Review of Bovine Brucellosis and Its Public Health Significance

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Research Article

Abstract
Brucellosis is an infectious zoonotic bacterial disease caused by a member of the genus Brucella. The disease affects both animals and human beings resulting in a serious economic loss in the animal production sector and deterioration of public health. Bovine brucellosis is highly prevalent and has significant economic and zoonotic implications for the rural communities in consequence of their traditional lifestyles, feeding habits, and disease patterns. The possible sources of infections include all infected tissues, aborted fetuses, vaginal discharges, and potentially contaminated materials. The nature of the pathogenesis of the diseases lies in the presence of the bacteria in the cells and employing various methods to survive in the phagocytic cells. The disease can be transmitted from an infected host to susceptible animals in direct and indirect contact. Various methods are employed for the diagnosis of brucellosis including microscopic examination, culture methods, serological and molecular biology. The public health importance of brucellosis is much related to the infected animal species from which human transmission occurs. The economic importance of brucellosis depends upon the species of animal affected. It can cause considerable losses in cattle as a result of abortion and a reduction in milk yield. The most rational approach for control of Brucella abortus infection is by vaccinating young female animals. To deal with diseases like brucellosis, the public in general and high-risk groups, in particular, should be made aware of the zoonotic and economic importance of brucellosis through veterinary extension education.

Keywords: Bovine, Brucellosis, Public Health

1. Introduction
Since the earliest days of civilization, man is closely associated with animals and thus gave an opportunity for the inter communicability of microbial infections between humans and animals (Radiostitis et al., 2007). There are many diseases of cattle, which are transmitted to humans (Megersa et al., 2011). Among these, brucellosis (Bang’s disease, Contagious abortion, Malta fever, Undulant fever) is a highly infectious disease of humans and animals, which has been reported from many countries of the world including Ethiopia (Moti et al., 2013).
Ethiopia has one of the largest livestock resources in Africa, with a total cattle population of 47.6 million (CSA, 2012/13). Livestock contributes more than 30% of the agricultural gross domestic product and 19% in export earnings. Oxen provide draught power to cultivate grain crops in rural agriculture, which is the backbone of the economy. The comparatively huge livestock resources of the country and the economic return gained from this subsector do not coincide, because of prevalent infectious diseases, among other factors. Bovine brucellosis is one of these infectious diseases and has been reported from several parts of the country (Asmare et al., 2010).

The disease is caused by Gram-negative, facultative intracellular bacteria that can infect many species of animals (Teshome et al., 2003). Brucella comprises ten species; six of the ten can be isolated from terrestrial mammals: B. abortus, B. melitensis, B. suis, B. canis, B. ovis and B. neotomae (John et al., 2010).

Brucellosis in cattle is usually caused by biovars of Brucella abortus. In some countries, particularly in southern Europe and western Asia, where cattle are kept in close association with sheep or goats, the infection can also be caused by B. melitensis (Godfroid et al., 2013). Occasionally, B. suis may cause chronic infection in the mammary gland of cattle, but it has not been reported to cause abortion or spread to other animals (Shirima, 2005).

Bovine brucellosis is clinically characterized by abortion and retained fetal membrane (RFM) in cows and orchitis and epididymitis in bulls (Acha and Szyfres, 2003). Sources of infection include aborted fetuses, fetal membranes, vaginal discharges, and milk from infected cows. The most common route of transmission in cattle is through direct contact with an aborting cow and the aborted fetus or by indirect contact with contaminated fomites. Ingestion of contaminated pasture, feed, fodder, and water may also play a secondary role (Bhat et al., 2010). It is an economically important disease of livestock causing reproductive wastage through infertility, delayed heat, loss of calves, reduced meat and milk production, culling, and economic losses from international trade bans (Mangen et al., 2002).

Humans are almost exclusively exposed to brucellosis through contact with animals and food of animal origin or transmitted via human contact with secretions, predominantly through calving and abortions, this disease can also be spread through the consumption of contaminated, unpasteurized dairy products. Although the disease is characterized by febrile illness in humans, it is difficult to diagnose solely from the clinical picture, due to its similarities to other febrile diseases, such as malaria or typhoid. Brucellosis is endemic in many countries and is responsible for considerable economic and health burdens (Bandara and Mahipale, 2002).

Although the disease has been eradicated from most of the developed countries, it is still a major public and animal health problem in many developing countries, where livestock is a major source of food and income (Acha and Szyfres, 2003). The incidence of human brucellosis is correlated with the level of incidence in domestic animals (Bhat et al., 2010). Human cases occur after ingesting raw milk and milk products and coming into close contact with infected animals. Human brucellosis can be a very debilitating disease, although the case fatality rate is generally low (Acha and Szyfres, 2003).

The aims of this paper:
- To review cattle brucellosis
- To review the zoonotic importance of brucellosis

2. Literature Review
2.1. The Causative Agent

Brucella is a Gram-negative, facultative intracellular bacteria that can infect many species of animals, including humans. Ten species are recognized within the genus Brucella. There are six ‘classical’ species, B. abortus, B. melitensis, B. suis, B. ovis, B. canis, and B. neotomae, and another four species have been recognized more recently (Corbel et al., 1997).
Brucella abortus is the causative organism for bovine brucellosis. Brucella abortus is mainly infective for cattle, but occasionally other species of animals such as sheep, swine, dogs, and horses may be infected. Cattle can also become infected by B. suis and B. melitensis when they share pasture or facilities with infected pigs, goats, or sheep. The infections in cattle caused by heterologous species of Brucella are usually more transient than that caused by B. abortus (Acha and Szyfres, 2003).

2.1.1. Resistance and survival properties
Under appropriate conditions, Brucella organisms can survive in the environment for a very long period. Their ability to withstand inactivation under natural conditions is relatively high compared with most other groups of non-spore-forming pathogenic bacteria (Joint FAO and WHO, 1986). B. abortus is sensitive to pasteurization temperatures and its survival outside the host is largely dependent on environmental conditions. The pathogen may survive in an 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 feces, and eight months in liquid manure tanks (Maudlin et al., 2009).

Survival is prolonged at low temperatures and organisms will remain viable for many years in frozen tissues. Brucella in aqueous suspensions is readily killed by most disinfectants. A 10g/l solution of phenol will kill Brucella in water after less than 15 minutes of exposure at 37°C. Formaldehyde solution is the most effective of the commonly available disinfectants, provided that the ambient temperature is above 15°C (Joint FAO and WHO, 1986).

2.1.2. Occurrence and prevalence of infection
Bovine brucellosis is widespread throughout the world except for the developed countries where eradication has been achieved (Joint FAO and WHO, 1986). Many countries have made considerable progress with their eradication programs and some have eradicated the disease (Radostits et al., 2000). The disease has been eradicated in Finland, Norway, Sweden, Denmark, Netherlands, Belgium, Switzerland, Germany, Australia, and Hungary, Romania, and Bulgaria as well as other countries. Most European countries are free of bovine brucellosis (Acha and Szyfres, 2003).

It is of major economic importance in most developing countries, which have not had a national brucellosis eradication program (Radostits et al., 2000). In addition, the policy of many developing countries of importing exotic high production breeds without having the required veterinary infrastructure and the appropriate level of development of the socio-economic situation of the animal holder aggravates the situation (Seifert, 1996). In most developing countries, resources have not been sufficient to control brucellosis. Although the information on prevalence is inadequate, there are indications of a very high incidence in many areas, particularly in tropics; in countries, that can least afford the loss in milk production and animal protein that accompanies this disease (Joint FAO and WHO, 1986).

The disease is prevalent in many countries of Africa (Seifert, 1996). The reason for the high prevalence is probably due to the fact that many countries have not yet started control or eradication schemes (Radostits et al., 2000). A prevalence of 5% and 9.9% were reported in Uganda, Tanzania, and Kenya (Kagumba and Nandokha, 1978) respectively. Four hundred ninety-nine sera sampled from cattle in the Djibouti Republic revealed a prevalence of 4% (Radostits et al., 2000). In Southern Sudan, 6.5% prevalence was found in Dinka cattle of which 9.4% of female Dinka cattle have been slaughtered because of infertility caused by brucellosis (Hui, 1994). In Eritrea, 5.6% of seropositive animals to Brucella species have been detected in cattle (Omer et al., 2000).
2.1.3. Possible risk factors for infection

2.1.3.1. Animal risk factors
The susceptibility of cattle to B. abortus infection is influenced by the age, sex, and reproductive status of the individual animal. Sexually mature pregnant cattle are more susceptible to infection with the organism than sexually immature cattle of either sex. Susceptibility increases as the stage of gestation increases (Colibaliy and Yamego, 2000). Most animals infected as adults remain infected for life. Herd size and animal density are directly related to the prevalence of disease and difficulty in controlling infection in a population. Calving practices also play a major role in the spread of brucellosis. Separate calving pens allow for minimizing exposure of uninfected animals. Whether a herd raises its own replacement animals or purchases replacement animals affects the potential for introduction into the herd (Radostits et al., 2006).

2.1.3.2. Pathogen risk factors
Brucella abortus is a facultative intracellular organism capable of multiplication and survival within the host phagocytic cells. The organisms are phagocytized by polymorphonuclear leucocytes in which some survive and multiply. The organism is able to survive in macrophages because; it has the ability to survive phagolysosomes. The bacterium possesses an unconventional non-endotoxin lipopolysaccharide which confers resistance to antimicrobial attacks and modulates the host immune response. These properties make lipopolysaccharide an important virulence factor for Brucella survival and replication (Jepkosgei, 2016).

2.1.3.3. Occupational risk factor
Laboratory workers handling Brucella cultures are at high risk of acquiring brucellosis through accidents, aerosolizing, and/or inadequate laboratory procedures. In addition to this, abattoir workers, farmers, and veterinarians are at high risk of acquiring the infection (Chain et al., 2005).

2.1.3.4. Management risk factors
The spread of the disease from one herd to the other and from one area to another is almost always due to the movement of an infected animal from an infected herd into a non-infected susceptible herd (Colibaliy and Yamego, 2000). Large numbers of organisms are shed from the reproductive tract when infected cows abort. In cows that lactate following the abortion, milk, including colostrum, is an important source of infection, and bacteria are excreted intermittently in milk throughout the lactation period. The fluid in hygromas caused by Br. abortus infection may contain large numbers of organisms, but because of being restricted to the lesion they do not seem to be important in the spread of the disease (Thomson and Tustin, 1994).

2.1.4. Source of infection and transmission
2.1.4.1. Sources of infection
The risk associated with exposure of susceptible animals to the disease following parturition or abortion of infected cattle depends on three factors: the number of organisms excreted the survival of these organisms under the existing environmental condition and the probability of susceptible animals being exposed to enough organisms to establish infection. Brucella abortus achieves its greatest concentration in the contents of the pregnant uterus, the fetus, and the fetal membranes after birth (Radostits et al., 2000). In addition, vaginal discharge and to a lesser extent, farm areas contaminated by fecal matter of calves fed on contaminated milk could be considered as the main source of infection (Acha and Szyfres, 2003). Infected animals also shed
organisms in the milk. Therefore, raw milk or raw milk products of bovine origin are ready sources for infections in humans. There can be also accidental self-inoculation with live *Brucella* vaccine strains that result in the disease (Genene et al., 2009).

### 2.1.4.2. Mode of transmission and route of infection

The most common route of transmission is the gastrointestinal tract following ingestion of contaminated pasture, feed, fodder, or water. Moreover, cows customarily lick after birth, fetuses, and newborn calves, all of which may contain a large number of organisms and constitutes a very important source of infection. Bulls do not usually transmit the infection from infected cows to non-infected mechanically (Thrusfield, 1995). The use of infected bulls for AI constitutes an important risk since the infection can be spread to many herds (Acha & Szyfres, 2003). Humans are infected from drinking raw or un-pasteurized infected milk, from exposure to infected discharges or tissues (Robinson and Production, 2003).

### 2.2. Pathogenesis

*B. abortus* has predilection in the pregnant uterus, udder, testicle and accessory male sex glands, lymph nodes, joint capsule, and bursa. After the initial invasion of the body, localization occurs initially in the lymph nodes. *B. abortus* is phagocytized by macrophages and neutrophils in an effort by the host to eliminate the organism. However, once inside the phagocyte, *B. abortus* is able to survive and replicate. The phagocyte migrates via the lymphatic system to the draining lymph nodes where *brucella* infection causes cell lysis and eventual lymph node hemorrhage following exposure (Edmonds et al., 2001). Because of vascular injury, some of the bacteria enter the bloodstream and subsequent bacteremia occurs, which disseminates the pathogen throughout the body. If the infected animals are pregnant, *B. abortus* will colonize and replicate in high numbers in the chorionic trophoblast of the developing fetus. The resulting tissue necrosis of the fetal membrane follows the transmission of bacteria to the fetus. The net effect of chorionic and fetal colonization is abortion during the last trimester of pregnancy (Kushwaha et al., 2016).

The preferential localization to the reproductive tract of the pregnant animals is due to the presence of unknown factors in the gravid uterus. These are collectively referred to as allantoic fluid factors that would stimulate the growth of *Brucella*. Erythritol, four-carbon alcohol, is considered to be one of these factors (Edmonds et al., 2001) which are elevated in the placenta and fetal fluid from about the fifth month of gestation (Thomson and Tustin, 1994). The preferential replication of *Br. abortus* in the extra-placental site within trophoblasts of the chorioallantoic membrane results in rupture of the cells and ulceration of the fetal membrane. The damage to placental tissue together with fetal infection and fetal stress will induce maternal hormonal changes. As a result, abortion occurs principally in the last three months of pregnancy, the incubation period being inversely proportional to the stage of development of the fetus at the time of infection (Xavier et al., 2009).

### 2.3. Clinical Signs

#### 2.3.1. Clinical signs in animals

The incubation period varies between 14 and 120 days (Azad et al., 2003). Primary clinical manifestations of brucellosis among livestock are related to the reproductive tract. In highly susceptible non-vaccinated pregnant cattle, abortion after the 5th month of pregnancy is a cardinal feature of the disease (Radiotitis et al., 2000). Retention of placenta and metritis are common sequels to abortion. Females usually abort only once, presumably due to acquired immunity (Edmonds et al., 2001).

In cattle, *B abortus* causes abortions, stillbirths, and weak calves. The placenta may be retained
and lactation may be decreased. Epididymitis, orchitis, and testicular abscesses are sometimes seen in bulls (Mantur and Mangalji, 2004). Infertility occurs occasionally in both sexes, due to metritis or orchitis/epididymitis. Hygromas, particularly on the leg joints, is a common symptom in some tropical countries. Arthritis can develop after long-term infections. Systemic signs do not usually occur in uncomplicated infections, and deaths are rare except in the fetus or newborn. Infections in non-pregnant females are usually asymptomatic, but pregnant adult females infected with \textit{B. abortus} develop placentitis, which normally causes abortion between the fifth and ninth month of pregnancy. Even in the absence of abortion, there is heavy shedding of bacteria through the placenta, fetal fluids, and vaginal exudates (Aparicio, 2013).

![Fig. 1: Epididymitis in Bulls (a) and abortion in a cow (b)](image)

Source: (Edmonds et al., 2001).

![Fig. 2: Hygromas on leg joints](image)


### 2.3.2. Symptoms of human brucellosis

The most common symptoms of brucellosis include undulant fever in which the temperature can vary from 37.8°C in the morning to 40°C in the afternoon; night sweats and weakness. Common symptoms also include insomnia, anorexia, headache, constipation, sexual impotence, nervousness, encephalitis, arthritis, endocarditis, orchitis, and depression (Quinn et al., 2002). Spontaneous abortion is seen mostly in the first and second trimesters of pregnancy in pregnant women infected with \textit{Brucella}. Lack of appropriate therapy during the acute phases may result in localization of \textit{Brucella} in various tissues and organs and lead to sub-acute or chronic disease which is very hard to treat (Bosilkovski et al., 2007).

### 2.4. Diagnosis

The diagnosis of brucellosis always requires laboratory confirmation. It is made possible by direct
demonstration of the causal organism using microscopic examination, culture methods, animal inoculation (identification of the agent), direct demonstration of antibodies using serological techniques, and molecular methods (Sathyanarayanan et al., 2011).

2.4.1. Microscopic examination and culture methods
Specimen of fetal stomach, lung, liver, placenta, cotyledon, and vaginal discharges are stained with Gram stain and modified Ziehl Nelson stains. Brucella appears as a small red-colored, coccobacillus in clumps. Blood or bone marrow samples can be taken cultured in 5-10% blood agar is used. To check up bacterial and fungal contamination; Brucella selective media are often used (Bax et al., 2007). The selective media are nutritive media, blood agar-based with 5% seronegative equine or bovine serum. On primary isolation, it usually requires the addition of 5-10% carbon dioxide and takes 3-5 days incubation at 37°C for visible colonies to appear (Gall et al., 2003).

2.4.2. Animal inoculation
Lab animals such as guinea pigs are intramuscularly inoculated 0.5-1ml of suspected tissue homogenate and sacrificed at three and six weeks post-inoculation and serum is taken along with the spleen and other abnormal tissue for serology and bacteriological examination (Pappas et al., 2006).

2.4.3. Serological diagnosis
Body fluid such as a serum, uterine discharge, vaginal mucus, and milk or semen plasma from suspected cattle may contain different quantities of antibodies of the IgM, IgG1, IgG2, and IgA types directed against Brucella (Corbel, 2006).

2.4.3.1. Milk ring test
It is cheap, easy, simple, and quick to perform. It detects lacteal anti-Brucella IgM and fat globules from milk and forms a red ring in a positive case. However, it tests false positive when milk that contains colostrums, milk at the end of the lactation period, milk from cows suffering from abnormal disorder or mastitis. Milk that contains a low concentration of lacteal IgM, IgA, or lacks the fat clustering factors, tests false negative. Because lacteal antibodies rapidly decline after abortion or parturition, the reliability of milk ring test using 1ml milk to detect Brucella antibodies in individual cattle or intact milk is strongly reduced (Nielson et al., 2001). Although the milk ring test was performed with 8ml milk, it improved the detection of brucellosis in the tank milk. It may test false positives when races of colostrums are present in tank milk (Abubakar et al., 2012).

2.4.3.2. Rose Bengal Plate Test (RBPT)
It is a spot agglutination technique. It does need special laboratory facilities and is simple and easy to perform. It is used to screen sera for Brucella antibodies. The test detects specific antibodies of the IgM and IgG type. Although the low PH (3.6) of the antigen enhances the specificity of the test and temperature of the antigen and the ambient temperature at which the reaction takes place may influence the sensitivity and specificity of the test (Australian Veterinary Emergency Plan, 2005).

2.4.3.3. Complement Fixation Test (CFT)
The CFT test is highly specific but it requires highly trained personnel as well as suitable laboratory facilities. It measures more antibodies of the IgG1 type than antibodies of the IgM type. It is the most reliable diagnostic test now in routine use for individual animals. It is relatively insensitive to
antibodies resulting from strain 19 immunizations (vaccinations). The CFT is widely used and accepted as a confirmatory test although it is complex to perform, requiring good laboratory facilities and adequately trained staff to accurately titrate and maintain the reagents. There are numerous variations of the CFT in use, but this test is most conveniently carried out in a microtiter format (Nielson et al., 2001). Either warm or cold fixation may be used for the incubation of serum, antigen, and complement: either 37°C for 30 minutes or 4°C for 14–18 hours. A number of factors affect the choice of the method: anti-complementary activity in serum samples of poor quality is more evident with cold fixation, while fixation at 37°C increases the frequency and intensity of prozones, and a number of dilutions must be tested for each sample (Xavier et al., 2009).

2.4.3.4. ELISA test
Indirect enzyme immunoassay (ELISA) is the serological test group that is used to determine the prevalence of brucellosis in surveys. It is a test that offers excellent sensitivity and specificity with a minimum of equipment and sources in kit form. It is more suitable than the complement fixation test for use in smaller laboratories and now it is used for the diagnosis of a wide range of animal and human diseases (Mantur and Mangalgi, 2004). Although in principle ELISAs can be used for the tests of serum from all species of animal and man, results may vary between laboratories depending on the exact methodology used. Not all standardization issues have yet been fully addressed. For screening, the test is generally carried out at a single dilution. It is also important to note that ELISAs are only marginally more specific than RBPT or CFT (Alehegn, 2015).

2.4.4. Molecular methods

2.4.4.1. Polymerase Chain Reaction
Polymerase chain reaction (PCR) assays can be used to detect Brucella DNA in pure cultures and in clinical specimens, i.e. serum, whole-blood and urine samples, various tissues, cerebrospinal, synovial, or pleural fluid, and pus (Colmenero et al., 2010). Direct detection of Brucella DNA in brucellosis patients is a challenge because of the small number of bacteria present in clinical samples and inhibitory effects arising from matrix components. Basic sample preparation methods should minimize inhibitory effects and concentrate on the bacterial DNA template (Queipo-Ortuno et al., 2008).

2.5. Differential Diagnosis
There are many potential causes of abortion in cattle. Infectious causes of abortion include viral diseases such as Infectious Bovine Rhinotracheitis, Rift Valley fever; and infections with other organisms such as Trichomonas fetus, Neospora caninum, Campylobacter fetus, Listeria monocytogenes, various Leptospira species (Poester et al., 2013).

2.6. Post Mortem Findings

2.6.1. Gross Findings
In cows, the main sites of infection are the endometrium of the uterus and the fetal placenta. The uterus appears normal externally but the endometrium is invariably infected. The intercotyledonary areas of the placenta are generally thickened with yellow gelatinized fluid and may be ulcerated, appear like leather, and have mucoid or fibrino-purulent deposits on the surface. Placental cotyledons are hyperemic and may have areas of yellow–grey necrosis and be covered with sticky brown exudates (Mahajan, 2013). The uterus of infected cows is characterized by brownish fluid, with exudate consistent with a necrotizing placentitis and the uterus can also show fibrinous necrotic exudates and multifocal hemorrhages (Luzzi et al., 1993).
The fetus is usually swollen, with blood-tinged fluid found subcutaneously and in the body cavities; the umbilical cord may be thickened and swollen (Poester et al., 2013). Other lesions include fibrinous pleuritis and peritonitis, bronchopneumonia, and splenitis (Solera, 1995). Fibrinous pericarditis has been described as a significant fetal lesion in brucellosis (Mahajan, 2013).

Fig. 3: Uterus from a *Brucella abortus* infected cow immediately after abortion shows several necrotizing and hemorrhagic acute placentitis.  

Fig. 4: *Brucella abortus* infected aborted fetus with an acute diffuse severe fibrinous pleuritis.  

In Bulls, *B. abortus* causes infection and swelling of the testicles that may not be obvious, but increasing pressure results in necrotic foci that grow and coalesce and may lead to total testicular necrosis with sequestration by inflammatory thickening of tunica. *B. abortus* may also infect the accessory sex glands. *Brucella* in cattle may localize in the carpal and other bursae where hygromas containing large numbers of bacteria may be found (Poester et al., 2013).

2.6.2. Microscopic findings

In Cows, when examined microscopically, the membranes and cotyledons contain many mononuclear cells with some neutrophils and the chorionic epithelial cells are packed with the bacteria. Anabnormally firm attachment of the chorionic villi of the placenta results from necrosis and enlargement of the maternal villi and the presence of inflammatory exudates (Poester et al., 2013).

Necrotic neutrophilic placentitis with perivascular infiltrates is the most frequent microscopic change in experimentally infected cows and inflammation is associated with large numbers of *B. abortus* cells inside macrophages and trophoblasts (Mahajan, 2013).
2.7. Treatment
Effective treatment for animals with brucellosis is not known to date (Weidmann, 1991). The treatment of brucellosis in the cow has generally been unsuccessful because of the intracellular sequestration of the organisms in lymph nodes, the mammary gland, and reproductive organs and the bacteria are facultative intracellular which survive and multiply within the cells (Radostits et al., 2000). Generally, treatment of infected livestock is not attempted because of the high treatment failure rate, cost, and potential problems related to maintaining infected animals in the face of ongoing eradication programs (Edmonds et al., 2001). Man can be treated with antibiotics (doxycycline with rifampicin), however, relapses are impossible (Smits and Kadri, 2005).

2.8. Prevention and Control
Prevention, control, and eradication of brucellosis are a major challenge for public health programs. Although controlled or eradicated in a number of developed countries, re-introduction of brucellosis remains a constant threat, while in others, especially in the developing world, this disease continues to exert its devastating impact perpetuating poverty. The strategies for preventing brucellosis have to be adapted to the animal production system (Seifert, 1996). The successful prevention of this disease, which is so difficult in cattle production in the tropics, requires that, as far as possible, all available steps are taken to combat it (Weidmann, 1991).

2.8.1. Vaccination
The WHO has long been involved in brucellosis surveillance and control, including research and development of vaccines to prevent animal brucellosis (Munir et al., 2010). Systematic vaccination of animals is recommended where the prevalence is greater than 5% (Holveic et al., 2007). The vaccine increases individual resistance to systemic infection, and in infected animals decreases the probability of placental infection, abortion, and massive shedding of infectious organisms (Ibrahim, 2010).

The following are some of the vaccines available against brucellosis:

**Brucella abortus S19 Vaccine:** The most widely used vaccine for the prevention of brucellosis in cattle is the *Brucella abortus* S19 vaccine, which remains the reference vaccine to which any other vaccines are compared. It is used as a live vaccine and is normally given to female calves aged between 3 and 6 months as a single subcutaneous dose of $5–8 \times 10$ viable organisms. A reduced dose of organisms can be administered subcutaneously to adult cattle, but some animals will develop persistent antibody titers and may abort and excrete the vaccine strain in the milk. Alternatively, it can be administered to cattle of any age as either
one or two doses of 5 × 10 viable organisms, given by the conjunctival route; this produces protection without a persistent antibody response and reduces the risks of abortion and excretion in milk when vaccinating adult cattle (Mantur and Mangalgi, 2004). 

*Brucella abortus* S19 vaccine induces good immunity to moderate challenge by virulent organisms. The vaccine must be prepared from USDA-derived seed and each batch must be checked for purity (absence of extraneous microorganisms), viability (live bacteria per dose), and smoothness (determination of dissociation phase). Seed lots for S19 vaccine production should be regularly tested for residual virulence and immunogenicity in mice (Mantur and Mangalgi, 2004).

**Brucella abortus strain RB51 vaccines:** This is a recently developed vaccine and has replaced *Br. abortus* strain 19 in a number of countries as the approved calf hood vaccine because it does not interfere with serological evaluation (Edmonds et al., 2001). *Brucella abortus* strain RB51 is a live stable rough mutant of *Br. abortus* strain 2308, which lacks much of the lipopolysaccharide O-side chain and has been investigated as an alternative to strain 19 vaccines (Radostits et al., 2000). Adult vaccinations with *Br. abortus* strain RB51 only rarely causes abortion. One way to reduce the side effects of RB51 is to reduce the dose. When using the reduced dose of this vaccine (1 × 10 colony-forming units [CFU]), on late pregnant cattle, no abortions or placentitis lesions are produced.

### 2.8.2 Test and slaughter

It involves recognition of all animals which have responded immunologically to a *Brucella* infection and subsequent culling of the reactors. According to Weidmann (1991) these methods could be achieved when the rate of infection is reduced to an acceptable level (about 1-2%). Part of the scheme has to be careful control of all animals which will be newly added to the herd as well as a production system that prevents contact with infected neighboring farms and/or contaminated feed or pasture.

#### 2.8.3 Pasteurization

The most rational approach for preventing human brucellosis is the control and eradication of the infection in animal reservoirs. *B. abortus* is inactivated by pasteurization and its survival outside the host is largely dependent on environmental conditions. The pathogen may survive in the 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 feces, and eight months in liquid manure tanks. Bacterial survival is prolonged at low temperatures and organisms will remain viable for many years in the frozen carcass (Maudilin et al., 2009). Pasteurization of dairy products is an important safety measure where this disease is endemic. Unpasteurized dairy products and raw or undercooked animal products (including bone marrow) should not be consumed (Abbas and Aldeewan, 2009).

#### 2.8.4 Hygienic Prophylaxis

Experience shows that vaccination alone cannot bring about the eradication of the disease (Weidmann, 1991). From the epidemiology of the disease, important steps were derived at an early stage as hygienic prophylactic measures. These include:

- The isolation of calving animals in separate calving pens which are subsequently disinfected with 2.5% formalin (Weidmann, 1991; Thomson and Tustin, 1994).
- Wet and well-grassed calving camps should be avoided, and vehicles used for transporting infected animals should be disinfected after use (Thomson and Tustin, 1994).
- Aborted fetuses, placentas, and uterine discharges must be disposed of, preferably by incineration (Weidmann, 1991; Thomson and Tustin, 1994; Radostits et al., 2000).
All cattle, horses, and pigs brought to the farm should be tested, isolated for 30 days, and retested (Radostits et al., 2000).

Cows, which are in advanced pregnancy, should be kept in isolation until after parturition, since occasionally infected cows may not show a positive serum reaction until after calving or abortion (Thomson and Tustin, 1994; Radostits et al., 2000).

Replacement stock should be purchased from a herd free of brucellosis (Thomson and Tustin, 1994).

Chlorhexidine gluconate is an effective antiseptic against Br. abortus and it is recommended for washing the arms and hands of animals attendants and veterinarians who are exposed to contaminated tissues and materials (Weidmann, 1991; Thomson and Tustin, 1994; Radostits et al., 2000).

3. Significance of the Disease
3.1. Economic Significance

Endemic brucellosis in low-income countries of sub-Saharan Africa and South Asia has multiple economic implications across agriculture and public health and broader socio-economic development sectors. Efforts to control the disease in low-income countries must take a different approach. Simply replicating past successes in brucellosis control and eradication in high-income countries will not work. Low-income countries have at least a ten-fold higher burden of infectious disease from a wide variety of pathogens (Mc Dermott et al., 1987). The assessment of the economic aspects of brucellosis, with emphasis on the low-income countries of Africa and Asia, is structured in three main parts. The first describes an overall framework for the economic assessment of disease burdens and the impacts of potential control programs. The second part systematically reviews available animal, human, and joint burden estimates from studies conducted in these regions. The third section provides estimates, when available, of different costs associated with brucellosis illness and its control. This section also comments on tools and approaches for assessing control programs that are of relevance to low and middle-income (Zamri-saad and Kamarudin, 2016). When brucellosis is detected in a herd, flock, region, or country, international veterinary regulations impose restrictions on animal movements and trade, which results in huge economic losses. The economic losses as well as its zoonotic importance are the reasons why programs to control or eradicate brucellosis in cattle (OIE, 2008).

In Ethiopia, information on losses specifically through brucellosis in the different types of production systems is sparse, except for Tariku (1994) who reported an annual loss from brucellosis estimated to be 88,941.96 Ethiopian Birr ($5231 equivalent) among 193 cattle, largely due to reduced milk production and abortions.

3.2. Public Health Significance

*Brucella abortus*, *B. melitensis* and *B. suis* are highly pathogenic for humans (Abubakar et al., 2012). Brucellosis remains the most common zoonotic disease in the world, with more than 500,000 new cases reported annually (Godfroid et al., 2013); the actual number of cases, including undetected and unreported cases, is believed to be considerably higher (Al Dahouk et al., 2013). Brucellosis is often a neglected disease despite being endemic with high zoonotic potential in many countries (Poester et al., 2013). The prevalence of human brucellosis differs between areas and has been reported to vary with standards of personal and environmental hygiene, animal husbandry practices, and species of the causative agent, and local methods of food processing (Chugh, 2008).

The Brucellosis 2003 International Research Conference estimated that 500,000 human infections occur per year worldwide, with incidences ranging from less than one case per 100,000 population in the UK, the USA, and Australia, through 20 to 30 cases per 100,000 in southern

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European countries such as Greece and Spain, to more than 70 cases per 100,000 in the Middle Eastern States such as Kuwait and Saudi Arabia. The majority of reported human brucellosis cases are caused by *B. melitensis*, *B. abortus*, and *B. suis*, in occurrence order, novel and atypical *Brucella* are also being investigated (Al Dahouk et al., 2013).

As compared to the study of animal brucellosis, the study of human brucellosis in Ethiopia is sparse with even less information on risk factors for human infection. For instance, out of 56 cases with a fever of unknown origin, two (3.6%) were reported to be positive for *B. abortus* antibodies by RBPT and CFT (Jergafa et al., 2009). A study conducted in traditional pastoral communities by Ragassa and others (Pal et al., 2017) using *B. abortus* antigen revealed that 34.1% of patients with febrile illness from Borena, 29.4% patients from Hammer, and 3% patients from Metema areas were tested positive using Brucella IgM/IgG lateral flow assay. Studies conducted in a high-risk group such as farmers, veterinary professionals, meat inspectors, and artificial insemination technicians in Amhara Regional State (Bifo et al., 2020), Sidama Zone of Southern People Nations and Nationalities State (Degefu et al., 2011), and Addis Ababa (Degefu et al., 2011) found a seroprevalence of 5.30%, 3.78%, and 4.8% by screening sera from 238, 38 and 336 individuals respectively. The discrepancy between and others might be due to the difference in milk consumption habits and the sensitivity of test methods used (Ferede et al., 2011).

Humans may become infected by ingestion of unpasteurized cheese or milk, by direct transmission through contact with infected animals, or by handling specimens containing *Brucella* spp. in the laboratory. It is also transmitted to humans by the consumption of raw dairy products and by direct contact with the skin or mucosa during parturition and abortion (Ferede et al., 2011).

In South Sudan, a fraught with several potential risk factors could fuel the dissemination of brucellosis to livestock and humans (Lado et al., 2012). The traditional pastoralist’s practice of assembling several herds into cattle camps with close livestock-human interactions is one of the key milestones. Moreover, poor awareness is a risk milestone to the occurrence and perpetuation of brucellosis in livestock which could create human health hazards (Ibrahim, 1990). Further brucellosis risk indicators including the rampant animal herder’s practice of vulval blowing, to facilitate milk letdown during cow milking (figure 6), and the practice of direct udder-to-mouth consumption of raw milk (figure 8) could exacerbate human brucellosis. Drinking raw milk was significantly associated with brucellosis while drinking boiled milk was protective (Lado et al., 2012). Hence active public health education on the benefits of boiling milk before consumption is imperative.

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*Fig. 6: Blowing through the vulva to enhance milkletdown in the Terekeka*

Source: Lado et al. (2012).

*Fig. 7: Direct sucking of raw milk from cattle camps in Terekeka County.*

Source: Lado et al. (2012).
4. The Status of Brucellosis in Ethiopia

Ethiopia, located in Eastern Africa, is predominantly an agrarian country with over 85% of its population engaged in agricultural activity. The country has diverse agro-ecological zones, which have contributed to the evolution of different agricultural production systems. Animal husbandry forms an integral part of agricultural production in almost all ecological zones of the country (Haileselasie et al., 2010).

In Ethiopia, a number of works have been done on the sero-prevalence of brucellosis in different parts of the country. However, the economic impact of the disease on animal productivity and production is not yet assessed. So far, only one study has been made at Chaffa State farm, Wollo, from 1987 to 1993. The same paper indicated that there was an annual loss estimated to be 88,941.96 Eth. Birr due to reduced milk production and abortions in the farm on 193 study animals (Tolosa et al., 2007).

A serological investigation on the prevalence of brucellosis in Ethiopia has been carried out in different parts of the country for the last 31 years. Pioneer surveys were done by Meyer (1980) and reported a positive reaction of 39% out of 1010 cattle owned by the Institute of Agricultural Research (IAR). The survey conducted by Gebre-mariam (1985) on the prevalence of bovine brucellosis in four different farms around Addis Ababa showed that 18.4% were positive reactors out of 178 tested animals. In the Borena zone of the Oromia region, the highest seroprevalence (50%) was documented using ELISA (Alem and Solomon, 2002).

In Ethiopia, most research done on brucellosis has been focused on intensive dairy cattle herds in urban and peri-urban areas. The World Organization for Animal Health (OIE) reported a prevalence of 20%; the prevalence was higher around large towns than in rural areas (Dando et al., 2001). Since the first report of brucellosis in the 1970s in Ethiopia, the disease has been noted as one of the important livestock diseases in the country (Ibrahim et al., 2010). A large number of articles have been published reporting individual seroprevalence ranging from 1.1% to 22.6% in intensive management systems (Asmare et al., 2010) and 0.1–15.2% in the extensive management system (Dinka and Chala, 2009). In zebu cattle of the central highlands, a prevalence of 4.2% was reported (Bekele et al., 1989).

<table>
<thead>
<tr>
<th>S. No</th>
<th>Author</th>
<th>Year</th>
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5. Conclusion and Recommendation

Brucellosis is worldwide and has a high prevalence in different areas of Ethiopia. Brucellosis affects both animals and humans, has a very high economic and public health impact. Its impact on public health is very well related to the infected animal species from which human...
transmission occurs. The disease transmits from infected animals to human beings through several routes. It is a special hazard to occupational groups. It causes considerable losses in cattle as a result of abortion and a reduction in milk yield. Even though the disease is prevalent in Ethiopia, few reports in humans are available. This may be due to the absence of appropriate diagnostic facilities. Prevention, control, and eradication of brucellosis are a major challenge for public health programs. The strategies for preventing brucellosis have to be adapted to the animal production system. The successful prevention of this disease, which is so difficult in cattle production in the tropics, requires that, as far as possible, all available steps taken to combat it are Vaccination, Test and Slaughter, Pasteurization, and hygienic prophylaxis. Based on the above conclusion the following recommendations were forwarded:

- Public education on the transmission and source of infection of the disease needs to be undertaken.
- The necessary precautions should be taken to reduce occupational risks.
- Pasteurization of milk should be widely practiced to prevent human infection.
- Isolation of aborted animals and proper disposal of aborted fetuses and fetal membranes, preferably, by incineration.
- The isolation of calving animals in separate calving Pens
- Strict movement control of animals from one area to another in order to prevent the spread and transmission of the disease from infected cattle to the non-infected ones.

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