A Cross-sectional Study on Bovine Tuberculosis in Small Holder dairy farms of Guto Gidda District, East Wollega Zone, Western Ethiopia

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Abstract: Bovine tuberculosis (BTB) is a contagious chronic and debilitating disease of cattle that can infect humans, other domestic animals and some wild life. The present study was aimed to estimate the prevalence of BTB and associated risk factors in small holder dairy farms of Guto Gida district in East Wollega Zone, Western Ethiopia. A cross-sectional study design was carried out using comparative intradermal tuberculin test from January to September 2014, in small holder dairy farms Guto-Gida district to determine the prevalence of BTB. Purposive sampling technique was used. The current study showed that from a total of 295 cattle tested, 24 (8.14%) were found to be positive for BTB. Out of the total examined animals, 60 (20.3%) were males. The effects of different risk factors (like sex, age, breed type, and body condition score) for the occurrence of BTB were investigated. The difference in reactivity to the CIDT test among the study participants in different age groups was statistically significant (P-value = 0.027) showing higher risk of BTB in older animals when compared to the younger ones (OR=4.03, 95% CI, 1.17-13.85). This study revealed the significance of the disease in the study area. Farm owners and residents are usually in close contact with these animals and also consume raw milk regularly which predispose for high infection risks to them. Thus, further detailed epidemiological studies should be done to investigate the link between bovine and human tuberculosis in the study area in order to design appropriate strategic prevention and control measures.

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Introduction

Bovine tuberculosis (BTB) is described as a contagious, chronic and debilitating disease. Among the livestock species, cattle mostly are affected by the principal cause *Mycobacterium bovis* (*M. bovis*), which is genetically related to and clinically indistinguishable from TB caused by *Mycobacterium tuberculosis* [1].

Mycobacterium tuberculosis is the most common cause of human TB, but an unknown proportion of the cases are due to M*ycobacterium bovis* (*M. bovis*) [2]. The role of *M. bovis* in human TB is well established in developed world, though it is not well known in developing countries [3-4]. In Africa to which 82% of human and 85% of animal population live in area where BTB is either partly or not controlled at all [1], it was a significant zoonotic human pathogen that aggravate the 'triple trouble' of HIV/AIDS, TB infection and malnutrition [5]. Similarly in Ethiopia, the disease is expected to have a significant public health impact as 95% of the farmers are still keeping zebu cattle using the traditional animal husbandry system, consuming animal product raw and often shares the same sheltering with animals [6].

It causes great economic losses concomitant to the actual biological constraints like low productivity, low genetic potential and prevalent disease. In Ethiopia, BTB has been an epidemic since 1967 with prevalence ranging from 15.6% to 50% reported in dairy farms around the country [7]. Recently, BTB prevalence was 5.15% in slaughter animals, in abattoir based study [8].

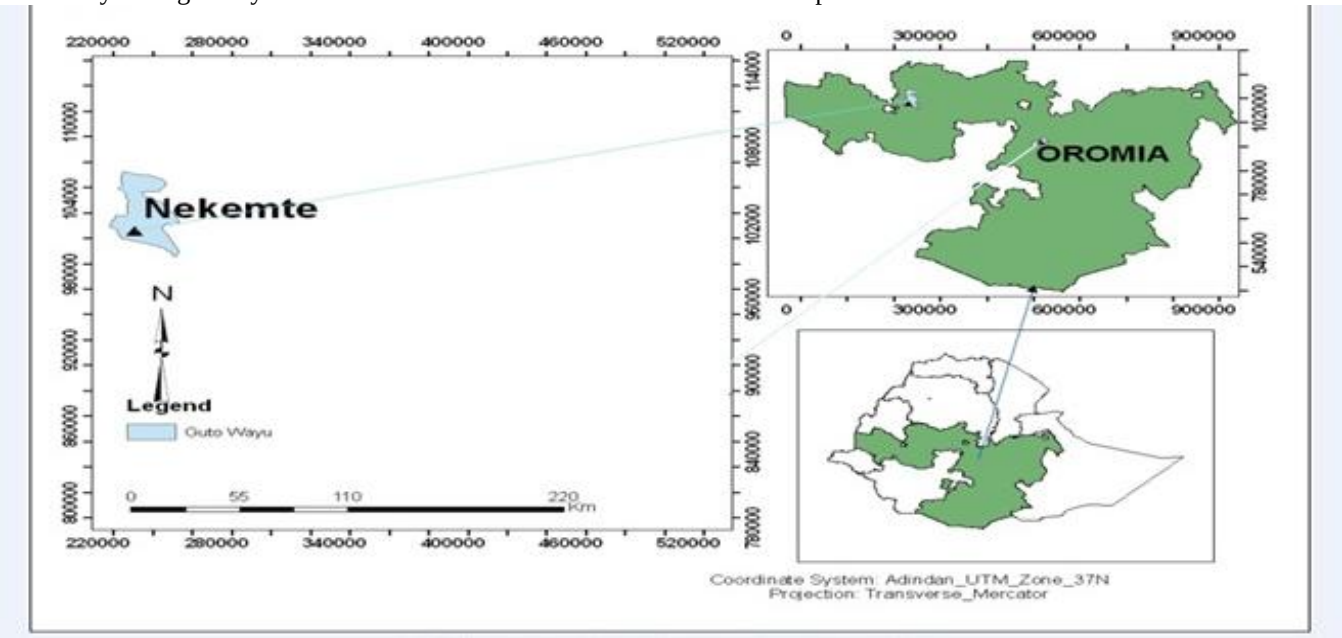
Mycobacterium bovis is an intracellular pathogen of Macrophages and other cells of monocytic type. Whilst the diagnosis of many infectious diseases of animals relies on the detection of humoral (antibody) response to the infectious agent, the predominant immunological response in *M. bovis* infected cattle is affected by T- lymphocytes [9]. This cell-mediated immune (CMI) response is both the mechanism of defense and the cause of chronic inflammation (granulomas) characteristic of *M.bovis* infections [10].

Therefore, ante mortem test of cellular immunity can identify *M.bovis* infected animals earlier and have great sensitivity than the antibody based assays evaluated to date. The two standard tests currently approved for the diagnosis of TB in cattle, namely the in- vivo intra dermal tuberculin test and the in- vitro γ-interferon assay, follows this principle [11].

Diagnosis of *M. bovis* infection in cattle is often based on history, clinical and necropsy findings and tuberculin test. For confirmatory diagnosis, culture and biochemical property have been used. To differentiate *M. bovis* from other members of *M. tuberculosis* complex, nucleic acid recognition method using polymerase chain reaction (PCR) and DNA genome sequence have been used [12-13]. Several new laboratory procedures like lymphocyte proliferation and γ-interferon assays have been introduced as aid in the diagnosis of mycobacterium infections. However; because of technical problems and cost, with the exception of tuberculin test, they have not come into widespread use in veterinary diagnostic laboratories and in fields on large scale basis [14].

The effective ante mortem surveillance for BTB primary relies on the detection of the infected cattle at an early stage by the use of sensitive immunodiagnostic test [15-16]. Intradermal tuberculin test detects a delayed type hypersensitivity reaction to the intradermal injection of tuberculin [17]. It is the only method in general use today and the ante mortal test prescribed by the office international des Epizooties (OIE) for international trade of cattle [16]. Tuberculin test has been used for the diagnosis of BTB for more than 100 years. Limitation of the test has been recognized for many years, but the amount of published data available to validate its performance is still limited. This is primarily because of the fact that experiment to establish the sensitivity and specificity of the test are expensive and labor intensive as it require slaughter of animals and postmortem examination [18].

In Ethiopia, tuberculin test is used for the diagnosis of BTB in live animals. Despite the limitation of the test, it remains the primary ante mortem diagnostic tool for TB in cattle, providing a cost-effective and reliable means of screening entire cattle populations. Since there was lack documented information in the study area, this study was designed to determine the prevalence of bovine TB and associated risk factors in small holder dairy farms of Guto Gidda District in Eastern Wollega Zone, Western Ethiopia.



Source: [19]

Figure 1: Map of the study area

Materials and Methods

Study area

A cross-sectional study was conducted from January to September 2014 in small holder dairy farms Guto-Gida district, East Wollega Zone, Western Ethiopia. The district is found at 331 kms west of Addis Ababa on Addis-Assosa road. It lies between 9° 5'N and 36° 33'E, on an altitude of 2088 mails and enjoys an average annual rainfall of 1200 mmHg (Figure 1). The average daily minimum and maximum

temperatures range between 15 and 27° C, respectively [19]. Agriculture is common in the region particularly; coffee, maize and sorghum are widely practiced, whereas livestock constitutes the primary sources of income for the community. According to the district animal statistics agency the animal population of the district accounts: 26,553 cattle, 101 horse, 28 mule, 930 donkey, 1960 sheep 878 goat and 8599 poultry.

Study animals

The study was conducted on all cross and local breeds kept in the small holder intensive dairy farms of the district purposively that varies in age, sex, breed, body condition and management.

Sampling and sample size determination

Convenient sampling method was employed so that the district was selected purposively based on their livestock population in farms (both local and crossbred types) and ease of accessibility and all the animals in the farms were tested. A total of 295 dairy animals were tested by comparative intradermal tuberculin test and for validation o this study.

Study design

CIDT test was used mainly to differentiate between animal infected with *M.bovis* and those sensitized to tuberculin due to exposure to other mycobacterium or related genera. Two sites at the middle of the neck were shaved and cleaned 12 cm apart on the left side of the neck. The area was examined for the presence of any gross lesion. The skin fold at the two sites were measured by a caliper and recorded. Each animal was then injected 0.1ml (2500 IU/ml) avian tuberculin PPD (Veterinary Laboratories agency Addleston, surrey KT153NB) and 0.1ml (2500 IU/ml) bovine tuberculin PPD (Veterinary Laboratories agency Addleston, surrey KT153NB) intradermaly at the anterior and posterior part respectively. The sites were examined and the skin thickness was measured 72 hours post injection. The results were interpreted in accordance with the recommendations of the OIE [20]. Briefly, when the skin thickness is increased by 4mm or more at bovine PPD injection site regardless of the increase at avian site, the animal was considered as positive for BTB and when the skin thickness is increased at sites, the difference of increase at bovine (B) and increase at Avian (A) sites was considered. Thus, when ΔB-ΔA is less than 2mm, between 2mm and 4mm or 4mm and above, the animal was considered as negative, suspect/doubtful or positive, respectively.

During the data collection, the individual animal identification number, origin, breed and sex was recorded. Age category was estimated by using the dental eruption and wear as described [21] Body condition score was made using method developed for zebu cattle [22].

Data analysis

The collected data was entered into Microsoft Excel spread sheet and analyzed by using STATA version 11software. Relationships between test result and potential predictor variables were assessed using bivariate and multivariate logistic regression model. P-values of less than 0.05 were considered statistically significant.

Results

Demographic characteristics of the study participants

Table 1: General characteristics of the study subjects (n=295)

Characteristics	Frequency	Percent (%)
Sex		
Male	60	20.3
Female	235	79.7
Age		
Adult	193	65.4
Young	102	34.6
Breed		
Cross	238	80.7
Local	57	19.3
Body condition		
Good	62	21.0
Medium	196	66.4
Poor	37	12.5
Test result		
Positive	24	8.14
Negative	271	91.9
Total	**295**	**100**

The comparative intradermal tuberculin test (CIDT) in the current study showed that from a total of 295 cattle tested, 24 (8.14) were found to be positive for BTB (Table 1). Out of the total examined animals, 60 (20.3%) were males and 235 (79.7%) were females. The effects of different risk factors (like sex, age, breed type, and body condition score) for the occurrence of BTB were investigated. Accordingly, even though there was no significance difference between both sexes a higher prevalence of BTB in female animals were recorded (6.67% in males and 8.51% in females). The difference in reactivity to the CIDT test among the study participants in different age groups was statistically significant (P-value = 0.027) showing higher risk of BTB in older animals when compared to the younger ones (OR=4.59, 95% CI, 1.23-17.20) (Table 2).

Table 2: Crude and Adjusted odds ratios for various factors that might affect the test result

Characteristics	Test result (%)		Crude OR	95% CI	P-value	Adjusted OR*	95% CI	P-value
	+Ve No	-Ve No						
Sex								
Male	4 (6.67)	56 (93.33)	1	------	-----	1	-----	-----
Female	20 (8.51)	215 (91.49)	1.30	0.43-3.96	0.642	0.79	0.23- 2.70	0.709
Age								
Young	3 (2.94)	99 (97.06)	1	-----	-----	1	-----	-----
Adult	21(10.88)	172 (89.12	4.03	1.17-13.85	0.027	4.59	1.23-17.20	0.024
Breed								
Cross	23(9.66)	215(90.34)	1	-----	-----	1	-----	-----
Local	1(1.75)	56 (98.25)	0.56	0.16-1.95	0.363	0.53	0.15- 1.86	0.318
Body condition								
Good	5 (8.06)	57(91.94)	1	-----	-----	1	-----	-----
Medium	15 (7.65)	181(92.35)	0.95	0.33-2.71	0.916	1.05	0.36-3.10	0.926
Poor	4 (10.81)	33 (89.19)	1.38	0.35-5.51	0.647	1.66	0.40-6.92	0.484

OR = Odds Ratio, CI = Confidence interval, N = number * = All the variables in the table are included in the model.

Discussion

In this study, the overall individual animal prevalence of BTB in small holder dairy farms of Guto- Gida district was 8.14% by using CIDT test. This result was in line with the findings reported by the previous researches conducted in other parts of the country that indicated an average prevalence rate of 15% BTB ranging from 3.4% in small holder production system up to 50% in intensive dairy productions [23-24, 7, 25-26].

Among the potential associated risk factors, age of animals was identified by numerous studies in both developed and developing countries. The duration of exposure increases with age; older animals are more likely to have been exposed than younger ones, as shown by several cross-sectional studies carried out in Zambia, Tanzania, and Chad [27-31], which was in agreement with the present finding in the current study areas. This was reasoned out by [32], in that animals might get infected at a young age, but only expresses the disease clinically when they are adults. Moreover, [33], further argued and explained the late occurrence of disease in that Mycobacteria can remain in a latent state for a long period in an infected animal before reactivation at an older age.

The Zebu (*Bos indicus*) breed cattle are thought to be much more resistant to BTB than European cattle [34]. The result of the present study concurs where we found the prevalence to be 1.75% in local breed, whereas 9.66 % in crossbred cattle from Holestin Fresian and local breeds. Sex has only been mentioned as a risk factor in studies carried out in Africa. Opinions diverge regarding its influence on the susceptibility to a *M. bovis* infection. A cross-sectional study conducted from 2006 to 2007 on 1470 animals in Uganda revealed significantly more females positive to the skin test than males [30], which is in line with the result of the present study.

In contrast to the current result and other cross-sectional studies, a cross-sectional study conducted in Tanzania from 1994 to 1997, which included 5692 indigenous and 244 exotic cattle, revealed that male cattle were significantly more affected by BTB than female animals [28]. Justification given for this as explained by the same author was that male cattle are mostly used as draught oxen, which are kept longer in the herd than females. Due to this particular longevity, it is more probable that they get in contact with infected cattle from other affected herds and in turn get infected. This would imply that contact between herds is a major source for bovine TB transmission.

Conclusion

In conclusion, the prevalence reported in the current study only using CIDT test revealed that BTB is present and is well-established in small holder dairy farms in the district. Farm owners in the study areas are usually in close contact with their animals and also consume raw milk regularly which may predispose for the disease. Thus, the livestock agency of the district and the researchers should give awareness creation to the farm owners on the relevance of screening test for BTB before purchasing the animals. Moreover, further detailed epidemiological studies should be done to investigate the link between bovine and human tuberculosis in the study area in order to design appropriate strategic prevention and control measures.

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