



SEROPREVALENCE STUDY OF BOVINE BRUCELLOSIS IN ASSELA GOVERNMENT DAIRY FARM OF OROMIA REGIONAL STATE, ETHIOPIA

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ABSTRACT

A cross sectional study was conducted to determine the sera-prevalence of bovine brucellosis and to assess the risk factors that contribute for its occurrence in Assela government dairy farm. The study animals were comprised indigenous (Aris, Borena) and cross bred. All eligible animals in the farm, which were above 6 months of age were sampled and the serum collected were first screened by Rose Bengale Plate (RBP) test and all reactor sera were further tested by Complement Fixation Test (CFT). Information concerning age, sex, parity, breed and origin of the animals were gathered from record book in the farm. Moreover, information on abortion, retained placenta, still birth and the overall husbandry system of the farm were gathered. Out of the 304 serum samples collected to 43 animals become positive with RBP test and the reactors were further subjected to CFT for confirmation and all were found to be positive with an overall prevalence of 14.14% (N=43). The result of chi square analysis showed that there was significant variation in the prevalence of the disease between the two ages (or=7.3, and $P < 0.001$). The result also indicated that there was significant difference in the sera positivity of brucellosis between the two sexes with 16.94% prevalence of the disease in female (41) and 3.2% in male animals (n=2) (5.3, and $P < 0.01$). The present study also revealed sera prevalence of 23.6% in the local cattle and 11.2% in cross bred once that were kept in the dry farm. Chi square analysis should that the difference in prevalence between the two breeds was highly statically significant with 2.1 and P value of 0.008. Parity was also identified as one risk factor in the farm in which significantly higher prevalence of brucellosis was observed in cows that gave birth more than once (28.6%) than first calf heifers (0%) (or=5.4 and P value =0.000). Furthermore there was statically significant association between sera positivity to brucellosis and history of previous abortion ($p < 0.001$) and retained fetal membrane and still birth ($p < 0.05$). In conclusion, all the risk factors assessed in the farm were found to be significant in the distribution and occurrence of the diseases. Control measures are also recommended.

KEYWORD:- Bovine, Brucellosis, Cattle, RBPT, CFT, Assela, Intensive farm.

INTRODUCTION

As one of the developing countries, Ethiopia depends on natural resources for its economic and social stability. Among these resources, livestock is the primary agricultural economic sector to which the farmers live is directly or indirectly interrelated (Fukadu, 1999). The country, Ethiopia has an enormous livestock resources with the total contribution of 15% of gross domestic product and 33% of the agricultural output. Current estimates show that there are 41.5 million heads of cattle, 28.2 million sheep and goats, 5.8 million equine species, 1 million camels and over 42 million poultry. The population of dogs and cats not known; however, each houses hold in the rural area owns one or two dogs and owns a cat. There are also an estimated 4.6 million house honey bee colonies contributing to the livelihood the rural population. The contribution of finishing at least 12 in land lakes and dams and many other rivers is also large. Recent advances in contribution of small ponds and hydroelectric dams are giving another opportunity for fish farming (DADA, 2006). However, there is no substation progress in livestock production due to various reasons. The most common constraints are poor animal husbandry

practice, low genetic potential of the indigenous breed, and above all the prevalence of different diseases in the entire country. The presences of these diseases make the livestock industry less efficient, especially diseases having negative impact on livestock export trade (Fukadu, 1999). In developed countries, animal trade is ruled by strong veterinary regulation and do not allow importing any animal species from the country having transmissible diseases or from unknown diseases status countries. In most Africa and Asian countries, the regulation is relatively low or totally absent as a result, the incursion of exotic diseases is high in these countries. Therefore, to use the huge livestock resource by reducing the impact of major animal diseases having economic and zoonotic importance such as brucellosis, tuberculosis, PPR and etc, surveillance is essential. In this study, brucellosis is taken as important problems to be investigated (Fukadu, 1999). Bovine Brucellosis is a major zoonotic disease widely distributed in both humans and animals especially in the developing world (WHO, 1997). Bovine Brucellosis is a disease of cattle usually caused by *Brucella abortus* less frequently by *Brucella melitensis* and rarely by *Brucella suis* (Radostits et al., 1994). It was first isolated and identified

in 1897, from bovine fetus and fetal membranes by Danish veterinarian, Fredrick Bang. The infection of cattle caused by the organ since then has been known as "bang's disease" or bang's abortion disease (Williams and Wikins, 1997).

Bovine Brucellosis is primarily a reproductive disease characterized by a abortion last trimester or birth of unthrifty newborn in the female and orchitis and epididymitis with frequent sterility in male (Radostits *et al.*, 1994) means of transmission in both female and male is through ingestion and direct or indirect contact, environment contamination by fetus, fetal membrane vaginal secretion like pasture forage, water, stable etc while the means by which man get infected are through indirect contact of materials contaminated by fetus, fetal membrane, vaginal secretion and directly through ingestion of raw milk and fresh cheeses (PAHD-WHD, 2001). Bovine brucellosis is found world wide but most European countries are free of bovine brucellosis (Garcia-Garrillo and Lucero, 1993). In the rest of the world rate of infection vary greatly from country to another and between regions within a country. The highest prevalence seen in dairy cattle in Latine America and other developing countries like Africa including Ethiopia is most serious disease (Chukwu, 1987; sierfert, 1996).

In Ethiopia different researches have been carried out to study prevalence of bovine brucellosis in many parts of the country with prevalence rate of 18.4% Kibru (1985) to 1.31% Mulugeta (2006) while the prevalence in humans of occupational workers was 2.4% Tadele (2004) and all negative Mulugeta (2006). Brucellosis, occurring world wide in domestic and game animals as well as in humans, creates a serious economic problem for the intensive and extensive livestock production system. The economic losses due to bovine brucellosis arises from slaughter of cattle herds that are infected with brucella, loss of calves due to abortion, birth of weak calves that die soon after birth, impaired fertility, decrease milk yield, endangering animal export trading of a nation, government costs incurred for research and eradication programs (Chukwu, 1987). It is estimated that bovine brucellosis causes a 20% to 25% loss in milk production as a result of interrupted lactation due to abortion and delayed conception (PAHO-WHO, 2001). Besides the reduction in milk production, there is also loss of calves, temporary or permanent infertility and interferes with the breeding program due to culling of valuable cows. This is of greater importance in beef herds where calves represent the sole source of income (Radostits *et al.*, 1994). Available information indicates that brucellosis is one of the most serious diseases in cattle in Latin America as well as other developing areas. Official estimates put annual losses from bovine brucellosis in Latin America at approximately US \$ 600 million (PAHO-WHO, 2001). The annual economic loss due to brucellosis in cattle was estimated to be 150 million french francs in Cote D'Ivoire, US \$ 233.88 million in Nigeria and US \$ 33.4 million in Kenya, Tanzania and Uganda (Chukwu, 1987; Shirima *et al.*, 2007). In central Africa, an incidence of infection in cattle of above 30% has been found and economic losses of the yearly income of the animal holder have been calculated of up to 6% (Seifert, 1996). The objective of this study was to

determine the prevalence of bovine brucellosis and to assess risk factors associated with the occurrence of the disease in Asella governmental dairy farm.

MATERIALS AND METHODS

The study was conducted in Asella, government dairy farm. Oromia regional state, which is located at 175 km south east of Addis Ababa. The altitude and annual rainfall of the area ranges from 502_ 4130 MSL and 200_ 400mm respectively with mean annual temperature of 22.5°C. It is one of the highest populated area in Ethiopia with estimated human population of 2,521,349 and live stock population of Cattle- 82190, Sheep – 51292, Goat- 811479, Poultry- 562915, Equine- 22055. All the eligible animal, which were greater than 6 month and no history of vaccination was subjected. A total of 304 animals were tested with RBPT and confirmed via CFT. A cross sectional study was conducted on both pure and cross breeds using serological test (Rose Bengal Plate test (RBPT) and Complement fixation test (CFT) was done in and around Asella government dairy farm. 304 animals were tested out of which 62 were male.

Blood sample collection

About 60ml of blood was collected from the jugular vein of each animal using plain vacutainer tube and allowed to clot over night in a slant position at room temperature, finally the serum sample were decanted and stored at -20°C until it tested. The RBPT was performed according the standard procedure. Serum stored at -20°C was first removed and let to melt then 30ml serum was taken from each sample using micropipette and again 30ml commercial prepared antigens taken and place in opposite site of each micropipette wells. Mix thoroughly and left for 4 minutes, finally positive result were categorized based their degree of agglutination. The CFT was done at National veterinary Institute, Debraezeit, Ethiopia according to the protocol recommended (OIE, 2004).

RESULTS

Overall sera prevalence

A cross-sectional study to determine the sera prevalence of brucellosis was conducted between October 2007 and March 2008 in Asella governmental dairy farm. Serum samples were collected from cattle comprising both indigenous and exotic breeds and that were above 6 months of age. All the collected sera were subjected to RBP test and forty three (n=43) sera were found to be positive. Upon further testing of the RBP test positive sera with CFT all sera (n=43) became positive. Thus, the subsequent statistical analyses were based on the 43 sera that were positive to both RBPT and CFT (serial interpretation) test result. The overall sero-prevalence of brucellosis in the farm was 14.14% (n=43)

Potential Risk Factors

Sex

Sex is believed to have implication in the epidemiology of brucellosis. Thus comparison was made on the sera prevalence of male and female animals to assess the existence of any association between the prevalence and sex. Accordingly the sera prevalence of female was 16.94% (n=41) and that of males 3.2% (n=2) as shows in table 1. Statistical analysis revealed significant difference

in sera positively between the two sexes (OR=5.4, p=0.006).

TABLE 1. Sera prevalence of brucellosis in cattle analyze according to sex

sex	No. tested	CFT Positive	Prevalence (%)
Male	62	2	3.22
Female	242	41	16.94
Total	304	43	14.14

Pearson $\chi^2=7.65$, $p=0.006$, OR=5.4

Age

Animals in this study were categorized in two groups, the first group comprising animals with age six month to five years and second group containing animals older than five years. Age is supposed to have some association with the occurrence of brucellosis. Because, sexual maturity is very important for the rapid multiplication of brucellosis organisms. Thus, the two age groups were compared for sera positively which is indicated in table 2. The sera prevalence of the disease in the age groups 6 month -5 years and greater than 5 years was 4.65% and 26.512% respectively. Highly statistically significant difference was observed between the age groups with OR of 5.7 and $p=0.000$.

TABLE 2. Sera prevalence of brucellosis in cattle analyzed according to age

sex	No. tested	CFT Positive	Prevalence (%)
6 month-5 years	172	8	4.65
>5 years	132	35	26.51
Total	304	43	14.14

Pearson $\chi^2=29.39$, $p=0.000$, OR=5.7

Breed

There is still controversy among different authors on the issue of breed susceptibility to brucellosis. The present study attempted to look in o the existence of any association between sera positively and breeds of the animals. Therefore, the prevalence of local and the cross breed animals was compared in (Table 3). The sera prevalence of local and cross breed cattle was calculated as 23.61 and 12.21 having a significant variation with OR of 2.1 and $=0.008$.

TABLE 3. Sera prevalence of brucellosis in cattle analyzed according to breed

sex	No. tested	CFT Positive	Prevalence (%)
Locale	72	17	23.61
Cross	232	26	12.21
Total	304	43	14.14

Pearson $\chi^2=6.96$, $p=0.008$, OR=5.1

Origin

The farm used a stock replacement from the local breeds mainly composed of (Arsi and Borena). The study tried to see whether there was a significant association in the introduction of new animals and sera prevalence of

brucellosis in the farm. The result of univariate logistic regression indicated that a sera prevalence of 31.03% 13.95% and 11.20% for Borena, Arsi and cross breeds respectively. Borena breeds followed by Arsi was significantly affected with a significant association of (OR=2.7 and 1.2, $p=0.0023$) (Indicated in Table 4).

TABLE 4. Sera prevalence of brucellosis in cattle analyzed according to Origin

sex	No. tested	CFT Positive	Prevalence (%)
Borena	29	9	31.03
Arsi	43	6	13.95
Cross	232	28	11.20
Total	304	43	14.14

Test statistics: Pearson $\chi^2=12.40$, $p=0.002$

Parity

Parity of an animal was one of the potential risk factors in the study area. Results of statistical analyzed that there was a significant variation with (OR=2.76, $P< 0.000$) as it is indicated in Table 5.

TABLE 5. Sera prevalence of brucellosis in cattle analyzed according to Parity

sex	No. tested	CFT Positive	Prevalence (%)
No parturition	110	6	5.45
Single parturition	10	0	0
>one parturition	122	35	28.86
Total	242	41	16.94

Test statistics: Pearson $\chi^2=24.31$, OR=5.2 $p=0.000$
 Result of fisher’s exact test showed that history of previous abortion and retained placenta were significant (p , 0.001 and 0.05 respectively).

DISCUSSION

The individual sera prevalence of Bovine brucellosis in cattle in the intensive dairy farm, conducted by different researchers in different parts of the country, Ethiopia, indicated that it was fluctuating between 18.4%, in and around Adiss Abeba (Kibru, 1985) and 0.14%, in North Gondar (Tadesse, 2003). Then highest sera prevalence obtained by Kibru (1985) was extremely reduced in the successive reports, while the present study revealed there was a great rise from the previous reports. As it is indicated 1.9% in and around Shola(Asegid, 1987), 7.62% in Arsi region (Bayleyegn, 1987), 8.1% in and around Addis Ababa (Yilkal, 1998), 1.5% in south Eastern Ethiopia (Abay, 1999), 0.14% in Gondar (Tadesse, 2003), 2.26% in Sidamo zone (Kassahun, 2006), 0.2% (Tolosa, 2004) and 1.13% in Addis Ababa and Sululta abattoirs (Mulugeta, 2006). Except a report conducted by Taye(1991) in Arsi region which has the same epidemiological circumstances to this study obtained a similar serological prevalence of bovine brucellosis (14.2%) the rest have had significantly low prevalence. Otherwise the raise in prevalence of the present study

agrees with characteristics of the husbandry system. According to FAO-WHO(1986) the level of brucellosis infection tends to be relatively high in intensive farms, where those have indigenous cattle or introduced breeds. It has been also reported that the risk of infection increase with change from the pure extensive to more intensive form of cattle management (Thinn and Wundt, 1976). Further more, the uphealing of the present prevalence might also be due to the spread of the disease because of movement of infected animals from infected herd to none infected susceptible herd (Radositits *et al*, 1994). On top of this the presence of unvaccinated animals in the infected herd, herd size, population density, maternity pen do have a direct association with the elevate of the prevalence of the disease (Blood *et al.*, 1979) and Nicoletti, 1980, additionally the carelessness in the hygienic measures such as isolation and disposal of infected animals, aborted fetus, retained placenta and uterine discharge disseminate the disease with in the herd. Different studies reported that older animals were more susceptible than younger animals (Yilkal, 1998; Bekele, 2000; Tolosa, 2004) which agrees with the present study. There was also report that indicated the serological response of male animals is limited and the test of infected male animals were usually observed to be non reactors or shown to be low antibody titers (FAO-WHO, 1986). One research finding publicized that male cattle are more resistance than female (Nicoletti, 1980) however the apparently high sera prevalence figure in female animals compared to male animals in this study agrees with the works of (Yilka, Kubuafor *et al.*, 2000, Bekele, 2000).

A study conducted by Gebretsadik (2005) in Tigray region state, Ethiopia, showed that there was not significant variation among female animals categorized in different parities however, other study done by Yilkal (1998) Indicated that there was significant variation with parity which agrees with the present study.

In respect to the breed of animals that had taken in this study, the local beads in general and Borena in particular obtained to be an animal with high sera positively in relation to breed prevalence. The variation local animals were highly affected might be due to the origin of the animal from the previously infected or exposed herds.

CONCLUSION AND RECOMMENDATION

The study indicated higher prevalence of bovine brucellosis in the Asella government dairy farm than previous prevalence obtained else in the country. All the risk factors in the study site showed a significant various that played role in dissemination and delivery of the disease in the farm. A statistical significant was observed in female animals than female animals. It also revealed that older animals were highly infected than younger animals and parity of individual animals was found to have direct association in dissemination of the disease to increase with the number of parity. The movement of animals or introduction of animals without testing and quarantine could also the spread of the disease.

The following recommendations are forwarded on the bases of the results and conclusion of this study.

- There should be separation of animals based on sex, age, breed, and parity to reduce the disease.
- Husbandry system of dairy farms should be modernized.
- Movement of animals from place to place and introduction of new animals without testing, quarantine, and retested should be banned.
- Further study of bovine brucellosis need to be conducted in and around Asella town to assess the status of the disease both in animals and humans, because currently the farm is acting as a source of milk and first heifer cows for the community of the town.

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