hygroma occurs mainly through ingestion of feed or water contaminated by have also been associated with brucellosis [4]. Human brucellosis uterine discharges, aborted fetuses or fetal membranes and even is a disease that may have variable clinical sign after exposure to through licking the genitalia of diseased animals. In addition, the bacteria; clinical manifestations may appear within five to sixty infected males can also spread the infection among females days. Infected patients with acute disease consisting of general through natural mating and artificial insemination. Brucella can symptoms, such as fever, malaise, sweats and lymphadenopathy pass through intact or injured skin and mucous membranes [7]. and hepato splenomegaly [8]. Chronic brucellosis is more severe Brucellosis is transmissible from animals to humans through form of the disease that can be associated with osteoarticular signs contaminated milk, raw milk products, meat or direct contact with including spondylitis, arthritis and osteomyelitis, or genitourinary infected animal blood, placenta, fetuses or uterine secretions, infection, such as orchitis, epididymitis, glomerulonephritis and handling infected animal fetus and placenta. Person to person kidney abscesses. Life-threatening complications comprise, in transmission is rare, but it being transmitted by close personal or descending order of frequency, neuro brucellosis, liver abscesses, sexual contact, blood donation, tissue transplantation and Bone and endocarditis [36]. marrow transfer [18].

### 2.3. Pathogenesis:

Brucella infection depends on natural resistance of the animal to delayed or missed because the clinical picture may mimic other the organisms, virulence of the Brucella specious and exposure infectious and non-infectious conditions. Thus, It is very difficult dose. Organisms enter animal hosts through skin abrasions, to make a diagnosis based on clinical signs despite abortions in the reproductive tracts, gastrointestinal tract, respiratory tract and third trimester being indicative of brucellosis; this is because other conjunctiva. In the alimentary tract the epithelium covering the infectious diseases such as leptospirosis, Rift valley fever and ileal Peyer's patches are sites of entry [38]. Brucella penetrates the Listeriosis can also cause abortion storms [37]. mucosal epithelium and transported as free bacteria or engulfed by phagocytic cells. After penetration and localized to regional lymph nodes it proliferates, disseminate haemogenously and localize in the reticulo endothelial [39]. Various mechanisms employed by The microorganism can be identified by microscopic examination Brucella organisms to survive inside the phagocytic cells is of stained smear from vaginal dis- charges, placenta, colostrum, inhibiting phagolysosome fusion, blocking bactericidal action of fetal stomach fluid or of the aborting cow's lochia, and the phagocytes and suppressing the myeloperoxidase H<sub>2</sub>O<sub>2</sub> halide abomasum of the aborted fetus using the modified Ziehl-Neelsen system [9]. They are taken up in phagosomes, re-main viable by (MZN) stain. Impression smears may be taken from freshly cut and suppressing phagosome-lysosome fusion, and inhibit apoptosis of blotted tissue surfaces, e.g. cotyle- dons, by firmly pressing the host cells. They multiply in vacuoles within the endoplasmic slide surface against the tissue. Allow to air dry and heat fix smears reticulum and spread to various organs, particularly into the cells [38]. Brucella is not a true acid- fast bacillus but show resistant to of the reticulo endothelial system, liver, urogenital tract, spleen and decolorization by week acids. They seem like short rods or skeletal muscle where they give rise to granulocytic inflammation coccobacilli, mostly arranged singly but occasionally in pairs or with or without necrosis or caseation [38].

Organisms spread through the hema-togenous route reaches the However, morphologically related micro-organisms, such as placenta and finally to the fetus. The preferential localization to the Chlamydophila abortus, Chlamydia psittaci and Coxiella burnetii reproductive tract of the pregnant animal is due to the presence of can mislead the diagnosis because of their superficial similarity. the allantoic fluid factors that would stimulate the growth of Accordingly, the isolation of B. melitensis on appropriate culture Brucella. Four carbon alcohols (Erythritol) is one of the factors media such as Farrell's selective media is recommended for an which elevated in the placenta and fetal fluid from end of second accurate diagnosis [41]. trimester of gestation. An initial localization within placentome adjacent to chorioallantoic membrane results in rupture of the cells The gold standard in the diagnosis of brucellosis is bacterial and ulceration of the membrane. The damage to placental tissue isolation (culture), which relevant under epidemiological point of together with fetal infection and fetal stress inducing maternal view. It requires long cultivation periods and great care during hormonal changes that cause abortion [38].

### 2.4. Clinical Signs of Brucellosis:

Clinical symptoms variation of brucellosis is typical consequence infections, particularly in research laboratories. All Brucella

of level of immunity, environmental influences, age, pregnancy status and virulence of the pathogen. Camel brucellosis is characterized by Abortion in pregnant Camels infected with Brucella organism's and non-Pregnant developed only mild,

### 2.5. Diagnosis of Brucellosis:

The diagnosis of brucellosis can be challenging and is frequently delayed or missed because the clinical picture may mimic other the organisms, virulence of the Brucella specious and exposure infectious and non-infectious conditions. Thus, It is very difficult dose. Organisms enter animal hosts through skin abrasions, to make a diagnosis based on clinical signs despite abortions in the reproductive tracts, gastrointestinal tract, respiratory tract and third trimester being indicative of brucellosis; this is because other conjunctiva. In the alimentary tract the epithelium covering the infectious diseases such as leptospirosis, Rift valley fever and ileal Peyer's patches are sites of entry [38]. Brucella penetrates the Listeriosis can also cause abortion storms [37].

#### 2.5.1. Bacteriological diagnosis:

The microorganism can be identified by microscopic examination of stained smear from vaginal dis- charges, placenta, colostrum, fetal stomach fluid or of the aborting cow's lochia, and the phagocytes and suppressing the myeloperoxidase H<sub>2</sub>O<sub>2</sub> halide abomasum of the aborted fetus using the modified Ziehl-Neelsen system [9]. They are taken up in phagosomes, re-main viable by (MZN) stain. Impression smears may be taken from freshly cut and suppressing phagosome-lysosome fusion, and inhibit apoptosis of blotted tissue surfaces, e.g. cotyle- dons, by firmly pressing the host cells. They multiply in vacuoles within the endoplasmic slide surface against the tissue. Allow to air dry and heat fix smears reticulum and spread to various organs, particularly into the cells [38]. Brucella is not a true acid- fast bacillus but show resistant to of the reticulo endothelial system, liver, urogenital tract, spleen and decolorization by week acids. They seem like short rods or skeletal muscle where they give rise to granulocytic inflammation coccobacilli, mostly arranged singly but occasionally in pairs or small groups. They appear as coccobacilli or short rods, usually arranged individually but sometimes in pairs or small groups [40].

However, morphologically related micro-organisms, such as Chlamydophila abortus, Chlamydia psittaci and Coxiella burnetii can mislead the diagnosis because of their superficial similarity. Accordingly, the isolation of B. melitensis on appropriate culture media such as Farrell's selective media is recommended for an accurate diagnosis [41].

The gold standard in the diagnosis of brucellosis is bacterial isolation (culture), which relevant under epidemiological point of view. It requires long cultivation periods and great care during handling any material containing Brucella organisms. Brucella Spp. is classified as a Biosafety level 3 organism, which manipulation should be performed in biosafety level-3 laboratories [9]. Brucellosis is one of the most common accidental laboratory

infections, particularly in research laboratories. All Brucella



strains are relatively slow growing and use of a selective medium, matrix in 96 well plates. i-ELISA has high sensitivity, but the e.g. Farrell's medium because of specimens from which isolations specificity can be rather low. Commercial kits using whole cell, S-best are heavily contaminated [38]. Specimens which used for LPS or the O-polysaccharide (OPS) as antigens have been Brucella isolation include milk (colostrum or milk within a week validated and results obtained from different assays are not always of calving) vaginal swabs; semen and aborted fetus are useful for comparable [44]. CFT allows the detection of anti-Brucella diagnosis of organisms at ante mortem. Samples collected at antibodies that are able to activate complement. Many authors necropsy include spleen, udder, pieces of uterus and testicular regarded the CFT as being the most sensitive and specific test for tissue, fetal stomach fluid, supra mammary lymph nodes (chronic brucellosis diagnosis because CFT antibodies remain in the serum and latent infections) and retropharyngeal (early infections) are for longer period of time than SAT antibodies. On the contrary, preferred, but iliac, pre scapular and parotid may be used. If some authors disclosed that this test is not highly sensitive but serological reactions are thought to be caused by S19 vaccine strain shows an excellent specificity. In the recent year CFT is then it is important to collect pre-scapular lymph nodes as well [9]. progressively being replace by ELISAs since it is difficult to be Demonstration of the bacteria is by staining with Gram-negative standardized. Nevertheless, CFT is a "prescribed test for trade" by stain or modified-Zeihl Neelsen staining florescent antibody test the OIE [3].

and polymerase chain reaction methods for Brucella species

identification[9]. B. Spp. colonies are elevated, transparent, FPA is simple and the rate of rotation of a molecule in solution is convex with intact borders, smooth, and a brilliant surface. The inversely proportional to its size. A small molecule will rotate colonies have a honey color under transmitted light. Optimal rapidly while larger molecules rotate more slowly. By attaching a temperature for culture is 37 °C whereas optimal pH ranges from fluorescing molecule to an antigen molecule, the rate of rotation 6.6 to 7.4. Some Brucella spp. requires CO<sub>2</sub> for growth. Typical can be measured using polarized light. The result is a measurement colonies appears 2 to 30 days of incubation, but a culture can only of the time it takes the molecule to rotate through a given angle. In be considered negative when there are no colonies appears 2 to 3 the case of brucellosis serology, small molecular weight subunit of weeks of incubation [38].

### 2.5.2. Serological diagnosis:

Serological tests frequently used to diagnose camel brucellosis reduction is proportional to the amount of antibody present [45]. include RBPT, CFT, ELISA (competitive and indirect), FPA and The SAT is simple, cheap and lack of sensitivity and specificity SAT. Different serological tests combination can increase mean that it should only be used in the absence of alternative diagnostic efficacy, although none of the serological tests can techniques. It has been used extensively for brucellosis diagnosis. differentiate Brucella species. False-positive or unspecific A suspension of Brucella possessing active antigen will agglutinate reactions with various other bacterial species may occur [3]. All when exposed to homologous Brucella antibody. This tests have limitations concerning specificity and sensitivity, agglutination forms clumps of bacteria which become especially when testing individual animals [10]. RBPT is known macroscopically visible. SAT is used to detect brucellosis by as the buffered Brucella antigen tests which rely on the presence measuring agglutinating antibodies of the IgM, IgG 1, IgG 2, and of antibodies against antigen of Brucella in the serum. The IgA types. The SAT can be used to detect acute infections, as principle is based on the ability of IgM antibodies bind to antigen antibodies of the IgM type usually appear first after infection and is markedly reduced at a low pH [42]. It is very sensitive and are more reactive in the SAT than antibodies of the IgG 1 and IgG suitable test for screening herds for brucellosis, but false positive 2 types. However, because the SAT may yield both false negative results due to vaccination with B. abortus strain 19 vaccine or cross or false positive results it effectively detects brucellosis only on a reactions with other bacteria. RBPT may not be absolutely reliable herd basis [46].

among commonly used serological diagnostic tests for brucellosis.

RBPT detected antibody in the sera of fifty percent of the animals Milk ring test (MRT) is serum agglutinations test used to identify suspected for brucellosis [3].

the accurateness of antibodies against Brucella spp. in milk. It suggested as a screening test to check Brucellosis is bulk tank milk. Competitive ELISA (c-ELISA) is the most sensitive test for the MRT is done by cream or whole milk [41]. Hematoxylin Brucella diagnosis of brucellosis. Doubtful or positive samples with RBPT stained cells are added to milk and incubated for the reaction. MRT were further confirmed by c-ELISA [33]. c-ELISA using a detects the IgM and IgA immunoglobulin. False adverse reaction commercial DNA extraction kit according to the manufacturer's in abnormal milk is due to mastitis, milk from the late lactation due protocol. Gene amplification was performed in a thermal cyler. c- to the presence of colostrum. Low concentration of lacteal ELISA, using S-LPS or OPS as antigens, are used for brucellosis antibodies or lacking fat, clustering, and factors in milk may also serology. Different antiglobulin-enzyme conjugates, substrate/ cause a false-negative result. Despite all these problems, the milk chromogens and antigens are prepared from different smooth ring test is very successful, it is the method of choice in dairy herds, Brucella strains. The c-ELISA uses a monoclonal antibody specific and it is a low-cost screening test as compared to other [40].

for one of the epitopes of the Brucella spp. OPS antigens have higher specificity, but slightly lower sensitivity than i-ELISA. This assay is an excellent confirmatory assay for the diagnosis of

brucellosis in most mammalian species [43]. Indirect enzyme Molecular techniques are important tools for diagnosis and linked immunosorbent assay (i-ELISA) is most commonly used epidemiologic studies, providing relevant information for system depends on enzymes for detection and consists of smooth identification of species and biotypes of Brucella spp. allowing Lipopolysaccharide (S-LPS) preparation attached to a polystyrene differentiation between virulent and vaccine strains. Molecular

### 2.5.3. Molecular diagnosis:



detection of *Brucella* spp. can be done directly on clinical samples including virulence of the infecting strain, size of inoculum, age, without previous isolation of the organism. In addition, these sex, pregnancy, species, and immune status of the [48].

techniques can be used to complement results obtained from

phenotypic tests. Despite the high degree of DNA homology **2.6.1. Humoral immune response:**

within the genus *Brucella*, several molecular methods, including

PCR, PCR restriction fragment length polymorphism (RFLP) and humoral immune response plays an important role in immunity to Southern blot, have been developed that allow, to a certain extent, provide protection. Protective mechanisms of humoral immunity differentiation between *Brucella* species and some of their biovars against intracellular pathogens may rely on combination of factors [47].

PCR based techniques have been developed in recent years and are protection against primary infection is less explicit. Innate or in use as alternative diagnostic tests for brucellosis. They are based alternate immuno-protective mechanisms that precede on the detection of specific sequences of *Brucella* spp. DNA in development of humoral immunity are sufficient to control clinical samples. PCR techniques have lower diagnostic sensitivity primary infection and the synergistic and inhibitory contributions and higher specificity than culture methods hence best results are of specific antibodies need to be further explored [49]. obtained when the two are combined [37].

#### 2.5.4. Allergic skin test:

Allergic skin test (AST) is an allergic test that measures cellular immune response which has been used by some researchers, initial antigenic stimulus but is soon followed by IgG antibody. particularly on Bactrian camels in the former USSR. AST based IgG1 immunoglobulin is the most abundant in serum and exceeds on a delayed type hypersensitivity reaction with a maximum the concentration of IgG2. The magnitude and duration of the sensitivity at 72 hours post inoculation increase in the thickness of antibody response following immunization is directly related to the skin at the site of inoculation. The antigen does not induce animal's age at immunization and the number of organisms administered. immune system and not interfere in the diagnosis of the disease Following immunization with the standard dose of strain 19 during and decrease the of false-positive reactions. The skin test is highly calf hood, IgG antibody concentrations usually decline to specific and weak sensitivity. Thus, it is often suggested for use at diagnostically insignificant levels over 3 - 6 months [11]. the herd level as a positive test in unvaccinated animals [33 and 40].

#### 2.5.5. The 2-Mercaptoethanol test:

The Mercaptoethanol Test (2-MET) are two forms that use either chronic nature of many diseases caused by intracellular pathogens, 2-mercaptoethanol or dithiothreitol. Dithiothreitol has an effective adaptive response is necessary to control disease. recommended, because of the toxicity of 2-mercaptoethanol. The Several components of the immune system contribute to protection disulfide of IgM is being condensed to the manometric molecule against intracellular pathogens. Cell mediated immune response and unable to agglutination essentially calculate IgG unable to helps to remove the infection and creates memory component to agglutinate. However, IgG can also be decreased in the procedure, that specific antigen in the host, which is an essential property in providing false-negative results. Though in general, reduction of long lasting vaccination response [50] critical for protection IgM increases specificity. The test not suggested for the global against *Brucella* and other intracellular pathogens such as trade due to not eradication vaccinal antibodies. The 2-MET is, Chlamydia, Francisella, and Mycobacterium [49]. however, used prominently for national control and eradication programs [41]

#### 2.5.6. Laboratory animal inoculation:

Animal inoculation may be either through abraded skin or T-cell mediated protection is primarily associated with a Th1 T-subcutaneously in guinea-pigs or, preferably, through the digestive cell response and persistence (chronic brucellosis) with a Th2 tract or nasal (aerosol) intravenously, or intra peritoneal routes in response [49]. Macrophage-derived cytokines which are mice. The spleen of mice is cultured seven days after inoculation, interleukin 1 (IL-1), IL-12, and tumor necrosis factor alpha (TNF- while serum samples of guinea pigs are subjected to specific tests ) plays important role in control of early *Brucella* spp. infection by three and six weeks after inoculation. It is noteworthy however, IFN- pathway [50]. Immune response can control *Brucella* that in laboratory animal gastric acid can interfere with the infection by IFN- activates the bactericidal function on *Brucella* infectivity of *Brucella* [41].

### 2.6. Host Protective Immune Response:

Infection with *Brucella* usually results in the induction of both IgG2a and IgG3 engulf the pathogen to promote phagocytosis and humoral and cell-mediated immune responses. The magnitude and degradative endocytic compartments [50]. Cytokines are likely to duration of these responses can be affected by many factors exert maximum effect early in infection and balance enhancing

IgG1, IgG2, IgM, and IgA are the immunoglobulin isotypes present in animal serum. The first immunoglobulin produced after an initial heavy infection or strain 19 immunization is IgM. This

can usually be detected in the first or second week following the

#### 2.6.2. Cell-mediated immune response:

*Brucellae* are facultative intracellular bacteria that survive and replicate in both phagocytic and non-phagocytic cells. Due to the

Phagocytic cell process and presenting antigens to initiate T-cell responses which play a major role in acquired specific resistance to intracellular bacteria determines the resolution of infection [51].

Macrophages and T-cells play crucial roles in protection. Helper



immunity and exacerbating disease. The combined transfer of immune serum and cells has given better protection than that provided by serum or cells alone given prior to the challenge [51].

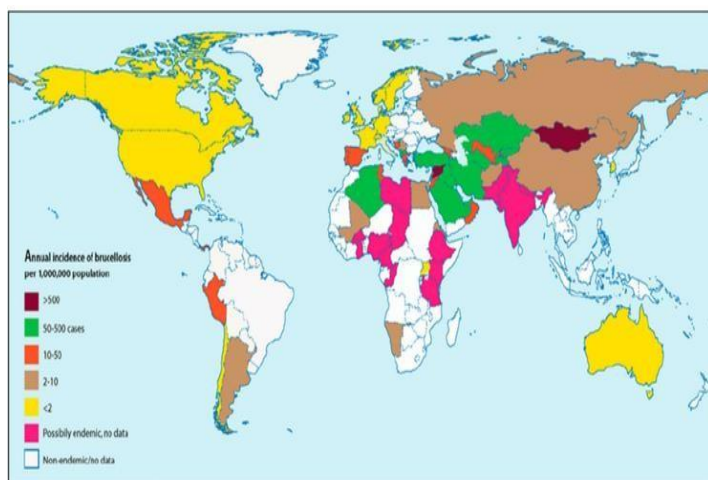
**2.7. Importance of Camel Brucellosis:**

**2.7.1. Public health importance:**

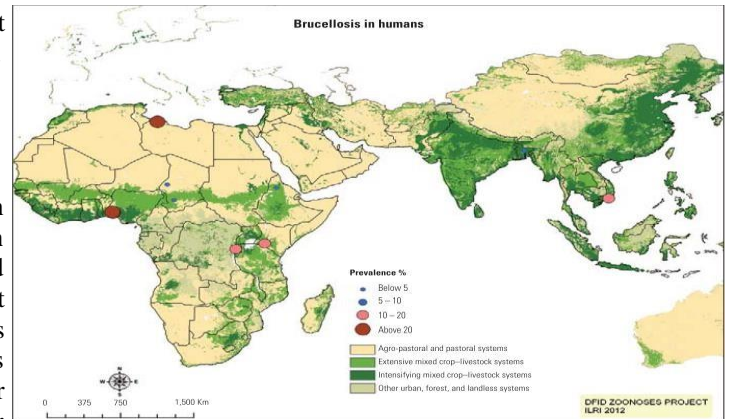
Brucellosis is an important zoonotic disease that has been shown to cause human ailments for over one and half centuries. It has been known to be caused by *B. melitensis*, *B. abortus*, *B. suis* and occasionally by *B. canis* [11]. It is the second most important zoonotic disease after rabies which is more severe in human beings than domestic animals. Brucellosis is transmissible from animals to humans through contaminated milk, raw milk products, meat or direct contact with infected animal blood, placenta, fetuses or uterine secretions, handling infected animal fetus and placenta. Person to person transmission is rare, but it being transmitted by close personal or sexual contact, blood donation, tissue

transplantation and Bone marrow transfer [18]. Human brucellosis is a disease that may have variable clinical sign after exposure to the bacteria; clinical manifestations may appear within five to sixty days. Infected patients with acute disease consisting of general symptoms, such as fever, malaise, sweats and lymphadenopathy and hepato splenomegaly [8]. Chronic brucellosis is more severe form of the disease that can be associated with osteoarticular signs including spondylitis, arthritis and osteomyelitis, or genitourinary infection, such as orchitis, epididymitis, glomerulonephritis and kidney abscesses. Life-threatening complications comprise, in descending order of frequency, neuro-brucellosis, liver abscesses,

The incidence and prevalence of brucellosis in humans has been reported from various countries of the world (figure 1 and figure 2). The incidence and prevalence vary partially depending on the living standards and habits of the community. For example residents of the Wajir County in Kenya drink camel urine since they believe that it eliminates all the illness in the body but this practice contributes to the transmission of camel brucellosis [3]. It is usually considered an occupational disease for those engaged in handling infected animals, such as veterinarians, laboratory staff, farmers, and abattoir workers [25].



**Figure 1:** Incidence of human brucellosis in world  
**Source:** [3]



**Figure 2:** Prevalence of human brucellosis in world  
**Source:**[52]

Pastoralists in Ethiopia consume raw milk, which contributes to the transmission of this disease among human and animals. more than 75% of the animal owners do not know about zoonotic Camel brucellosis [3]. The pastoral community in Ethiopia is traditionally, they consume raw animal products, sharing dwelling with their animals and poor management practices are highly prone to this disease. low awareness of the disease in general may result in high degree of transmission of the disease to human [53]. A cross-sectional study conducted by [12] in Mehoni District, Southeastern Tigray, from a total of 120 camel owners participated in the interview, about 91% (109) drank fresh raw milk regularly infected (11% of the total) (88.33% (106). The risk of Brucella contact to their animals (OR = 8.07, CI 95%; 0.476, 137.014) [12].

**2.7.2. Economic importance:**

Camels are primarily the domestic animals of pastoral communities that ensure food security. They produce milk, meat, hair and hides, and serve as a draught animal for agriculture and transport people and goods [11]. Generally brucellosis cause significant loss of productivity through low herd fertility as a result of abortions, sterility, late first calving age, long calving interval time and comparatively low milk production [9]. The costs associated with medical care of *Brucella* infected humans and the duration of time the infected people are out of work account for financial losses [11]. The disease can also have an impact on export and import of animals constraining livestock trade and is an impediment to free animal movement [12].

**2.8. Status of Camel Brucellosis in Ethiopia:**

Camel population in Ethiopia is around 1.16million, out of which, 434,291 inhabits in Afar region, 353,124 in Somali region and 239,357 in Oromia region [1]. Camel production could be a profitable venture for utilizing the vast arid and semi-arid areas of Ethiopia, where other animals survive with difficulty, especially due to the recurring drought conditions. Under such environmental conditions, camels thrive and form a source of milk and meat. But, complete exploration of camels for milk and meat production would only be possible when their reproductive performance is properly understood and improved [54]. In Ethiopia, brucellosis is endemic and the disease is highly prevalent in cattle, camels and



Origen	Prevalence	Test	Reference
Akaki Abattoir	6.5%	RBPT	[58]
	4.5%	CFT	
Afar	12.2%	RBPT	[54]
	4.1%	CFT	
Tigray	5.80%	RBPT	[12]
	3.37%	CFT	
Somali	4.9%	RBPT	[56]
	0.0%	CFT	
Dire Dawa	1.9%	RBPT	[59]
	1.6%	CFT	
Borana	12.5%	RBPT	[42]
	3%	i-ELISA	
Fentale	9.2%	RBPT	[60]
	9.1%	CFT	
Bale	0.6	CFT	[61]

**Table 3:** Prevalence of camel brucellosis in Ethiopia  
**2.8.1. Risk factors of camel brucellosis in Ethiopia:**

Previous investigations carried out showed that mixing of camels with other domestic animals during the time of migration, at watering time or at night enclosure is an important risk factor that contributes to the transmission and spread of the disease from infected animals to healthy ones [6]. The sero-prevalence of camel brucellosis has been shown to be higher in camels that have contact with cattle, sheep [54]. There are higher chances of brucellosis transmission from ruminants to dromedaries as they live in free range in promiscuity in the bush and at water points [42].

Such husbandry practices are common feature of some of the pastoral communities of Ethiopia. For example, there is free commingling of camels with ruminants in Borana pastoral areas. This might have contributed to the occurrence of camel brucellosis in the area. Studies also revealed that herds with larger size (>50 camels) had higher prevalence (36.84%) than medium (15.38%) and small sizes. Brucella seropositivity increased with large herd size while the chances of contact between animals' increases during calving or abortion. Thus, herd size and density of animal population together with poor management are directly related to high prevalence of brucellosis [5]. Investigation done by [58] on seroprevalence and risk factors of brucellosis in camels brought for slaughtering at Akaki abattoir, disclosed that age of

small ruminants in pastoral and agro-pastoral areas [5]. Brucellosis has been reported in camels from pastoral areas; where the prevalence was quite vary ranging between 1.9 to 12.5% for RBPT and 0.00 to 4.5% for CFT as shown in Table 3. This variation in prevalence of camel brucellosis can be attributed related to different factors such as difference in animal husbandry and management systems practiced by pastoral society [55]

Study conducted by [42] in Yabello and Gomole districts of Borana Zone, revealed seroprevalence of 12.5% using RBPT for screening from which 3% of them were confirmed to be positive by using Indirect Enzyme-Linked Immunosorbent Assay (i-

ELISA). A similar cross-sectional study conducted by [56] in Jigjiga and Gurusum districts of Fafan Zone, Somali Regional State showed seroprevalence of (4.9%) in camels when RBPT was used to screen the sera samples. Among those positives samples by RBPT, (0.4%) of them were confirmed positive by CFT. A cross-sectional study conducted by [54] in three selected districts of Afar region of Ethiopia also revealed similar seroprevalence of camel brucellosis. These authors sampled 245 camels from the two districts and their observation revealed that 4.1% of them were confirmed to be infected by *Brucella* spp. by CFT. A similar study conducted by [57] in Mehoni district, Southeastern Tigray in which seroprevalence of 5.80% and 3.37% was observed using RBPT and CFT, respectively. Previous study investigated by [57]. However, investigation done by [58] on seroprevalence and risk factors of brucellosis in camels brought for slaughtering at Akaki abattoir, serum samples from 201 apparently health camels were positive for brucellosis, of these, 9 (4.5%) were confirmed to be seropositive for brucellosis by CFT.

All these investigations showed that camels reared in all pastoral and few agro-pastoral areas of Ethiopia are infected with *Brucella*. Although the sample sizes considered and the geographical areas covered were limited, the previous results showed that brucellosis is well entrenched in camel population in the areas. This has important implication for public health particularly for those who are occupationally associated with camels.

## 2.9. Treatment:

*Brucella* organisms are Gram-negative coccobacilli which are sensitive to many broad-spectrum antibiotics [18], but the use of antibiotics is forbidden in many countries because of uncertainty about the infective status and antibiotic resistance. Treatment is unlikely to be cost-efficient or therapeutically effective because of the intracellular sequestration of the organisms, mainly in lymph nodes [33]. Treatment for human brucellosis includes administration of Tetracycline (five hundred gram every six hours orally) administered for at least six weeks, Doxycycline (a long acting tetracycline analogue) in dose of hundred gram every twelve hours orally with amino glycoside for the first two to three weeks of therapy. Other antibiotic used for treatment are Streptomycin, Gentamicin, Rifampicin, Fluoroquinolones, Trimethoprim or sulfamethoxazole in combination with another agent, such as doxycycline, rifampicin or streptomycin [18].

## 2.10. Prevention and Control:

The control and prevention of brucellosis depend on animal species involved, *Brucella* species, management practices and availability and efficacy of vaccines. The options to control the disease include immunization, testing and removal, and improving management practices and movement control[18]. Thus, control by herd immunization and vaccination of calves at four to eight months of age is helpful. Test and slaughter policy can be followed in counties where intensification is practiced [9]. From diagnostic base initial control measures including testing, quarantine and slaughter with vaccination implemented to reduce high prevalence [18]. In Endemic area, treatment can successfully eliminate





shedding of organisms from long term carriers, but it is believed to be economically unviable [52]. Effective vaccine against brucellosis in camels and other ruminants is live attenuated *B. abortus* S19 and *B. melitensis* Rev-1 proved [33]. Disadvantage of both vaccines are causing abortion, pathogenic to human beings and interference with serological tests. The non-smooth strains of *B. abortus* RB51 and *B. melitensis* M111 have recently been introduced into some countries. These vaccines are said to be safe and do not interfere with serological tests [9].

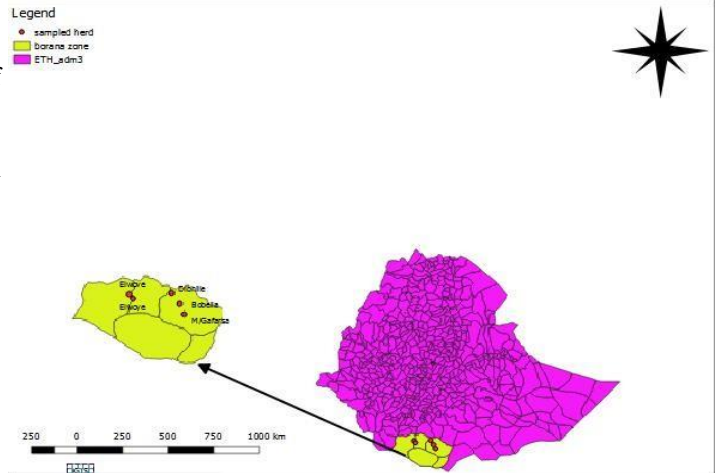
*B. abortus* “strain 19” or S19 (here after, S19) is an effective vaccine to prevent brucellosis until it was replaced by RB51. *Brucella* Strain 19 maintains its smooth appearance derived from the presence of the extracellular lipopolysaccharide (LPS). *B. abortus* strain RB51 vaccine has been developed in United States and tested for its efficacy and safety. This mutant strain of *B. abortus* does not produce cross-reacting antibodies in vaccinated animal that are detected in the routine surveillance tests. It means that animals vaccinated with RB51 remain negative on the brucellosis surveillance tests and do not give false positive results. This is because *Brucella* strain RB51 is rough as it lacks the lipopolysaccharide O chain, this feature gives it an advantage because it does not induce the antibodies that are detected by official diagnostic tests, resulting in the differentiation of vaccinated from infected animals [62]. Control of brucellosis in pastoral settings is difficult because of inaccessibility of public and veterinary health services, close contact between animals and their owners, ingestion of unpasteurized dairy products, and seasonal changes in livestock composition. Economic and cultural dependence of pastoral communities on their livestock implementing strategies based on culling infected animals is not acceptable, because animals are primary source of livelihoods. Therefore, the disease has a stable transmission level and tends toward persistence and endemic stability [63].

### 3. Materials and Methods:

#### 3.1. Study Area:

This study was conducted in Borana zone, which is among the 20 zones found in Oromia National Regional State. The zone has thirteen pastoralist District namely, Arero, Dhas, Dillo, Dirre, Dubluk, Eelwoye, Gomole, Guchi, Miyo, Moyale, Taltale, Yaballo and Wachile, and one town administration Yabello town. Borana zone is located 4° 3' to 5° N latitude and 37° 4' E to 38° 2' E longitudes and the landscape is characterized by slightly undulating peaks up to 2000 meters above sea level (masl) in some areas. It shares common boundaries with Guji zone in the east, Somali National Regional State in south east, southern Nation's Nationalities and Peoples Rational State in the west and one international boundary with Kenya in the south [64]. The area is characterized by bimodal pattern of rain with annual average precipitation ranging from 300mm to 700mm. the main rainy season locally known as "Ganna" extending from mid of March to May and small rainy season termed "Hagayya" from mid of September to mid-November. The other two seasons are the cool dry season "Adoolessa" extending from June to August and the major dry season "Bona" extending from December to February. Animal husbandry in the region is characterized by extensive pastoral productions system and seasonal mobility. Cattle are the dominant animal species followed by goats, camels and sheep [65]. Two districts namely Arero and Elwoye were selected purposively

for this study (Figure 3).



**Figure 3:** Map of Ethiopia and Borena pastoral zone (Developed from Ethiopian shape files using QGIS).

#### 3.2. Study Design:

A cross-sectional study design was conducted from November 2020 to April 2021 by using serological tests, the RBPT and i-ELISA to estimate the prevalence of *Brucella* infection in camels in the two selected districts. Information on each sampled camel including age, sex, herd size, parity, history of abortion, body condition, herd composition and physiological status of camels were record individually. Interview of pastoralists using questionnaires was conducted to assess the community knowledge and perception on camel brucellosis.

#### 3.3. Sample Size:

The sample size for this study was estimated by the formula given by Thrusfield (2007);  $N = [1.96^2 P_{exp} (1 - P_{exp})] / d^2$ , Where: n= sample size,  $P_{exp}$ = minimum expected prevalence, 1.96= the value of Z at 95 % confidence interval d= desired accuracy level of 5 %. Therefore, by using the above formula and taking the previous prevalence of 3 %, the minimum sample size at 95 % confidence interval and at 5 % precision or accuracy level, the sample size is calculated to be 45 per district. However, the sample size increased to 315 (increased three times) to increase the precision of the estimates.

#### 3.4. Sampling Method:

The sampling method used in this study was multistage sampling to select peasant associations (PAs), villages (Peasant associations) and herd and then finally the camels. The districts from the zone was selected purposefully based on camel population and abortion history and accessibility the districts to the main road by vehicles. Five pastorals is associations were selected randomly from the two districts selected. From these pastoral associations accessible herds were selected from which 315 were selected conveniently.

#### 3.5. Sample Collection:

##### 3.5.1. Blood collection:

About 8 mL of whole blood was collected from the jugular vein,



using plain vacutainer tubes and needles, from each camel aged six months and above. Each sample was labeled using codes specific to the individual animal and herd information. The tubes were tilted on a table overnight at room temperature to allow clotting.

Serum was collected by decanting [66]. The serum was stored at 20 °C in Yaballo Regional Veterinary Laboratory. During blood sample collection individual animal history including age, sex, herd size, parity; history of abortion, body condition and herd composition and herd size were recorded.

**3.6. Laboratory Techniques:**  
Based on the recommendations of World Organization for Animal Health (OIE) Rose Bengal Plate test (RBPT) and indirect enzyme linked immunosorbent assay (i-ELISA) were used in this study. The i-ELISA used in this study employ purified LPS antigen with good sensitivity [44].

### 3.6.1. Rose Bengal plate test:

Equal volume (30 µL) of stained antigen and test serum were mixed and rotated gently up to four minutes on a white tile or enamel plate. Based on the absence and presence of agglutination due to an antigen and antibody complex the result was read as positive or negative. To detect micro-agglutination results of RBPT magnifying glass was used and interpreted as 0, +, ++ and +++.

### 3.6.2. Indirect enzyme-linked immunosorbent assay:

The screened samples that positive by RBPT were further confirmed by i-ELISA to detect Brucella antibodies. In this study commercial i-ELISA kit (ID. Vet innovative diagnose ID Screen® Brucellosis Serum indirect Multi-species, BRUS-MSvar 1014GB) used to detect antibodies directed against *B. melitensis*, *B. abortus* and *B. suis* using short incubation method was used. A wash solution was dispensed into each well in 96 well plate pre coated with inactivated antigen *B. abortus* LPS. Specimen and the controls were added into the plate diluted at 1: 20. This mixture was gently shaken, covered with plate sealing tape and incubated at 37 °C for 30 minutes. Each well was washed with the wash solution approximately 300 µL three times to avoid drying of well between washing. The conjugate was added into each well, covered with plate sealing tape and incubated at 37 °C for 30 minutes. The plate with all its wells was re-washed three times with wash solution approximately 300 µL. The substrate was added into each well at room temperature 26 °C for 30 minutes incubated in dark. Finally, 100 µL stop solution was added and the ELISA reader machine was read plate [67].

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ELISA reader machine was measure Optical density (OD) at a wavelength of 450 nm. To assess the quality of a plate, the OD was not exceeding 2.00 for positive control and 0.500 for negative control. Results were calculated as percentage of the ratio between the corrected sample OD and positive control OD (S/P-ratio). S was the OD of the test sample minus the OD of the negative control (NCx), over P: the OD of the positive control (PCx) minus the OD of the NCx.  $S/P \% = 100 \times (\text{Sample} - \text{NCx}) / (\text{PCx} - \text{NCx})$ . A cut-off 5). Female camels with the history of abortion had higher

of  $\geq 80\%$  according to the manufacturer was to be considered for positive test samples [44].

**3.7. Questionnaire Survey:**  
Structured questionnaire was used to assess the awareness of the community (both owners and herders) about brucellosis in camels. The structure of the questionnaires focused on the perception and knowledge of the pastoral community about brucellosis in camels and was written in English and translated to local language (Afaan Oromoo). During pre-testing, additional information was gathered and some of the questions were modified. In total, forty-five (45) pastoralists whose animals were test for brucellosis were interviewed. The information gathered by the questionnaire was related the potential routes of transmission in animal and human, clinical signs in animal, species it affects, and measures taken to prevent and control the disease.

### 3.8. Ethical Clearance:

Written informed consent was obtained from all participants and legal guardians of minors. This study was approved by the Institutional animal care and use committee of Addis Ababa University College of veterinary medicine and agriculture.

### 3.9. Data Management and Analysis:

Data generated from the survey and laboratory investigations were recorded and coded using a Microsoft Excel spreadsheet (Microsoft Corporation) and analyzed using STATA version 13.1 for Windows (STATA Corp. College Station, TX, USA). The association between explanatory and outcome variable was analyzed at individual animal level by using logistic regression.

Associated risk factors and seroprevalence analysis was conducted with multivariable logistic regression and chi-square test model respectively. Prevalence was compared with the chi-square test as appropriate. Odds ratio was used to assess the strength of association between exposures variables associated with the disease in animals. For analysis of the effects of reproductive parameters and seroprevalence the analysis was conducted using mixed-effect logistic regression methods. The significance level was set at 5% and 95% confidence level where P value < 0.05 was set statistically significant.

## 4. Results:

### 4.1. Seroprevalence of Brucellosis in Camels and Associated Risk Factors:

Of a total of 315 dromedary camels (53 male and 262 female) tested by using RBPT 29 (9.21 %; 95 % CI: 6.25 – 12.95) of them were found positive. When the positive samples were subjected to i-ELISA 9 (2.86 %; 95 % CI: 1.31 – 5.35) of them gave positive results for Brucella infection. The prevalence was higher in camels tested from Elwoye district 4 (3.17 %; 95 % CI: 0.87 – 7.93) than those tested from Arero district 5 (2.65 %; 95 % CI: 0.86 – 6.06). The results serological test was given in Table 4.



prevalence brucellosis than those without history abortion, which was statistically significant difference (OR = 6.24; 95 % CI: 1.08 - 35.86) (Table 6).

Variables	No. examined	No. Positive	Prevalence	X <sup>2</sup>	P-value
<b>District</b>					
Arero	189	5	2.65%	0.08	0.783
Elwoye	126	4	3.17%		
<b>Sex</b>					
Female	262	8	3.05%	0.24	0.645
Male	53	1	1.89 %		
<b>Age</b>					
Young	83	1	1.20 %	1.87	0.1716
Adult	53	1	1.89%		
Old	179	7	3.91%		
<b>Herd size</b>					
Small	106	1	0.94%	9.02	0.0027
Medium	167	3	1.8%		
Large	42	5	11.9%		
<b>Parity</b>					
No parity	87	1	1.15%	1.87	0.1710
Single parity	62	2	3.23%		
Two and more	113	5	4.42%		
<b>Reproductive problem history</b>					
Abortion	40	4	10 %	5.51	0.014
RFM	36	2	5.56%	0.74	0.358
Stillbirth	30	1	3.33%	0.01	0.925
<b>Body condition</b>					
Poor	148	5	3.38%	0.22	0.6380
Medium	84	2	2.38%		
Good	83	2	2.41%		
<b>Herd composition</b>					
Camel only	48	1	2.08%	0.68	0.4101
Camel & Bovine	49	1	2.04%		
Camel & shoats	92	2	2.17%		
All specious	126	5	3.39%		

**Table 4:** Results of Univariable analysis to identify risk factors

Risk factor	Odds ratio	Std. Err.	Z	P>z	[95%Conf. f.]	Interval ]
<b>District</b>						
Elwoye	1.160226	0.9025108	0.19	0.848	0.252589	5.32931
<b>Sex</b>						
Female	0.6221431	0.8071894	-0.37	0.715	0.048923	7.911663
<b>Age</b>						
Adult	1.927058	2.832757	0.45	0.655	0.108053	34.36789
Old	5.588583	7.002644	1.37	0.170	0.4794279	65.14484

Body condition	Odds ratio	Std. Err.	Z	P>z	[95%Conf. f.]	Interval ]
Medium	0.7789702	0.6905897	-0.28	0.778	0.137055	4.427379
Good	0.9508267	0.8709119	-0.06	0.956	0.1579215	5.724814
<b>Herd size</b>						
Medium	1.829599	2.194525	0.50	0.615	0.1743316	19.20152
Large	17.03541	19.68525	2.45	0.014	1.769079	164.043
<b>Herd composition</b>						
Camel & Bovine	0.9690467	1.460472	-0.02	0.983	0.0505219	18.58703
Camel & Shoats	1.346338	1.753916	0.23	0.819	0.1047774	17.29979
Camel, Shoats & Bovine	1.572648	1.928595	0.37	0.712	0.1421582	17.39766

**Table 5:** Results of multivariable analysis to identify risk factors

Risk factor	Odds ratio	Std. Err.	Z	P>z	[95% conf.]	Interval]
<b>Parity</b>						
Single parity	1.788536	2.331348	0.45	0.656	.1389827	23.01625
More than one	2.295611	2.770359	0.69	0.69	.2156067	24.44186
<b>Abortion</b>						
	6.23754	5.566409	2.05	0.040	1.084919	35.86158
<b>Stillbirth</b>						
	.6958434	.7892473	-0.32	0.749	.0753446	6.426445
<b>RFM</b>						
	.5955831	.5905866	-0.52	0.601	.0852868	4.159134
<b>_cons</b>	.0117072	.0117072	-4.42	0.000	.0016303	.084068
<b>_cons</b>	<b>.0117072</b>	<b>.0117072</b>	<b>-4.42</b>	<b>0.000</b>	<b>.0016303</b>	<b>.084068</b>

**Table 6:** Result of association between seroprevalence and reproductive parameters.

**4.2. Results of Questionnaire Survey:**

**4.2.1. Sociodemographic characteristics of respondents:**

A total of 45 respondents interviewed during this study which 27 (60%) of them were from Arero and 18 (40%) were from Elwoye districts. The majority 36 (80 %) of the participants was males and the remaining 9 (20 %) were females. When their age is considered 53.33% participants were between 25 to 45 ages. Majority of the participants were camel owner 29 (64.44%) while other is camel herder 16 (35.56%). Most of the animal herders had greater knowledge about the brucellosis than camel owner which was statistically significant (P<0.05) (Table 7).



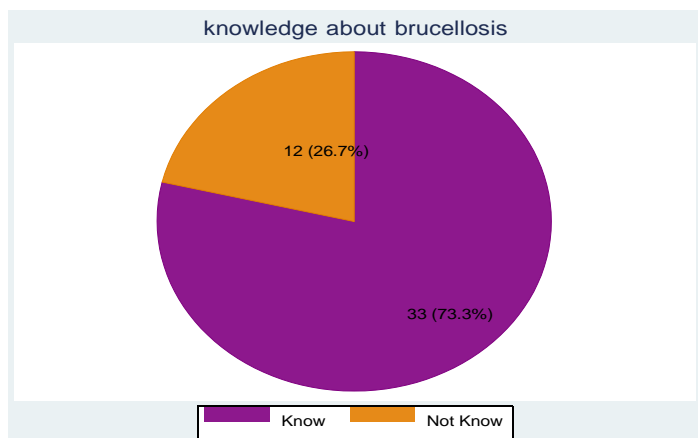
Demography		Respondents	Level of Knowledge	
			Frequency (%)	p-value
Gender	Male	36	26(72.2%)	0.7
	Female	9	7(77.8%)	
Age	< 25	13	12(92.3%)	0.07
	25-45	24	17(70.8%)	
	>45	8	4(50%)	
Occupation	Owner	29	18(62%)	0.013
	Herder	16	15(93.8%)	
District	Arero	27	21(77.8%)	0.4
	Elwoye	18	12(66.7%)	

**Table 7:** Level of knowledge regarding animals brucellosis compared with socio-demographic

**4.2.2. Level of knowledge and perception on brucellosis:**

The level of respondents' knowledge regarding brucellosis was high; 33 (73.33 %) respondents knew about the disease which is locally known as "salleessa/salleessisa" (figure 4). Most of them had heard about brucellosis from their family, neighbors and Personal observation 31 (93.94 %) whereas others got information from traditional healers 4 (12.12 %) and animal health workers 1 (3.03 %).

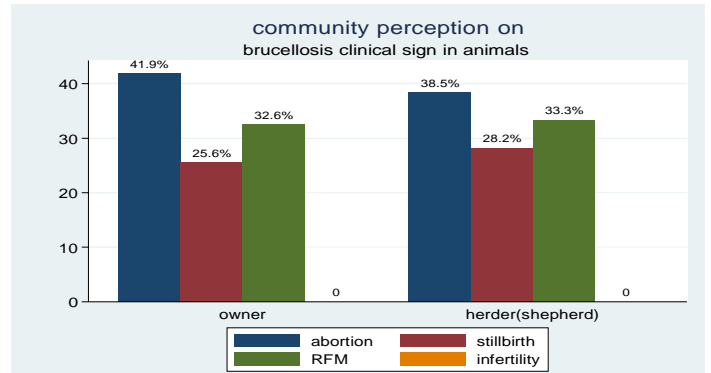
The pastoral communities have been living with their animals for generations and have built enormous indigenous knowledge on animal health problem. Knowledge transfer from the animal health worker to the society is a key intervention for the prevention, control and eradication of disease. The eminent gap of perception of the society about animal disease was due to the absence of well-designed attempt of animal health extension service, Poor infrastructure that constrain access to mobile livestock communities, integration of the CAHWs system into the veterinary service and limited resources to maintain service delivery.



**Figure 4:** Respondents' knowledge about brucellosis

The participants disclosed that they have been aware about the symptoms of brucellosis in camels. Although few differences was observed in signs described by the pastoralists, the majority

revealed that common signs of brucellosis include abortion, RFM and stillbirth with 100%, 81.82% and 66.67%. There was similar knowledge between camel owners and herders with regard to recognition of signs of brucellosis in camels (figure 5).



**Figure 5:** Respondents' knowledge about sign of brucellosis in camel between owner and herder

Variable	Category	Percent %	Frequency(N=45)
Do you know brucellosis	Yes	73.33%	33
	No	26.67%	12
Your source of information	Health care workers/ CAHWs	3.03%	1
	Neighbors/family/Personal observation	93.94%	31
	Traditional Healer	12.12%	4
	FM-Radio	-	-

**Level of Knowledge from who claimed knowledge of brucellosis**

Variable	Category	Percent %	Frequency(N=33)
Animal species it affect	Camel	100%	33
	Cattle	100%	33
	Goat	100%	33
	Sheep	21.21%	7
	Wild animals	-	-
Symptoms in animals	Abortion	100%	33
	Retain placenta	81.82%	27
	Stillbirth	66.67%	22
	Swollen leg joints	-	-
	Infertility	-	-
	reduced milk production	-	-
Do you know brucellosis as zoonotic disease	Yes	18.18%	6
	No	81/82%	27
Symptoms in man	Fever	-	-
	Joint pain	3%	1
	Headache	-	-

**Table 8:** Respondent Level of Knowledge and perceptions regarding brucellosis

**4.2.3. Respondent knowledge of risk factor**

The majority 27 (81.82 %) of the participants were not aware about the transmission methods of brucellosis between domestic animals



and humans. Concerning the zoonotic nature of brucellosis only 6 (18.18 %) of respondents knew that it is transmitted from animals to humans, of which 5 (15.15 %) mentioned consumption of raw milk as most common mode of transmission from. Transmission from animals to animals was mentioned by 9 (27.3%) of the pastoralists to by mixing of different animals species and contact with aborted materials was indicated by 2 (6.06 %) of the respondents. Different and common overall seasonally occurrence of brucellosis in animals mentioned by respondents are Major Rainy Season (“Ganna”), Cool Dry Season (“Adololessa”), Short Rainy Season (“Haggaya”) and Major Dry Season (“Bona”) with 93.94%, 24.24%, 21.21% and 9.09%. The details of the results of questionnaire are presented in (Table 9).

Variable	Category	Percent(%)	Frequencies (N33)
Source of infections for animals	contact with aborted material	6.06%	2
	Sharing the same pasture/water	–	–
Sex more affected	Introducing brucellosis infected animal into a herd	–	–
	Mixing with brucellosis infected or different domestic animals	27.3%	9
Age Brucellosis more common in animal	Male	–	–
	Female	100%	33
Season Brucellosis more prevalent	Young (< 3 years )	–	–
	Adult (3 - 4 years)	–	–
	Old (> 4 years)	100%	33
Transmission methods to man	Major Rainy Season (“Ganna”)	93.94%	31
	Cool Dry Season (“Adololessa”)	24.24%	8
	Short Rainy Season (“Haggaya”)	21.21%	7
	Major Dry Season (“Bona”)	9.09%	3
Transmission methods to man	Consuming raw milk	15.15%	5
	Consuming raw meat/ blood	–	–
	Contact with aborted fetus	–	–
	No idea	84.85%	28

**Table 9:** Respondent level of knowledge about brucellosis risk factor

**4.2.4. Community practice regarding Brucella Prevention and control:**

All Respondents 100% described that aborted material and other excreta are handled with bare hands, and they did not use any protective material while handling parturient livestock, removing placenta and other aborted materials. With limited knowledge about their responsibilities in the prevention and control of zoonotic disease, animal and human health care workers are not equipped to advise the public on appropriate prevention and control strategies. Some of this lack of knowledge can be explained

Variable	Category	Percent (%)	Frequency(N=33)
Use personal protective when Delivery assistance or contact aborted material	Yes	–	–
	No	100%	33
Proper disposing aborted foetus/fetal membrane	Yes	–	–
	No	100%	33
Mating assistance	Yes	100%	33
	No	–	–
Intervention you take if animals have brucellosis	Isolate	–	–
	Cull	6.06%	2
	Self-treatment	15.15 %	5
	Take to clinic	–	–
	Do nothing	78.79 %	26
Raw milk consumption	Yes	100%	33
	No	–	–
Raw meat consumption	Yes	100%	33
	No	–	–
Milk usage	For Sale	100%	33
	For family	100%	33

**Table 10:** Community practice regarding brucellosis in study area

Camel brucellosis sero-positive in herd level was insignificant (p > 0.05) different association between respondents those who know and not know brucellosis (Table 11).

Respondents knowledge level	OR	Std. Err.	Z	P> z	[95% Conf. Interval ]
Know Brucellosis	Yes	-	-	-	-
	No	3.625	2.94	1.59	0.113
Constant.	0.137	0.07	-3.71	0.000	0.05 - 0.39

**Table 11:** Univariable logistic regression analysis of respondent level of knowledge regarding Brucellosis for Brucella seropositivity found at herd level

**5. Discussion:**

Camel production has been considered an important economic activity in Borana pastoral area and remains so in the future. However, the optimal utilization of this important resource can be impaired by infectious diseases such as brucellosis. Brucellosis affects the productivity and reproductive efficiency of animals through reduction of milk production, abortion and decreased fertility [68]. This study provides important information on the



occurrence of brucellosis in camels in Borana pastoral zone. herd considered as all the animals in the herd were productive. Although the prevalence observed is low, it is not without impacts. Therefore, low milk yield and infertility of some animals was not Since animals and humans live intimately in the area sometimes considered as a threat to the overall productivity which allowing sharing shelters the occurrence of brucellosis in camels has them to keep chronically sick animals and less productive. These important implication for public health. Thus, it is an addition to sick animals have other values attributed to them, such as infertile the existing information on brucellosis in livestock. Previously the animals being valued for their size or animal with low milk yield occurrence of brucellosis has documented in other livestock being thought of as calmer. Consequently, as a result of these species [61]. The prevalence of brucellosis observed in this study attitudes the spread of brucellosis in the herd is high [75]. Lower in camels is in close agreement with the 2.43 % prevalence prevalence in Small herd size could be associated with grazing at reported by [69] in Jijiga and Babile, eastern Ethiopia; the 2.09 % the pasture near to enclosure without long distance movement, prevalence reported by [5] in Afar, Northeastern Ethiopia; the easy to manage and identify sick animal which minimize reports of [42] and [70] who observed a 3 % prevalence in southern predisposing factors and avoid contact with other herd.

Ethiopia and that of [12] and [70] who reported similar prevalence in camels in Tigray, Northern Ethiopia. However, the results of this Abortion, retained fetal membrane and stillbirth were reproductive study are higher than that findings of [57], [59], [71], [4], [72] and problems obtained in the history of the adult female camels with that of [11] who reported lower sero-prevalence from different prevalence of, 10 %, 5.56% and 3.33% respectively. Statistically parts of the country. On the other hand the results this study is significant ( $p < 0.05$ ) difference sero prevalence was observed in lower than some of the reports done elsewhere in the world. For abortions which close agreement with [54] in Selected Districts of instance, it is lower than the prevalence of 5.8 % reported from Afar, Ethiopia, [12] Mehoni District, southeastern Tigray, Sudan [9], 5.7% from Libya [25], 10.5% recorded in Nigeria [30], Ethiopia, [70] in Yabello district of Borena Zone, southern 11.5% in Egypt [66], 14% and 15.36% in Kenya [73] and [26], Ethiopia, [59] in and Around Dire Dawa City , eastern Ethiopia, respectively, 8.15% in Iran [16], and 9.09% in, Pakistan [24]. [11] in Fentale district Oromia Regional State, Ethiopia, [69] in

Jijiga and Babile districts, Eastern Ethiopia, [14] in the Sultanate

The difference observed could be due differences herding of Oman and [20] in Yemen. Camel herd kept in close contact with structure, the laboratory tests used and the sample size used for the bovine and small ruminants in the current study sero prevalence investigation. In the current study area camels and other livestock were 2.08% in camel kept alone, 2.04% camel kept with bovine, species owned by different individuals and communities are often 2.17% camel kept with small ruminants and 3.39% camel kept with herded together. In contrast in places where most of the previous bovine and small ruminants. However, no statistically significant studies were under taken there is clan-based herd segregation, difference was observed between these four camel groups. The which is likely to reduce introduction and spread of Brucella present finding was in line with the observation in Yabello District among herds or animals. The sensitivity and specificity of the of Borena Zone, Southern Ethiopia by [70] and in Southern confirmatory test also vary and may have contributed to the lowland of Ethiopia by [42]. The results of my study go parallel variation in prevalence. The differences could also be due to with the findings of [5] in Afar.

variations in animal management practices, the number of susceptible camels, presence of high number of camels in the herds Animal species diversification is common in Ethiopia and has and mixing of aborting camels with others and absence of economic and ecological advantages. However, it increases the accessible preventive veterinary services, close contact with chance of brucellosis and other disease transmission from other infected domestic and wild animals, population intensity, lack of infected ruminants to dromedaries [32]. spread of the disease awareness about the disease in camels [68]. In relation to among animal may be mixing of different species during husbandry practices, these animals are usually kept overcrowded movement for grazing and watering in the dry season as and reared in open system without differentiation of aborted and aggregating the animals around watering point might increase the pregnant ones and housed together with high stocking density, all contact between infected and healthy animals and thereby facilitate these factors play important role in the spread of the infection. The the spread of the disease [42]. Body condition of the camels was lack knowledge on the mechanisms of transmission of Brucella considered in this study to see the distribution of the infection in species might have caused Brucella infected animals used for different body condition. Even though, in this study body condition breeding purpose which serves as source of infection [74]. score was statistically not significant ( $P > 0.05$ ), high seropositivity was found in camels with poor 3.38% and medium 2.38% body

Herd size was highly statistically significant ( $P=0.017$ ) risk factor condition than camels with good 2.41% body condition. Body for camels brucellosis in this study. It is likely that the risk of condition of the camels was considered in underfed animals are disease transmission increased in a large herd size this is in expected to have a poor body condition that is manifested by accordance with the findings of [32] in the Afar region of Northeast decreased immunity against various infections [54]. Nutrition Ethiopia, [72] in selected districts of afar region, Ethiopia and [5] plays a great role in Immunity against various infectious diseases. in Selected Pastoral Districts of Afar, Northeastern Ethiopia. As Underfed animals are expected to have a decreased immunity that herd size increases, the chance of contact between animals is manifested by poor body condition [58].

increases leading to more chances of infection particularly during calving and abortion [54]. As herd size increases, the chances of Age was classified as young ( $< 3$  years), adult (3-4years) and old contact between animal also increases, leading to more chances of ( $>4$ ) based on sexual maturity to see the distribution of diseases in infection which is particularly more important during calving or each age group. Accordingly, this study reveals that brucellosis abortion when maximum brucellosis contamination occurs. Thus, infection may occur in animals of all age groups, but commonly herd size and density of animal population together with poor persists in old camels (3.91%) than in adult (1.89%) and young management are directly related to infection rate [5]. Owing large (1.20 %). Although no statistically significant difference ( $P > 0.05$ )



was observed between each age groups, being higher in the previous reports in Ethiopia [81], Uganda [82] and Kenya [83] but sexually mature age group. Susceptibility to brucellosis in camels differed from others studies conducted in Egypt [84], Malawi [85], of different age groups slightly higher in older animals which is in Pakistan [86] and [87]. Pastoralist communities have been living agreement with the previous reports of [59], [71], [32], [76], [54], with their animals for generations and have built enormous [69], [4], [14] and [78] [44] is infection may occur in animals of all indigenous knowledge with animal health problem but concerning age groups but persists commonly in older and adult animals which brucellosis being zoonosis Only 6 (18.18%) participants could sexually mature due to presence of growth factors (erythritol and correctly identify that diseases can be transmitted from livestock hormones) which favors the multiplication of pathogen in sexually to humans.

mature animals [58]. Animals which older and adult are at risk of infection due to sexual mating and diseases transmission [54].

The accessibility of information related to brucellosis, the main sources of information in study area were from family, neighbor Brucellosis in all age groups of camels indicates that infection and personal observation 31 (93.94 %). About 4 (12.12 %) and 1 started early in life probably through sucking and persisted into (3.03 %) of participants said that they have heard about brucellosis adulthood [78]. Younger animals tend to be more resistant to from traditional healer and health care workers or CAHWs infection due to less development sex hormones and erythritol respectively. FM-Radio were not a source the information for which stimulate the growth and multiplication of *Brucella* pastoralist community in study area. This in line to Surveys organisms, tend to increase in concentration with age and sexual conducted in Uganda [82]. Contrary to this finding the study in maturity [78]. Although a few latent infections, frequently clear Kenya [83] found CAHWs were the main source of information infections [4] and less exposure may occur. Sero-prevalence of for pastoralist. Knowledge transfer from the animal health care camel brucellosis according to sex was higher in female 8 (3.05 %) worker or CAHWs to the society is a key intervention for the than male 1 (1.89 %). This is consistent with previous study of [11], prevention and control of disease. The eminent gap of perception [4], [42] [58], [59], [71], [77], [72], [5], [70], Warsame et al., of the society about animal disease from animal health worker was (2012) from Ethiopia, [9] from Sudan, [79], [24] from Sindh due to the absence of well-designed attempt of animal health Pakistan and [19] in the south province of Jordan. Although no extension service, poor infrastructure that constrain access to statistically significant difference ( $P > 0.05$ ) was observed between mobile livestock communities, absence of integration system of CAHWs into the veterinary service and limited resources to maintain service delivery [88].

Females are at high risk of brucellosis than males due to their

usefulness in the production herds, females generally have a longer Almost all participants were knowledgeable about the brucellosis lifespan than males, and this may have increased exposure to the susceptibility of different animal species because of rearing bacterium [14]. Reduction of immunity in females during lactation, different livestock specious for diversification which has economic pregnancy and other reproductive stress may also contribute to and ecological advantages and increase community awareness higher prevalence in female camels [66]. There was no statistically regarding disease in livestock. Identifying brucellosis affected significant difference between the sex groups in the current study. species and its symptoms are crucial for livestock owners' These results may be associated with the effect of erythritol in both practices towards prevention and control measures of brucellosis. sex [66]. The number of breeding males kept by the pastoralists in In this study all participants who claimed to know brucellosis had the camel herds is very small on which random sampling method high level of knowledge of animal brucellosis clinical signs, was applied and this predictably bias the statistical analysis [32]. mostly recurrent abortion, RFM and stillbirth in contrasts with On the contrary [70], [4], [12] and [69] in Ethiopia and [80] in studies in Kenya [83], conversely very low knowledge of the Sudan and [78] in Mongolia reported the occurrence of infection is symptoms of brucellosis was fund infertility, reduced milk higher in male than female animals. This might be due to the production and swollen leg joints which similar to studies done in number of breeding males kept by the pastoralists in the camel Uganda [82] and Egypt [89]. The major risk factors of brucellosis herds of the present study was very small on which random transmission among different animal specious was mentioned by 9 sampling method was applied and [70].

(27.3%) respondents as mixing animal with brucellosis infected different livestock and through contact with aborted material was Parity was statistically insignificant difference ( $P > 0.05$ ) in this identified by 2 (6.06%) of the participants. With regard to public study. Prevalence of she-camels with the history of no parturition, health importance of brucellosis majority 27 (81.82 %) of the Primiparous and Pluriparous were 1.15%, 3.23% and 4.42%, participants were not sure that brucellosis can be transmitted from respectively. Therefore, this is consistent with the previous study animals to humans. Only 5 (15.15%) participants could identify by [32] and [54] in Afar, [70] in Yabello District of Borena Zone, that brucellosis can be transmitted from livestock to humans via Southern Ethiopia, [42] in Southern lowland of Ethiopia and [43] raw milk consumption. It comparable with earlier findings in in selected districts of Punjab, Pakistan. This might be due to Ethiopia [90] and [91], Pakistan [86], Kenya [83] and Uganda [82]. repeated exposure of the she- camels to parturition and other physiological stress increases the probability of acquiring *Brucella* In pastoral community raw milk consumption was regularly used infection [70]. On the contrary [59] reported the occurrence of as a replacement for drinking water however, consumption of raw infection is higher in single parturition than no parturition two and animal products and close contact with animals were not perceived more female animals [54]. Identifying the level of community as risk factors for a disease. This relates to a long-standing perception regarding brucellosis contributes to control at the traditional practice engaging in increase transmission of disease. human-animal interface through awareness creation. Low awareness about brucellosis transmission by eating habits or Understanding of participants toward brucellosis in animal was animal management practices makes communities vulnerable to seen in a large proportion of (73%) participants. This is in line with disease, threatens livestock and cause economic losses [92].



Knowledge gap that lead community to engage in high risk throughout my life.

practice such as improper disposal of aborted materials on field and assist their animals in the parturition without any protective wearing identified as priority problem in study area. These findings

were in the same line with [81] and [93] in Ethiopia. Combined factors of handling abort material without protective gloves and camel brucellosis in Borana zone was low but it is enough to affect poor cleaning practice could pose a great risk of disease spread to animal health, human health and the economy. Among the human and animals. Aborted fetuses and the placenta Proper potential risk factors assessed only large herd size and history handling decrease incidence and environmental transmission of abortion were significantly associated with camel brucellosis in the brucellosis. According to our study, 2 (6.06%) participants may area. The majority of the community had moderate overall sell frequently aborted animals in their herds, potentially knowledge regarding camel brucellosis but most of the community increasing brucellosis transmission not only between households had no knowledge about the zoonotic importance of brucellosis, its in the same village, but also across larger geographical areas. This transmission mechanisms, consequences of consuming raw milk, finding is similar to study in Ethiopia [93], Jordan [94] and Egypt handling aborted fetus and fetal membranes without any protective [95]. Participant mentioned actions taken when confronted with materials. Therefore, the veterinary and public health authorities as aborting animal in the herd ware without seeking veterinary well as the extension service should take the results of this study services give treatment by themselves and majority of them would into account when planning livestock and public health do nothing without isolate brucella infected animal. Failure to isolate suspected animals has been one of the major risk factors for transmission of Brucellosis within and between herds [83].

Only a small proportion of respondents perceived that brucellosis was a serious disease in animals and humans but they had unfavorable attitude towards prevention of brucellosis. Lacks of awareness of brucellosis were more likely make them to engage in risky practices that could expose them to infection. Effective control strategies cannot be currently implemented due to the lack of awareness high-risk practices like close contact between animals, ingestion of unpasteurized dairy products, and different livestock composition. Economic and cultural dependence of pastoral communities on their livestock implementing strategies based on culling infected animals is not acceptable [63]. Because of a lack of community awareness high-risk activities and lack of effective prevention and control measures are currently unavailable. Control programs could be more successful by educating livestock owners/herder and change their behavior towards disease control and animal health. Knowledge, attitude and practices survey play a vital role in evaluating livestock owners' understanding and preparedness against such livestock diseases [87].

### 5.1 Study limitations:

As most of the survey was conducted in drought season, some of pastoralists refused to allow collecting blood sample while animals are in poor body conditions. Seasonal migration of livestock in Borana in search of good pasture and watering points could be associated with temporal variation of prevalence of the disease that was not assessed due to the cross-sectional design of the current study. The major limitation of the study was the small sample size of questionnaire which could affect the power of the study and external validity of the findings making it impossible to generalize findings even to the whole.

### 5.2 Acknowledgements:

Above all, all praises and thanks are for Almighty ALLAH, Most Merciful and Most Gracious, who's enabled me to perceive and pursue ideas of life, blessed me with good health and proper guidance. My special thanks also to my Father Giro Dika and my mother Loko Jarso, who has been giving me unpaid sacrifice

## 6. Conclusion and Recommendations:

The results of present study revealed that of the prevalence of factors of handling abort material without protective gloves and camel brucellosis in Borana zone was low but it is enough to affect poor cleaning practice could pose a great risk of disease spread to animal health, human health and the economy. Among the human and animals. Aborted fetuses and the placenta Proper potential risk factors assessed only large herd size and history handling decrease incidence and environmental transmission of abortion were significantly associated with camel brucellosis in the brucellosis. According to our study, 2 (6.06%) participants may area. The majority of the community had moderate overall sell frequently aborted animals in their herds, potentially knowledge regarding camel brucellosis but most of the community increasing brucellosis transmission not only between households had no knowledge about the zoonotic importance of brucellosis, its in the same village, but also across larger geographical areas. This transmission mechanisms, consequences of consuming raw milk, finding is similar to study in Ethiopia [93], Jordan [94] and Egypt handling aborted fetus and fetal membranes without any protective [95]. Participant mentioned actions taken when confronted with materials. Therefore, the veterinary and public health authorities as aborting animal in the herd ware without seeking veterinary well as the extension service should take the results of this study services give treatment by themselves and majority of them would into account when planning livestock and public health do nothing without isolate brucella infected animal. Failure to isolate suspected animals has been one of the major risk factors for transmission of Brucellosis within and between herds [83].

Therefore, based on the above conclusion the following recommendations were forwarded:

- Further epidemiological studies with isolation and identifications of *Brucella* biotypes involved in camel brucellosis
- Establish participatory epidemiology and further studies on factors affecting of the occurrence of brucellosis in camels
- Public awareness should be given for pastoral community on economic and zoonotic importance of brucellosis.
- Collaboration between public health and veterinary to increasing awareness about the disease symptoms (animal and human), transmission (animal and human), control and prevention methods.

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