

Seroprevalence of Camel Brucellosis in Export Farm, Ethiopia

Teferi Benti Moti

National Animal Health Diagnostic and Investigation Center, Sebeta, Ethiopia

Email address:

teferibenti58@gmail.com

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Abstract: Brucellosis is a zoonotic illness that affects both domestic and wild animals and is carried by members of the genus *Brucella*. Camels have been diagnosed with Brucellosis in almost every camel-raising country in Africa and Asia. Although camels are not the principal hosts for *Brucella* organism, they are susceptible. Humans get infected with the disease by direct or indirect contact with diseased animals or their products. A cross-sectional study was undertaken in Era camel export farm from November to December 2016 to determine the prevalence rate of camel brucellosis and associated risk factors. To screen for *Brucella* antibodies, the Rose Bengal Plate Test (RBPT) was used, and the complement fixation test (CFT) was used to confirm positive reactor samples. The Chi-square method was used to evaluate the data. A total of 1850 sera samples were taken from Dromedary camels for a serological analysis. It's likely that the Rose Bengal test will create cross reactivity, a confirmatory test must be performed. In the camel export farm, the total seroprevalence rate was 1.4 percent, with 1.73 percent (n=32) being screened by the Rose Bengal plate test and 1.4 percent (n=26) being verified by the complement fixation test. Age has a significant impact on the seroprevalence rate of camel brucellosis. Camels older than 4 years had a seroprevalence rate of 1.6 percent (22/1400) whereas camels less than 4 years had a rate of 0.9 percent (4/450). There are a statistically significant difference in age groups ($p < 0.05$). According to this study, brucellosis in camels could be a serious and widespread disease. As a result, newly purchased camels were inspected and confined until they were healthy, positive reactor animals were culled, and public health awareness was enhanced.

Keywords: Brucellosis, Camel, CFT, Export Farm, RBPT, Seroprevalence, Zoonotic

1. Introduction

Brucellosis is the generic name used for the animal and human infections caused by some species of the genus *Brucella*, mainly *Brucella abortus*, *B. melitensis*, and *B. suis* [1]. The camel (*Camelus dromedaries*, one-humped camel) plays a major socio-economic role within the pastoral and agro-pastoralist in dry and semi-dry zones of Africa and Asia [2]. The worldwide domesticated camelids population (dromedaries and Bactrian) is about 28 million [3]. About 80% of the camel population inhabits Africa, with 60% within the Eastern African countries (Sudan, Ethiopia, Somalia, and Kenya) the exporters of dromedary camels to the peninsula and Egypt [3]. Camels contribute significantly to the source of revenue for agro-pastoralists and therefore the pastoralists living within the insubstantial environments of Africa and Asia as sources of milk, meat, transportation,

leather, and wool [2] and riding as a tourist attraction and valuable racing animals. Racing is incredibly popular within the Arabian Gulf regions and is amid active camel trading [4]. Camels are more drought tolerant than the other animate being [5].

Camels are one of the livestock resources in Ethiopia, with a population estimated to be 4.8 million [6]. This number ranks the country third in Africa next to Somalia and Sudan and fourth within the world. In Ethiopia, camels are reared in Oromia, Somali, and Afar Regional states by pastoralists and agro-pastoralists within the arid and semi-arid areas [7]. Female camels account for 75% of the herd, while male camels are usually sold prematurely for slaughter [8]. Ethiopia exported live animals to some African countries and the Middle East as and source of income and foreign currency. The Ethiopian Government has been encouraging the establishment of the private sector of several livestock

export farms to increase the country's export share of the fast-expanding marketplace for live animals and meat. Era camel export farm is among live camel export farms established and located within the East Arisi zone, Oromia.

Despite their high productive potential, camels perform poorly management within the pastoralist, scarcity of feeding, slow production, and disease are constraints to the upper productivity. Camels graze and browse freely mixed with other livestock. These may expose to various bacterial and viral infections are anticipated. Among these, Brucellosis will be cross transmitted and spread between animals.

In Ethiopia, brucellosis has been reported in camels by different authors' [9], 11.9% and 7.6%; [10], 5% and 4.1%; [11], 5.8% and 3.37%, [12], 9.2% and 9.1%; [13], 12.2% and 4.1% and [14] 3.6% and 3.1% by RBPT and CFT respectively. Different studies showed that *B. abortus* and *B. melitensis* are the most frequently isolated from milk, aborted fetus and vaginal swabs of diseased camels [15].

Although camels aren't known to be primary hosts of *Brucella*, they're vulnerable to *B. abortus*, *B. melitensis*, and *Brucella ovis* [16]. The transmission of brucellosis in camels depends on the *Brucella* species being prevalent in primary animals sharing their habitat and husbandry [17]. The disease can cause significant loss of productivity, long calving interval time, low herd fertility, and relatively low milk production. Camel brucellosis poses a barrier to the export and import of animals constraining livestock trade and is an obstacle to free animal movement [18]. Camel brucellosis has considerable public health importance as camel milk is usually consumed raw within the pastoral areas and has close contact with their animals. *B. melitensis* has highly zoonotic potential, followed by *B. abortus* and *B. suis*. Infected camels often exhibit an acute or persistent febrile illness with a diversity of clinical signs that make the diagnosis of camel brucellosis difficult [19].

Brucellosis in camels is not well studied as compared with other domesticated animal species. Camel can get an infection from cattle, sheep, goats, and other species. Camel is at greater risk of developing the diseases after contact with infected large and small ruminants [20]. However, there is currently little information available about the epidemiology of the disease in camel, and its occurrence poorly estimated in the pastoral area of Oromia Regional State. Therefore, the aim of the present study was to explore the prevalence rate and potential risk factors of camel brucellosis in the Era camel export farm.

2. Materials and Methods

2.1. Study Area and Study Animals

The study was conducted from November to December 2016 in Era camel export Farm to determine the prevalence of *Brucella* infection. Era is located in the Rift Valley of East Arisi zone, about 125 km southeast of Addis Ababa, with an elevation of 1600-1700 meters above sea level. The annual rainfall ranges from 400 to 800 mm. The temperature range

is 13.9 to 27.7°C [21]. The target animals of this study were male camels under an extensive production system. Camels reach maturity at 3 to 4 years of age [22].

2.2. Study Design and Sampling Method

A cross-sectional study was conducted, using serological tests Rose Bengal Plate (RBPT) and Complement Fixation test (CFT) on camel sera. Study animals were male camels above one year of age with no history of vaccination against brucellosis. The purposive sampling method was employed in which a total camel of 1850 was sampled. The origin of Camels was from three zones, East shoa, Borena, and Bale zone of Oromia Regional state.

2.3. Blood Sample Collection

Using ordinary Vacutainer tubes and a disposable needle, approximately 5 to 7 ml of blood was obtained from each animal's jugular vein. The ear tag number of each individual animal was used to label each sample, and prospective risk factor information was acquired.

To separate serum from blood clots, collected sera were stored at room temperature overnight. The serum was collected into sterilized cryovials, labeled, and transported to the National Animal Health Diagnostic and Investigation Center (NAHDIC) in an icebox with dry ice and stored at -20°C until it was tested.

2.4. Serological Tests

The Rose Bengal Plate Test (RBPT) was employed as a screening test for *Brucella* antibodies, with positive results confirmed by the Complement Fixation Test (CFT).

2.4.1. Rose Bengal Brucella Antigen

An antigen prepared from *B. abortus*, USDA strain 1119-3) and stained with Rose Bengal dye and suspended in acid buffer (pH 3.65) was used to detect *Brucella* antibodies in serum as per the method described by [23] using a ceramic agglutination plate. The reagents were Rose Bengal *Brucella* antigen (Cenogenics, USA), positive and negative controls were employed. It detects antibodies against *B. abortus*, *B. melitensis*, and *B. suis* in serum samples. Results of RBPT were interpreted as 0, +, ++ and +++ as described by [24]. No agglutination = 0; + = barely visible agglutination (seen by using magnifying glass); ++ = fine agglutination and +++ = coarse agglutination. The samples with an agglutination +, ++, and +++ was reported as positive, whereas samples with no agglutination (0) were recorded as negative.

2.4.2. Complement Fixation Test (CFT)

ID vet, 310. rue Louis pasture, France *Brucella* antigen was used. Complement fixation test is a suspension of *Brucella abortus* biovar one Weybridge strain no. 99 inactivated by heat and phenol. The reagent was used for the detection of antibodies against *B. abortus*, *melitensis* and *Suis* in ruminant, equidae, suidae, camelids and wild carnivores. The technique was applied according to the [1]. Before the

test, reagents such as complement, antigen, and haemolysin were titrated according to OIE guidelines, 2018. The sera samples were de-complemented before test at 56°C for 30 minutes in water bath to make free of complement. The Complement fixation test has two steps, in the first step antigen, test sera and complement are mixed and incubated. In the second step an indicator system which consists of sheep red blood cells (SRBC) and amboceptor or lytic antibody (that sensitizes RBC to the action of complement) is added and incubated.

The Sera reacted positively by RBPT were subjected to a confirmatory test with Complement Fixation Test [23]. The CFT result was interpreted as Sedimentation of Sheep Red Blood Cells (SRBC) at the bottom of U-shaped microplate indicates positive results, and complete hemolysis of Sheep Red Blood Cells (SRBC) indicates negative [1]. Complement with 100% strong fixation reaction, more than 75% fixation of complement at a dilution of 1:10 and at least with 50% fixation of complement at a working dilution (1:5) classified as positive. ID vet *Brucella* positive and negative control sera were employed for CFT.

2.5. Data Analysis

The data collected was coded, entered into a Microsoft Excel spreadsheet and analyzed using IBM SPSS statistics version 20.0. The seroprevalence was calculated as percentage by dividing the numbers of animals seropositive for brucellosis (positive using CFT) by the total number of camels sampled. The degree of association between each risk factor was assessed using the Chi-square (χ^2) test. For all analyses, 95% confidence intervals (CIs) were computed and p -value < 0.05 was taken as significant.

3. Results

Camel brucellosis was found to have a seroprevalence rate of 1.4 percent. By Rose Bengal Plate Test, 1.73 percent ($n=32$) of 1850 sera samples were seropositive for *Brucella* infection, and 1.4 percent ($n=26$) were positive by complement fixation test. Positive Rose Bengal Plate Test (RBPT) samples were subjected to a confirmatory test, as recommended by [23], and the confirmatory test revealed that 81.25 percent of the samples (26/32) were positive. This could be due to cross-reactions between *Brucella* and other bacteria with antigen determinants that are identical to *Brucella*'s in the Rose Bengal test [25]. There was a prevalence rate of 1.6 percent (22/1400) in animals older than 4 years and 0.9 percent (4/450) in young camels under 4 years. Because adults had higher seropositivity than young camels, it was concluded that adult camels had higher seropositivity than young camels, implying that the disease may be a disease of sexually developed animals.

4. Discussion

According to the Complement Fixation Test, the total seroprevalence rate of camel brucellosis was (1.4 percent).

This finding was nearly identical to that of a previous study [7, 26-28], which found 1.2 percent and 1.8 percent, in the Borena lowlands, 1.6 percent and 1.8 percent in Dire Dawa, and 1.8 percent in the Ethiopian Somali area, respectively. However, it was lower than the earlier seroprevalence rate recorded in Ethiopia by RBPT and CFT in camel-rearing regions (Afar, Somali, and Oromia) [10-14, 29, 31]. The disparity could be explained by the sample size, the fact that only male animals were collected, and the agroecology of the sampled area. Due to the lack of erythritol in male animals, they are less susceptible to *Brucella* infection [32].

Adult camels have a higher prevalence rate than young camels. This result was consistent with the [12, 14, 29, 33-35] findings that adult camels have a higher seroprevalence rate than juvenile camels, which is statistically significant. According to a meta-analysis, the prevalence was higher in post-pubertal animals than in pre-pubertal animals [36]. In contrast, [10, 13] found that there is no substantial difference in animal age categories. In this study, there was a significant difference in camel brucellosis seropositivity between age groups ($P=0.00$).

On the other hand, younger animals are more resistant to infection and can sometimes clear a long-term infection, despite the possibility of latent infections [37]. *Brucella* organisms are stimulated to grow and multiply by sex hormones and erythritol, which tend to increase in concentration with age and sexual maturity [20, 38].

Animal age could be an important epidemiological element that influences brucellosis seropositivity [27, 39].

5. Conclusion

The seroprevalence in the present study revealed that brucellosis is a widespread and established disease in the camel farm and age is one of the risk factor. Brucellosis is a contagious illness that is also a serious zoonotic threat around the world. Various *Brucella* species are capable of infecting nearly all animal species, resulting in significant economic damage. Camels are valuable sources of money, milk, meat, transportation, leather, and wool, as well as recreational animals [40]. Traditional management approaches, such as mixing herds with other animals and allowing camels to freely migrate, are considered to be important in preventing disease spread. The consumption of raw camel milk is common in pastoral areas, and the sickness has become endemic due to a lack of information about the disease. As a result of the above mentioned finding, camels may be screened for brucellosis before joining the farm, the disease's state in camels and humans assessed, and the *Brucella* species implicated identified. To reduce the disease's risk, public health awareness and a secure husbandry management system should be implemented on a constant basis.

Competing Interests

The authors declare that they have no competing interests.

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