

## Preliminary Survey of Brucellosis in Commercial Dairy Cattle in Debre Zeit, Ethiopia

<sup>1</sup>Abrha Bsrat, <sup>2</sup>Baylegn Molla and <sup>3</sup>Berhe GebreEgziabher

<sup>1</sup>College of Veterinary Medicine, Mekelle University, Mekelle, Ethiopia

<sup>2</sup>Infectious Diseases Molecular Epidemiology Laboratory, Department of Veterinary Preventive Medicine, College of Veterinary Medicine, The Ohio State University, Columbus, Ohio, USA)

<sup>3</sup>FAO Office, Roma, Italy

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**Abstract:** This study was carried out to determine the sero-prevalence of bovine brucellosis in dairy animals and to assess possible risk factors that could promote its transmission to man in the study area. Cross sectional study design and conventional sampling strategy were employed. Rose Bengal Plate Test (RBPT) for screening and Complement Fixation Test (CFT) for confirmation were used. A total of 370 blood samples were collected from five farms of non-vaccinated, crossbreed dairy cattle of both sex and different age level. Furthermore, a total of 47 cattle attendants and owners in the farms were interviewed. The overall prevalence of brucellosis in the study area was 1.9% and all positive sera were from female cattle. Result of the questionnaire survey revealed a prevalence of 4.1% for abortion and 7.3% for retained fetal membranes. Among the interviewed participants 66% were found to have no knowledge of brucellosis, 34% wear protective gloves during handling aborted material and 57.5% responded that they used consuming of raw milk. Results of this study showed that bovine brucellosis in the study area is low and test and slaughter policy can be used in order to control the disease in the dairy animals of Debre Zeit.

**Key words:** Bovine Brucellosis • CFT • Dairy Animals • Prevalence • RBPT • Serum

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### INTRODUCTION

Ethiopia has the largest number of livestock in Africa and the most diversified domestic animal genetic resources. However, the performance of livestock as food producer in Ethiopia is poor [1]. However, their performance is low because of inadequate feed supplies, poor genetic, less reproductive efficiency and abundant diseases [2, 3].

Brucellosis is among the most important diseases confronting food animal production by causing loss of calves and high incidence of retained fetal membrane [4, 5]. Moreover, it decreased milk production, lowered conception rate, prolonged inter calving period, causes sterility, cull of animal due to breeding failure, endangering animal export trade of a nation, loss of man hours and medical costs and government costs on research and eradication schemes [6]. It is distributed worldwide but varies with different species and geographic areas [7].

In Ethiopia, information on economic and zoonotic importance of brucellosis is not well established quantitatively as compared to the degree of the risks of the disease expected due to high animal population of the country and the greater tendency of private as well as governmental farms to expand high producing exotic dairy farms to satisfy the ever increasing milk demand of the urban population [8]. However, several investigators have established the endemicity of bovine brucellosis in different parts of the country [9-17] indicating the disease was wide spread in both indigenous and exotic crosses.

Considering both socio-economic and public health importance of the disease and increasing of the demand for food of animal origin in growing cities of the country it is of important to know the magnitude of brucellosis and identify major potential determinants of the disease which intern assist in devising baseline information to develop national wide brucellosis information. Therefore, this study was carried out to determine the sero-prevalence of bovine brucellosis in dairy animals in the study area and

to assess risk factors that could promote its transmission to man; and based on the results to indicate the measures to be taken in control and prevention of brucellosis in the dairy farms of Debre Zeit.

## **MATERIALS AND METHODS**

**Study Area:** The study was conducted on 370 heads of Holstein cattle breeds of five intensive farms from December 2006 to May 2007 in Debre Zeit, Which is located at about 45 km south east of Addis Ababa. The town is located in the Eastern Shewa Zone of the Oromia Regional State and has a latitude and longitude of 8°45'N 38°59'E with an elevation of 1,920 meters above sea level. The long rainy season extends from June to September with an average rainfall of 800mm and the short rainy season extends from March to May. Annual average temperature ranges from 12-3°C to 27.7 °C with highest temperature is in May [18]. The study district was selected because of the concentration of Holstein cattle breeds in it and its significance as source of milk and milk products for Addis Ababa, Capital of Federal Democratic Republic of Ethiopia. Besides, the town posses farms like Genesis farm (established with an objective of being center of training for farmers from all corners of the country and to be a sample for all business owners who want to establish dairy farm through the whole country) and International Livestock Research Institute, ILRI, dairy farm (center for adaptation and crossing of Holstein breeds for distribution to the whole country).

**Study Population:** The target population was cattle, which consists of breeding females, replacement heifers and available bulls of exotic breeds. A total of 370 animals of above six month of age were sampled of which 50 were heifers, 3 bulls and 317 breeding females. Breeds of cattle in dairy farms were crosses of Holstein type. None of the animals tested were vaccinated against brucellosis.

### **Study Design**

**Sampling Methodology:** The study was a cross-sectional type. Five farms were selected by conventional sampling method based on the voluntaries of the owners and then animals with in the farms were selected using simple random sampling. The sample size of the dairy cattle was calculated according to Thrusfield [19] on the basis of 5.6% prevalence of bovine brucellosis in the study area [2]. The minimum sample size at 95% level of confidence, with 5% desired precision was calculated to be 81 dairy cattle. However, a total of 370 dairy animals were sampled.

**Collection of Blood Samples:** Blood sample was obtained from jugular vein of each sampled animal using plain vacutainer tube and needle. This was done according to the protocol given by OIE [20].

**Serology:** Rose Bengal plate Test (RBPT) was used as a screening test of the serum samples for the presence of Brucella agglutinins using the procedure described [21]. RBPT Brucella antigen (Institution Pourquier, France), positive control and negative control sera (National Veterinary Institute (NVI), Debre Zeit, Ethiopia) were used. All RBPT positive sera samples were confirmed by CFT following the protocol by MAC MILLAN (1990). Compliment fixation test antigen (Central Veterinary Laboratory, Weybridge), Complement and Amboceptor (Biomérieux, France), 2% sheep RBC, positive and Negative control (NVI, Debre Zeit, Ethiopia) were used. CFT is the recommended confirmatory test for brucellosis with high specificity whereas RBPT is the recommended screening test with high sensitivity [22].

**Questionnaire Survey:** A total of 47 cattle attendants and owners in the farms were interviewed to assess risk factors for the transmittion of brucellosis from cattle to cattle and possibility of humane acquiring brucellosis in the study area. Standard questionnaire format was designed to obtain information about the awareness of cattle attendants in the farms and /or farm owners on the risk of brucellosis transmittion from cattle to human. The contribution of herd management practice (herd size, use of maternity pens, breeding system, disposal of aborted materials, animal replacement strategies and fate of aborted cows, history of vaccination, feeding system and health service) and other risk factors for the occurrence and dissemination of brucellosis was assessed. During each sample collection the sex, breed, source, history of abortion and retained fetal membrane was recorded. Furthermore, knowledge on brucellosis, use habit of protective gloves during handling of aborted material or not and consumption of raw milk was included in the questionnaire survey.

**Data Analysis:** The apparent prevalence rate was calculated by dividing the number of RBPT/CFT positive animals by the total number of animals tested. The association of brucellosis with history of abortion and retained fetal membrane was analyzed using chi-square test. P-value < 0.05 and 95% confidences interval (CI) were used to determine statistical significance.

Table 1: Prevalence of brucellosis and its distribution among age and sex groups in Debre Zeit (December 2006 - May 2007)

Factors		Animals examined	RBPT +ve (%)	CFT -ve (%)	Over all prevalence
Sex	Male	3	0 (0%)	0 (0%)	1.9%
	Female	367	7(1.9%)	7(1.9%)	
Age (yrs)	6month-3yrs	52	0 (0%)	0 (0%)	Yrs = years
	>3yrs-6yrs	108	1(0.9%)	1(0.9%)	
	>6yrs	210	6(2.9%)	6(2.9%)	

Table 2: Association of brucellosis with Abortion and Retained Fetal Membrane in Debre Zeit (December 2006 - May 2007)

Categories		RBPT +ve (%)	RBPT -ve (%)	Total (%)	X <sup>2</sup>	P-value
History of Abortion	Yes	4 (1.1)	9 (2.4)	13 (4.1)	60.8	< 0.001
	No	3 (0.8)	301 (81.4)	304 (95.9)		
History of RFM	Yes	4 (1.1)	19 (5.1)	23 (7.3)	32.0	< 0.001
	No	3 (0.8)	291 (78.6)	294 (92.7)		

## RESULTS

Of the 370 serum sample RBPT examined for brucellosis screening, 7(1.9%) were positive which were in turn positive for CFT and all of them were female with high prevalence of those age group above 6 years (Table 1).

Among 317 breeding females RBPT screened, 13(4.1%) for history of abortion and 24(7.3%) for retained fetal membranes were recorded showing statistically significant association with brucellosis distribution in the study site (Table 2).

Questionnaire survey on epidemiological risk factors showed that vaccination is not used in all sampled dairy farms. All the five farms used stall feeding and have veterinary professionals for health service provision. Eighty percent of the farms replace their dairy animals from their own with the rest 20% used both market and own farm, used artificial insemination as means of breeding system and have separate maternity pens. Indigenous knowledge of questionnaire respondents (cattle attendants and owners) on encouraging of brucellosis transmission from cattle to humans showed 66% (31/47) of them have no knowledge of brucellosis and 34% (16/47) wear protective gloves during handling aborted cows, retained fetal membranes. Furthermore, 57.5% (24/47) responded positively on the consumption of raw milk.

## DISCUSSION

The result of the present investigation agrees well with the reports of the prevalence 2.1% around Shoa in

central highland of Ethiopia [17], 1.8% in Amhara National Regional State (personal communication), 0.7% in Tigray National Regional state [14], 0.1% in North Gondar [13] and 1.5% in Addis Ababa. However, other studies carried out in different part of the country indicated relatively higher prevalence of the disease: 7.8% in Arsi [9], 8.1% around Addis Ababa [2], 11.6% in Sidama Region [10] and 12.5% in Wuchale-Jida [23]. This might be due to difference in awareness to dairy farm owners husbandry management. In line with this the questionnaire survey indicated that 80% of the assessed dairy farms follow culling strategy of aborted cows for any reason and replacement of dairy animals is mainly from their own herds that can lead to minimal introduction of brucellosis. Furthermore, in Debre Zeit, artificial insemination is being practiced and semen is supposed to be collected and processed from brucella free breeding bulls. Contact between farms was also negligible as more than 90% of the intensive farms had no contact with neighboring herds. Furthermore, new reports are coming from various animal species of different animal production at different settings of the country [24 – 27].

The absence of male reactor animals in this study agrees with the findings of other investigators [2,28-30] that could probably be due to the small number of sampled male (n=3) animals. However, it had been reported that males are usually more resistant than female cattle [2, 24, 31]. It was also reported that serological response of male animals to brucella infection is limited. Tests of infected male animals were usually observed to be non-reactors or showed low antibody titers [32]. Similarly, it was also stated that male cattle are more resistant than females [33].

Different factors are probably involved in sex susceptibility variation including physiological and behavioral difference. Because of the preferential growth of *Brucella abortus* in gravid uterus that can enter as it disseminated from the principal sites of carrier states (udder, supramammary lymph node). In latently infected cows depending on the number of pregnancy events and presence of infection, the organism can get sufficient contact with the lymphoid system to stimulate a significant immune response [5]. On the other hand, some locally infected bulls in their testes are known to be non-reactors or only had low antibody titers [31]. This suggests that serological test may under estimate *Brucella abortus* infection in males, because of the successful confinement of the bacteria in the testes and reticuloendothelia system [33]. Besides, the greater prevalence rate in females might be due to the behavior to lick or sniff their newly born calves, if infected, can lead to re-infection but bulls are usually kept separately [7].

#### CONCLUSION AND RECOMMENDATIONS

The study showed low evidence of *Brucella* infection. However, the positive animals can be a potential hazard to animals and human in the study area due to the increased demand for animal products like milk and meat. Most of the respondents consumed raw milk from potentially infected animals and have no knowledge of brucellosis and handle aborted fetus and fetal membrane with bare hands. Therefore, unless extensive extension service is launched to make the stock owners aware of the disease, the present prevalence of the disease can increase within short period of time.

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