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Assessment of bovine tuberculosis and its risk factors in cattle and humans, at and around Dilla town, southern Ethiopia

Gebremedhin Romha[1, *], Gebremedhin Gebre egziabher[2], Gobena Ameni[3]

[1]Dilla University, College of Agriculture and Natural Resource, Department of Animal and Range Science, P.O. Box 419, Dilla, Ethiopia
[2]Samera University, College of Veterinary Medicine, P.O. Box 132, Samera, Ethiopia
[3]Aklilu Lemma Institute of Pathobiology, Addis Ababa University, P.O. Box 1176, Addis Ababa, Ethiopia

Email address:
gebremedhinromha@yahoo.com (G. Romha), greatmedhin2010@gmail.com (G. G. egziabher), gobenachimidi@yahoo.co.uk (G. Ameni)

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Abstract: A cross-sectional study was carried out from February, 2012 to June, 2013 using comparative intradermal tuberculin (CIDT) test on 440 individual live cattle kept in 118 housholds/farms to determine the prevalence of, and to identify the risk factors associated with, bovine tuberculosis (BTB) infection in cattle and to assess the public awareness on its zoonotic importance through structured questionnaires at and around Dilla town, southern Ethiopia. The herd and individual cattle prevalence of bovine tuberculin positivity were 15.3% (95%, Confidence interval (CI): 8.7 to 21.8) and 4.3% (95%, CI: 2.4 to 6.2), respectively at cut-off greater than 4 mm. The herd prevalence of BTB varied significantly among herd size (P=0.019) and management conditions (P=0.031). Herds which had 5-9 and more than 9 cattle were six (Odds ratio (OR) = 6.4) and twelve (OR=12.0) times more reactive to bovine tuberculin testing than herds that had less than or equals to four cattle in their farms. Among the animal related risk factors only breed (P=0.020) had a significant association with the prevalence of bovine tuberculin positivity with the highest odds ratio in Holstein breeds (OR= 6.1) as compared to Zebu and cross (Holstein X Zebu) breeds. Less than one third (29.7%; 35 of 118) of the respondents recognized BTB, and only 22.9% (27/118) of the respondents had understanding of its zoonotic implication. Awareness rising of cattle owners about BTB and its transmission, and the zoonotic implication of BTB is of extreme importance for effective implementation of TB control measures.

Keywords: Bovine Tuberculosis, CIDT Test, DillaTown, Awareness, Risk Factors, Ethiopia

1. Introduction

It is well known that humans and animals have had close interactions. The interaction is becoming largely increased in the 21st century due to the shift from extensive rural production system into the combined urban and peri-urban intensified livestock husbandry to satisfy the rise in demand for animal products.This largely contributes to the ongoing transmission of shared infectious zoonotic diseases from cattle to humans [1]. Bovine tuberculosis among the principal zoonotic diseases [2] caused by *Mycobacterium bovis*, member of the *Mycobacterium tuberculosis* complex (MTC), which affects many vertebrate animals and humans, and characterized by progressive development of granulomas in tissues and organs [3,4].

Tuberculosis (TB) caused by bovine origin has emerged as a significant disease with the tendency for inter-species spread. Bovinetuberculosis has been significantly widely distributed throughout the world and has been a cause for great economic loss in animal production and the most frequent cause of zoonotic TB in man [5]. In developed countries, mandatory pasteurization of milk combined with tuberculin testing and culling (slaughter) of infected cattle resulted in dramatic decline in the incidence of human TB due to *Mycobacterium bovis (M. bovis)* [6]. In Africa; however, BTB represents a potential health hazard to both animals and humans, as nearly 85% of cattle and 82% of the human population live in areas where the disease is prevalent or only partially controlled [7]. In developing countries where BTB is still common and pasteurization of milk is not practiced, an estimated 10 to 15% of human TB cases are caused by *M. bovis* [8].

In Africa though BTB represents a potential health hazard to both animal and human populations as in most developing countries, *M. bovis* infection remains largely uninvestigated. Its epidemiology and public health significance remains largely unknown due to several factors including the high cost of testing programme, social unrest due to political instability, ethnic wars resulting in displacement of large numbers of people and animals, and a lack of veterinary expertise and communication networks [9 –14].

In Ethiopia, BTB is considered to be a prevalent disease in cattle populations. Tuberculin skin test survey indicates that the prevalence ranges from 0.8% in extensive rural farming systems that keep Zebu cattle to 50% in intensive husbandry systems (15-19).Few studies have also indicated the disease is zoonotic; transmitted from animal to humans and vice versa [9,16, 20].

Many studies have shown that there are many risk factors conducive to the spreading and persistence of BTB in developing countries such as demography, eating habits, living and socio-economic status of families, illiteracy, culture and customs, the existence of HIV/AIDS, and close proximity with animals [11,16,21-23]. Ethiopian milk consumers generally prefer raw milk (as compared to treated milk) because of its taste, availability and lower price. The zoonotic risk of BTB is often associated with consumption (ingestion) of dairy products based on unpasteurized milk infected with *M. bovis*. Also, aerosol transmission from cattle-to-human should also be considered as a potential risk factor [16, 19, 24, 25].

In Ethiopia, the epidemiology of BTB is not well established in livestock, and most studies have been focused mainly around the central part of the country. In order to embark the national BTB control program in the future, the epidemiology of the disease has to be assessed widely in all regions of the country where such study has not yet been conducted. The objectives of the present study are therefore to determine the prevalence of, and to identify the risk factors associated with, BTB infection in cattle and to assess the public awareness on its zoonotic importance at and around Dilla town, southern Ethiopia.

2. Materials and Methods

2.1. Description of the Study Site and Population

The study was conducted at Dilla town and its surroundings, southern Ethiopia. Dilla town is the capital of Gedeo zone, located 359 kms from Addis Ababa, the capital of Ethiopia.The geographical location of the research site is 5^0 to 7^0North latitudes and 38^0 to 40^0East longitudes in the escarpment of the Rift Valley at an elevation ranges from 1200 to 3175 meters a.s.l. The climate of Dilla town is characterized as warm humid temperate. Mean annual temperature ranges between 17^{o}C and 22.4^{o}C and mean annual rainfall between 1200 and 1800 mm. Dilla town is thus endowed with two rainy seasons, from March to May and from July to December [26]. The study populations were cattle originated from Dilla town and its surroundings and people who reared these animals.

2.2. Study Design and Sampling Method

A cross-sectional study was conducted from February, 2012 to June, 2013 using comparative intradermal tuberculin (CIDT) test on 440 live cattle to determine the prevalence of BTB and to identify the risk factors associated with BTB infection in cattle. Moreover, a questionnaire survey was carried out through structured questionnaires format to assess the knowledge and awareness of cattle owners on BTB and its zoonotic importance as well as the routes of transmission of the disease.The sample size for tuberculin testing was calculated using the cluster sampling formula described by Bennett et al. [27]. We assumed an intra-class correlation coefficient (rho) of 0.2, an expected prevalence of 11.6 % and standard error of 2.0%. The total sample size calculated for 118 households/farms was n=440 cattle.

The standard error (s.e.x) which measures the precision of our estimate is given by (i):

$$\text{s.e.x} = \sqrt{\frac{pqD}{n}} = \sqrt{\frac{pqD}{cb}} \qquad \text{(i)}$$

Rho (ρ) describes the rate of homogeneity, thus the variability is given by (ii):

$$\rho = \frac{(withinHerd\ Variation)}{TotalVaria\ tion} \qquad \text{(ii)}$$

$$D = 1 + (b-1)\rho \qquad \text{(iii)}$$

Where ρ = rho, s.e = standard error, D =design effect, b = samples per cluster, c = number of clusters, q =1-p and p = expected prevalence [28].

A list of households interested to participate in the study was established through the support of the agricultural and rural development office of the zone and used as a sampling frame. Thus, households/farms were considered as primary units; while individual animal were used as secondary units. The primary stage was sampling of households those who owned smallholder dairy farms. In secondary stage, animals within households were selected. Both the primary and secondary units were randomly selected (using random number technique).Thirty percent of each of the selected herds was included in the study. Accordingly, a total of 440 cattle belong to 118 households were selected in this study.

2.3. Study Methodology

Comparative intradermal tuberculin test was conducted on 440 live cattle using both avian and bovine purified protein derivates (PPD) (Lelystad Biological BV. Lelystad, Netherlands) kindly donated by the Animal Health and Veterinary Laboratories Agency (UK). Test procedures and interpretation of the results were done according to the procedures documented in OIE [4]. Management and other important data like animal age, breed, body condition score (BCS), sex, lactation status, pregnancy status, parity class and skin test measurements were recorded. Young cattle ≤6 months of age, and cows in late pregnancy and those recently calved were excluded in the study for fear of immune dysfunction that usually occurs in dairy cows starting around 3 weeks pre-calving to 3 weeks post-calving [29].

All animals in this study were thoroughly observed for their body condition. Body condition scoring was done according to the established guide lines [30]. The management of the farms was categorized as described by Ameni et al. [31] on the basis of housing condition (neatness, waste disposal, nature of the floor, presence of confinement), feeding (concentrate plus hay), possession of an exercise yard, and contact with other herds and provision with clean water. Structured questionnaires were prepared and administered to cattle owners or herders to collect data on the role of various risk factors for the occurrence and spread of BTB among cattle, and between cattle and human. A total of 118 cattle owners or farm workers were interviewed on the same day when their cattle were tested for BTB.

2.4. Data Analysis

Cattle skin test data at individual and herd level were recorded in database based on Microsoft® Excel for Windows 2007. Statistical analysis was carried out using STATA 11 Statistical Software (STATA Corporation, College Station, TX). Individual animal prevalence was defined as the number of positive reactors per 100 animals tested. The farm level prevalence was calculated as the number of herds with at least one-reactor animal per 100 herds tested. Chi-square ($\chi2$) and multivariate logistic regression analyses were used to examine the effect of risk factors on animal prevalence (P). A P-value < 0.05 was considered to be statistically significant.

2.5. Ethical Clearance and Consideration

Our research work was evaluated and approved by the Institutional Review Board (IRB) of the Aklilu Lemma Institute of Pathobiology, Addis Ababa University. The Reference Number of the approval letter is IRB/01/2012-13. Consent was obtained after the purpose of the study was explained to the owners of animals.

3. Results

3.1. Herd Level Prevalence and Risk Factors

The herd level prevalence of BTB was 18/118 (15.3%) (95%, CI: 8.7 to 21.8).The herd prevalence of BTB varied significantly among herd size (P=0.019) and management conditions (P=0.031) (Table 1). Those herds which had 5-9 and more than 9 cattle were six (OR=6.4) and twelve (OR=12.0) times more reactive to bovine tuberculin testing than herds that had less than or equals to four cattle in their farm (Table 1).

Table 1. *Evaluation of the association of risk factors to herd tuberculin test results at and around Dilla town, southern Ethiopia.*

Variable	Number tested	Number Positive (%)	$\chi2$ -value	P-value	OR(95%CI)
Herd size			7.9	0.019*	
<4	36	1(2.8)			1
5-9	39	6(15.4)			6.4(1.9-55.7)
>9	43	11(25.6)			12.0(1.5-38.5)
House hold history of HTB			0.3	0.581	
Absent	114	17(15.0)			1
Present	4	1(25.0)			4.0(0.2-89.7)
Management			4.7	0.031*	
Poor	34	9(26.5)			1
Good	84	9(13.1)			0.2 (0.1-0.9)
Presence of other livestock			0.04	0.841	
Absent	63	8(14.6)			1
Present	55	10(15.9)			2.8(0.2-2.2)
Presence of wild life			1.7	0.197	
Absent	91	16(17.6)			
Present	27	2(9.5)			1.3(0.1-6.2)

* Statistically significant (P <0.05), HTB= Human TB

3.2. Animal Level Prevalence and Risk Factors

The prevalence of bovine tuberculin positivity at individual animal level was 19/440 (4.3%)(95%, CI: 2.4 to 6.2) at cut-off greater than 4 mm. Among the animal related risk factors only breed (P=0.020) had a significant association with the prevalence of bovinetuberculin positivity. Holstein breeds were (OR= 6.1) six and three times more reactive to bovine tuberculin testing when compared to Zebu and cross (Holstein X Zebu) breeds, respectively(Table 2).

Table 2. *Evaluation of the association of selected animal risk factors with prevalence of bovine tuberculin positivity at and around Dilla town, southern Ethiopia* **.**

Variable	Number tested	Number positive (%)	χ2 Value	P-value	OR(95%CI)
Sex			0.12	0.726	
Female	382	17(4.5)			1
Male	58	2(3.5)			0.8(0.2-3.2)
Age (Yrs)			1.15	0.765	
<2	99	3(3.0)			1
2- 4	142	6(4.3)			1.4 (0.3-7.8)
4- 7	162	9(5.6)			1.9 (0.4-9.7)
>7	37	1(2.7)			0.9 (0.1-11.1)
Breed			7.80	0.020*	
Local	117	2(1.7)			1
Cross	240	9(3.8)			2.2(0.6-8.9)
Exotic	83	8(9.6)			6.1(1.3-29.5)
Body condition			0.17	0.920	
Poor	60	2(3.3)			1
Medium	69	3(4.3)			1.3 (0.2-8.5)
Good	311	14(4.5)			1.4 (0.3-5.7)
Lactation			1.04	0.307	
Non lactating	203	7(3.4)			1
Lactating	179	10(5.6)			1.7(0.6- 4.5)
Pregnancy			3.40	0.065	
Non pregnant	261	15(5.8)			1
Pregnant	121	2(1.7)			0.3 (0.1-1.2)
Parity class			3.51	0.173	
Heifer	169	4(2.4)			1
Parity 1-3	186	12(6.5)			2.8(0.9- 9.1)
Parity >4	27	1(3.7)			1.6(0.2- 14.7)

* Statistically significant (P <0.05)

3.3. Assessment of Cattle Owners Awarness on Bovine Tuberculosis

The knowledge on the role of various risk factors responsible for the occurrence and spread of BTB between cattle and people were assessed by a questionnaire on a total of 118 farm owners (households) and the result is depicted in Table 3. As can be seen from Table3, about 29.7% (35 of 118) of the respondents knew that cattle can have TB, and 22.9% (27 of 118) responded that bovine TB can transmit from animal to human and vice versa. The life style and consumption habit of the 118 respondents is shown in Table 4. Several demographic characteristics and other factors were considered to investigate their possible association with BTB recognition of the respondents. The awareness of the respondents regarding cattle infection with BTB and the transmission of BTB from cattle to man improved as the educational background of the respondents increased (data not shown).

Table 3. *Knowledge of cattle owners about bovine tuberculosis and its transmission to humans in the study area.*

Knowledge examined in Questionnaire	Number interviewed	Correct response (%)
TB can affect animals	118	35 (29.7)
TB is zoonotic	118	27 (22.9)
Drinking raw milk is source of infection for TB	118	31 (26.3)
Eating raw meat is source of infection for TB	118	29(24.6)
TB cough spray is source of infection for TB	118	27 (22.9)
Sharing the same house is source of infection for TB	118	26(22.0)

Table 4. *Life style, and milk and meat consumption habit of cattle owners in the study area.*

Habit of respondents	Number interviewed	Percent (%)
Milk drinking		
Boiled milk	118	19 (16.1)
Both raw and boiled milk	118	95 (80.5)
Do not consume milk at all	118	4 (3.4)
Meat eating habit		
Cooked meat	118	25(21.2)
Both cooked and raw meat	118	93 (78.8)
House sharing (live close to their animals)		
Not sharing	118	41 (34.8)
Sharing	118	77 (65.3)

4. Discussion

Discordant with the previous reports [19, 32, 33], this study discovered low prevalence of BTB in the study area. This could be due to the fact that the present study was conducted in smallholder farms with smaller herds, in which some animals were kept in the open air in contrast to previous studies, which were conducted on relatively large herd-sized farms. The transmission of BTB from cattle to cattle is largely influenced by herd size; the larger the herd size the greater the chance of transmission [34]. However, congruent with the result of the present study, low prevalences of BTB have been reported in smallholder dairy farms [29] and in cattle of rural livestock production systems in Ethiopia [18]. When larger proportion of the study animals was grazing in the field, the level of confinement is reduced to a certain degree, which in turn minimizes the rate of infection in the herd [35].

In this study herd tuberculin result showed a statistically significant association with herd management conditions suggesting that poor managerial inputs increase the risk of BTB. Previous studies similarly have reported higher infection rates in farms under poor management conditions [29, 30, 36]. It can therefore be generalised that the status of BTB could be improved by adopting sanitary measures that improve hygiene conditions on farms. Moreover, in the present study, pure Holstein breeds (OR= 6.1) were six and three times more reactive to bovine tuberculin positivity as compared to Zebu and cross (Holstein X Zebu) breeds, respectively which is similar with earlier reports [10,20,37].The probable reason could be the fact that genetically improved cattle suffer more severely from poor housing, under-and malnutrition and subsequently become more susceptible to infection [34].

In the present study, level of awarness of cattle owners about BTB showed that 29.7% of the respondents knew that cattle can have tuberculosis, and 22.9% recognized that BTB is zoonotic. Ameni et al. [29] have indicated that lack of understanding regarding the zoonotic of BTB, food consumption behavior and poor sanitary measures is the potential risk of BTB to public health. Likewise, in this study, more than 80% of the respondents were consuming raw milk. Humans acquire the infection primarily by ingesting the agent in raw milk and milk products, and secondly by inhaling it when there is close physical contact between the owner and his/her cattle, especially at night since in some cases they share shelters with their animals[38]. Moreover, in Scotland it had been reported that the incidence of *M. bovis* infection in cattle herds has been increased since 2000 suggesting a similar rise in the incidence of *M. bovis* infection in humans [39].

5. Conclusion

The study showed that prevalence of BTB in cattle was low as compared to prevalence of BTB in intensive dairy farms in central Ethiopia. Even though the prevalence was low, high prevalence was found in herds that were managed under poor managerial inputs and in larger herd size as well as in exotic breeds than their crosses and Zebu breeds. The majority of cattle owners in the study area lack awareness about BTB and its public health significance; a large portion of the public had habit of drinking raw milk and eating raw meat implying the possible potential of acquiring BTB. Awareness rising of cattle owners about BTB and its transmission, and the zoonotic implication of BTB is of extreme importance for effective implementation of TB control measures.

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