



Sero-prevalence and Associated Risk Factors of Brucellosis in Camel at Akaki Abattoir, Central Ethiopia

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ABSTRACT

The present study was delineated to investigate the seroprevalence and risk factors of brucellosis in camels brought for slaughtering at Akaki abattoir, Ethiopia during the period between December 2015 and April, 2016. Serum samples from 201 apparently health camels were initially screened for the presence of Brucella antibodies by the Rose bengal Plate Test (RbPT) and positive reacting sera were further confirmed by the Complement Fixation Test (CFT). Out of total 201 samples tested for RBPT, 13 (6.5%) were positive for brucellosis, of these, 9(4.5%) were confirmed to be seropositive for brucellosis upon further testing by CFT. There was significant difference in seroprevalence (based on CFT) of camel brucellosis in different age groups ($\chi^2=34$, $p < 0.05$), sexes ($\chi^2= 6.3$, $p < 0.05$), and animals with difference body score condition ($\chi^2= 11.85$, $p < 0.05$). The seroprevalence was significantly higher in animals of 5-9 years age group, females, poor body condition when compared to animals with other age groups, male animals and animal with medium or good body score condition respectively. The results of the present investigation indicate that Brucella spp. exists within the camels in Borena, Oromia region. Coordinated nationwide epidemiological surveillance in camel and other ruminants is required together with typing of infecting strains, thus enabling the transmission dynamics to be elucidated and initiating immunization campaigns, public health education and eradication strategies.

Keywords: Camel brucellosis, Seroprevalence, Akaki abattoir, Ethiopia

The camel (*Camelus dromedarius*, one-humped camel) play an important socio-economic role within the pastoral and agricultural system in dry and semi dry zones of Asia and Africa (Gwida *et al.*, 2012). Camels are a subset of huge livestock resources in Ethiopia with the population estimated to 2.3 million (CSA, 2007). This number ranks the country third in Africa after Somalia and Sudan and fourth in the world. In Ethiopia camels are reared in arid and semiarid areas in Borena, Somali, and Afar regions by pastoralists and agro-pastoralists (Mohammed *et al.*, 2011). Camels in the Oromia region are mainly kept for milk and meat production and transportation system.

Despite its high productive potential, camels perform poorly in the pastoral herd. Poor management, inadequate nutrients, slow production, and disease appear to be major constraints to the higher productivity of camels. Due to

over-browsing shrubs and recurrent drought, camels graze and browse freely mixing with other livestock. As a result of this exposure of camels to contagious diseases and various endo- and ectoparasites, illnesses are expected to increase. Among the diseases that can possibly be cross transmitted between cattle, goat, sheep, and camel, is brucellosis.

In Ethiopia, brucellosis has been reported in camels by various workers (Zewold and Haileselassie, 2012; Hadush *et al.*, 2013; Gumi *et al.*, 2013). Although camels are not known to be primary hosts of Brucella, but they are susceptible to *B. abortus*, *B. melitensis* and *Brucella ovis* (Seifert, 1996). Consequently, the prevalence depends upon the infection rate in primary hosts being in contact with them (Musa *et al.*, 2008). The disease can generally cause significant loss of productivity through late first

calving age, long calving interval time, low herd fertility and comparatively low milk production in camels. The disease poses a barrier to export and import of animals constraining livestock trade and is an impediment to free animal movement (Zinsstag *et al.*, 2011). In addition to economic importance, camel brucellosis has considerable public health importance as camel milk is often consumed raw in Ethiopia. *B. melitensis* is considered to have the highest zoonotic potential, followed by *B. abortus*, and *B. suis*. Infected camel often exhibit mild clinical signs which makes diagnosis of camel brucellosis difficult (Wernery, 2014).

Limited information is available on camel brucellosis in Ethiopia especially in Borena lowland of Oromia region. Therefore the present study was undertaken to estimate the seroprevalence of camel brucellosis in Akaki Abattoir by using serological tests RBPT and CFT and to elucidate risk factors associated with it.

MATERIALS AND METHODS

Study area

The study was conducted at Akaki Abattoir from December 2015 to April 2016, which is located in the southern outskirts of capital city, Addis Ababa. The average number of camels slaughtered in the abattoir was seven per day and 2500 per annum. Camel meat is not popular among residents in Addis Ababa but consumed mainly by Somali and other Muslim communities dwelling in the city. Camels brought for slaughtering at the Akaki Abattoir originated from Borena areas of Ethiopia. Borena is located in the Oromia National Regional State, about 500 km South of Addis Ababa. The climate of the Borena zone is semi-arid. According to regional estimates, the camel population in Oromia is 139,830 which represent 30.6% of Ethiopia's total camel population. Borena, the origin of study animals is situated at 600 km south of Addis Ababa on altitudes ranging from 500 to 2500 meters above sea level. The climate of Borena is semi arid. It has an annual rainfall of 450-650 mm in bimodal pattern with long rainy season between March and May and the short rainy season between October and November. The mean annual temperature varies from 19°C to 25°C with moderate seasonal variation (NMSA, 2003). Pastoralism

and agropastoralism are the two major livelihood ways practiced in the region.

Study animals

The study animals were apparently healthy camels brought for slaughtering at Akaki Abattoir during the study period. A total of 201 camels in the Abattoir were classified into three age groups; young age group (1-4 years old), adult age group (5-9 years old) and the older age group (10-15 years old (Abebe *et al.*, 2002). The camels slaughtered at the abattoir were transported from their areas of origin to the Akaki Abattoir on trucks and kept at the lairage for 1 to 7 days.

Study Design

A cross-sectional study was conducted at Akaki Abattoir from November 2015 to April 2016 to determine the seroprevalence of camel brucellosis. Census sampling method was employed. During sample collection, all necessary risk factors related to camel brucellosis were properly taken such as age, sex, and body condition.

Sample Size Determination

The sample size of the study animals were determined by using the formula given for census sampling methods (Thrusfield, 2005) by using an expected prevalence (Bekele, 2004)

$$n = 1.962 [pexp (1-pexp)]/d^2$$

Where,

n = required sample size;

p = expected prevalence (p = 1.8% = 0.018);

d = 5% = 0.05, 1.96 (CI = 95%).

Thus, the desired sample size for p = 0.018 will be n = 27. However, 201 camels were included in the study to increase accuracy, representativeness and randomness in the study animals.

Blood Collection and Serological tests

Blood samples (5 to 8ml) were obtained by jugular venupuncture using plain vacutainer test tubes from

properly restrained animals and were stored in an ice box. The blood samples were allowed to clot in a slanting position, then transported to the laboratory in a leak-proof container with ice packs. They were centrifuged at 1000 rpm for 5 mins. Sera were then decanted into 5 ml plastic tubes and stored in the refrigerator at -20°C until further processing took place.

The prevalence of brucellosis was determined by Rose Bengal Plate Test (RBPT) accordingly to standard procedures developed by Nilson and Dukan (1990) using *B. abortus* antigen (BIO-RAD, Marnes-la-Coquette, France) Institute puravier 326, Rue de la Galera 34097 montpellier cedex 5, France) as a screening test. All the sera samples were further confirmed by complement Fixation Test (cFT) (OIE, 2004) using *B. Abortus* antigen S99 (cVL, New Haw Wey bridge, and Surry KT 15 3Nb, UK), control sera and complement (bgvv, berlin, Germany), and 2% sheep RBC, prepared by the National Veterinary Institute, Ethiopia, were used in the study.

Data Analysis

Data obtained from serological tests was stored in Microsoft excel spreadsheet program. Descriptive and analytic statistics were computed using software SPSS Version 20.0. Logistic regression and Chi-square test (χ^2) were employed to identify possible risk factors associated with seropositive camel. The relationship of associated risk factors with positive serological test was computed using odds ratio (OR) signified by 95% confidence intervals (Thrusfield, 2005).

RESULTS AND DISCUSSION

The sero-prevalence and risk factors of camel brucellosis in the Akaki Kality abattoir based on RBPT and CFT are presented in Table 1 and Table 2. Out of total 201 samples tested for RBPT, 13 (6.5%) were positive for brucellosis. Of these, 9(4.5%) were confirmed to be seropositive for brucellosis upon further testing by CFT.

The logistic analysis of putative risk factors indicated that there was significant difference in seroprevalence (based on RBPT) of camel brucellosis in different age groups ($\chi^2=34$, $p<0.05$), sexes ($\chi^2=6.3$, $p<0.05$), and animals with difference body score condition ($\chi^2=11.85$, $p<0.05$). The seroprevalence was significantly higher in animals of 5-9 years age group, females, poor body condition when compared to animals with other age groups, male animals and animal with medium or good body score condition respectively. Further analysis of the different risk factors using CFT is indicated in Table 2. Similar association of risk factors were observed for CFT positive results.

Brucellosis is considered as one of great public health problem all over the world (Radostits *et al.*, 2007) with more than 500,000 human cases reported annually (Pappas *et al.*, 2006). The bacterial agent of brucellosis is classified by the CDC (2007) as a category (b) pathogen that has a potential for the development as a bio-weapon. Although Camels are highly susceptible to brucellosis caused by *Brucella abortus* and *Brucella melitensis*, little attention has been paid to this disease in camels as it provokes only few clinical signs in contrast to its clinical course in cattle (Mousa *et al.*, 1987)

Table 1: The association of assumed risk factors with dependent RBPT Brucella seropositivity in camel

Variables		No of tested animal	No of positive animal	Prevalence (%)	χ^2 Value	P-value	95% CI	OR
Age (Years)	1-4	104	0	0	37.08	0.00		
	5-9	34	10	29.4				
	10-15	63	3	4.8				
Sex	Female	118	12	10.2	6.5	0.011	1.18-72.85	9.28
	Male	83	1	1.2				
Body condition	Good	67	0	0	10.05	0.007		
	Medium	45	2	4.4				
	Poor	89	11	12.4				

Table 2: The association of assumed risk factors with dependent CFT Brucella seropositivity in camels

Variables		No of tested animal	No of positive animal	Prevalence (%)	χ^2 Value	P-value
Age (Years)	1-4	104	0	0	34.95	0.00
	5-9	34	8	23.5		
	10-15	63	1	1.6		
Sex	Female	118	12	7.6	6.3	0.01
	Male	83	1	0		
Body condition	Good	67	0	0	11.85	0.003
	Medium	45	2	0		
	Poor	89	11	10.1		

The camel brought to Akaki Abattior for slaughter purpose usually belongs to Borena low land of Oromia region. Borena lowland peoples keep camels primarily for milk production, transportation, and meat production (Demekie, 1998; Coppock, 1994). In the present study, all 201 camels were clinically normal at the time of sampling and according to the owners, none had previously shown clinical signs of brucellosis. The 4.5% seroprevalence of brucellosis in apparently healthy camels in the present study indicates that many infected camels might be silent carriers for brucellosis and their products may pose a serious health problem for consumers. This finding is in agreement with the results (4.2%) of earlier reports in same area (Teshome *et al.*, 2003). However the findings were higher than most of the previous reports in Ethiopia. For instance Bekele (2004), Gumi *et al.* (2013) and Gassese *et al.* (2014) reported a prevalence of camel brucellosis as 0.4-2.5%, 0.9% and 0.53% respectively in Borena, Oromia region. The seroprevalence result of the present study is lower than many of the earlier reports in Ethiopia [1.7% in Tigray (Bekele, 2004), 7.6 % in Afar (Zewold and Haileselassie, 2012)] and other neighbouring countries [2.0 to 15.4% was reported in Kenya (Wanjohi *et al.*, 2012), 19.4% in Jordan (Dawood, 2008), 30.5% in Sudan (Ahmed *et al.*, 2007), 7.61% in Egypt (Hassanain and Ahmed, 2012)].

Differences in seroprevalence of camel brucellosis observed by various researchers might be due to differences in herd size, sample size, agro ecological and management conditions, and the presence or absence of infectious foci, such as Brucella-infected herds, which

could spread the disease among contact herds. Moreover lower prevalence rates reported earlier could also be the results of tests with low diagnostic sensitivity (Baumann and Zessin, 1992). Furthermore cross reacting bacteria such as *Escherichia coli*, *Yersinia enterocolitica* and Salmonella serotypes (Garin-Bastuji *et al.*, 1999) have potential to affect serological findings when tests of low specificity are used. In camels there are yet no standard set for the diagnostic test protocol and diagnostic titre for brucellosis. (OIE, 2000) recommends the test procedure outlined for the diagnosis of bovine brucellosis to be applied for camels. Accordingly, RBPT is considered as satisfactory screening test (OIE, 2000; Quinn *et al.*, 2002). The highest specificity of CFT deserved it to be used as confirmatory test in serial testing (OIE, 2000). Therefore, the use of serial testing procedure initially screened all samples by RBPT, and then applying CFT on positive reactors as employed in the current test improves the efficiency of detecting brucellosis (Teshome *et al.*, 2003).

In the present study, seroprevalence of brucellosis was significantly higher ($\chi^2 = 6.3$, $p < 0.05$) in females than males which concurs with the findings of Junaidu *et al.* (2006), Maiti and Mohan (2013) and Mohamed *et al.* (2013). Higher prevalence of brucellosis in female camels may be associated to erythritol which stimulates the growth of *B. abortus* (Gyles and Prescott, 2004). Also relaxation of immunity in females is attributed to lactation, pregnancy and other reproductive stress which contribute to higher prevalence in female camels (Gyles and Prescott, 2004). Female animals also play an important role in disseminating the disease via uterine discharge and milk.

The role of males in the spread of disease under natural mating is not important (Radostits *et al.*, 2007).

A significantly higher prevalence ($\chi^2=34$, $p< 0.05$) was also observed in adult camels (23.5%) than in young camels (0%) or old camels (1.6%). This is in agreement with Dawood (2008) and Sisay and Mekonnen (2012) who reported a higher prevalence of brucellosis in adult than in young camels in southern province of Jordan, and Afar region in Ethiopia respectively. Young animals tend to be more resistant to infection and frequently clear infections although few latent infections may occur (Walker, 1999). The presence of growth factors (erythritol and hormones) favour infections in sexually mature animals (Quinn *et al.*, 2002).

Nutrition plays a great role in Immunity against various infectious diseases. Underfed animals are expected to have a decreased immunity that is manifested by poor body condition (Faye and Bengoumi, 2006; Radostits *et al.*, 2007). Therefore, body condition of the camels was considered during the study to see the distribution of the infection in different body condition scores. Accordingly, significantly higher seropositivity was observed in camels with poor body condition score than camels with medium or good body condition score ($P>0.05$). Similar findings were observed by Swai *et al.* (2011) in Tanzania.

CONCLUSION

The 4.5% seroprevalence of brucellosis in apparently healthy male camels in abattoir indicates that camels serve as permanent carriers of brucellosis and could be source of infection for humans. The existing scenario of brucellosis in camels of the study area calls for urgent capacity building of regional laboratories. Co-ordinated nationwide epidemiological surveillance is required together with typing of infecting strains, thus enabling the transmission dynamics to be elucidated and initiating immunization campaigns, public health education and eradication strategies. That will be possible only by including camels in the national program for control and eradication of brucellosis in Ethiopia.

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