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Prevalence and associated risk factors of bovine tuberculosis in ambo and Debre Berhan dairy farms, central Ethiopia

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Abstract
Bovine tuberculosis (BTB) is a chronic bacterial disease of animals and humans and is a major infectious disease among cattle, other domesticated animals, and certain wildlife populations in a which principally caused by *Mycobacterium bovis* and characterized by progressive development of tubercles in any tissue/organ of the body. A cross-sectional study on bovine tuberculosis was carried out in Debre Berhan and Ambo dairy farms from October 2013 to March 2014 to estimate the prevalence of Bovine tuberculosis. A total of 413 animals from 42 herds were subjected to comparative intradermal tuberculin test (CIDT). The result of CIDT indicates an overall prevalence of 7.02% (29/413) at individual animal level and prevalence of 26.19% (11/42) at herd level in both study area. Prevalence of 11.9% and 100% were recorded at individual animal level and herd level respectively in Ambo. Whereas in Debre Berhan the prevalence at individual animal level and herd level were 3.67% and 18.42% respectively. The prevalence of BTB within age group <2 year, 2-6 years, 6-10 years and ≥ 10 years was 1.33%, 10.41%, 4.44% and 3.70% respectively. This difference was statistically significant ($P< 0.05$). However there was no statistically significant difference ($P>0.05$) observed in prevalence of BTB between sex, farm size, breed and body condition of animals in the present study.

Keywords: Ambo, bovine tuberculosis, Debre Berhan, prevalence, tuberculin test

1. Introduction
Bovine tuberculosis (BTB) is a chronic bacterial disease of animals and humans and is a major infectious disease among cattle, other domesticated animals, and certain wildlife populations in a large number of countries [1]. This principally caused by *Mycobacterium bovis* and characterized by progressive development of tubercles in any tissue/organ of the body [2]. Several species of domestic animals and wildlife are susceptible to *M. bovis*, with cattle, goats and pigs being most susceptible, and sheep and horse having a high natural resistance [3, 4]. Even though cattle are considered to be the leading hosts of *M. bovis*, isolations have been made from various other livestock and wildlife species, and transmission to humans constitutes a community health problem [5, 6]. Aerosol exposure to *M. bovis* is considered to be the most common route of infection of cattle, but infection by ingestion of a contaminated material also occurs [7]. The standard technique for BTB detection in live animal is the comparative intradermal tuberculin test (CIDT) based on delayed hypersensitivity reactions. The CIDT test includes bovine and avian tuberculin and is used mainly to discriminate among animals infected with *M. bovis* and those sensitized to tuberculin due to exposure to other mycobacteria or related genera [6].
The incidence of BTB became uncommon in humans and cattle in developed nations [5]. However, it remains a significant disease in many developing countries where BTB is endemic, triggering substantial economic losses [8].
In Ethiopia, BTB is widespread in cattle. Prevalence varies from 3.5% to 50.0% depending on the geographical areas, the breeds, and the husbandry practices [9, 10]. Even though studies conducted so far have been incapable to establish the countrywide prevalence, mainly due to absence of adequate disease surveillance and diagnostic facilities, the rate of carcass condemnation due to tuberculosis has been growing over the past decade [11]. In addition to causing a high morbidity, BTB can also be a financial burden to farmers owning infected cattle; it has been advocated that cattle with BTB have a reduced productivity affecting milk yield and carcass value [12] as well as through reduced pulling power in traditional farming system [13].

BTB has also zoonotic potential [14] because of control measures are not applied or are applied sporadically, pasteurization practice is not adequate, consumption of raw milk/meat is a common practice, failure of culling of infected cattle herd, lack of awareness of its means of transmission, living of person with his/her animal in single house and also using infected cattle manure for plastering wall or floor and as source of energy for cooking can surges the chance of spread of tuberculosis as zoonosis in Ethiopia. It is estimated that in countries where pasteurization of milk is rare and bovine tuberculosis is common, 10 to 15% human cases of tuberculosis are caused by *M. bovis* [15].

For effective control, the basic information based on where *M. bovis* infection is present in a number of animal species in developing countries like Ethiopia [16] having the knowledge of distribution, prevalence, risk factors and zoonotic implication of the disease. Therefore the objectives of this study were to determine the prevalence of tuberculosis and associated risk factors in selected dairy farms at Ambo and Debra Berhan towns.

2. Materials and Methods

2.1 Study Area

The study was conducted in Debre Berhan and Ambo dairy farms. Debre Berhan town, East Shewa Zone of the Amhara Region, about 120 kilometers north east of Addis Ababa on the paved highway to Dessie, located at latitude and longitude of 9°41′N 39°32′E and an elevation of 2,840 meters above sea level. Ambo town is located in the West Shoa Zone, Oromia Region, 107 km West of Addis Ababa located at latitude and longitude of 8º 59'N 37º51'E and an elevation of 2101 meters. The maximum and minimum temperature of the area is 24 °C and 13 °C, respectively.

2.2 Study Design

A cross-sectional study was conducted from October 2013 to March 2014 to estimate the prevalence of Bovine tuberculosis in Debre Berhan and Ambo dairy farms.

2.3 Study population and Sampling method

After random selection of the farms, all individual cattle whose age greater than 6 months were sampled for the study and tested by Comparative Intradermal Tuberculin (CIDT) test. Sample size was calculated by using the formula given by [17], expected prevalence of 27.8% [18] at 95% confidence interval and 5% level of precision.

$$N = \frac{1.96^2 * P_{exp} (1-P_{exp})}{d^2}$$

Where P_{exp} = expected prevalence
d = absolute precision (5%)
n = sample size

Hence a total of 413 animals were sampled.

2.4 Method of Data Collection

2.4.1 Comparative Intradermal Tuberculin (CIDT)

All cattle older than six months, except clinically sick animals and cows one month pre-and post-partum, were tested by CIDT according to [6]. Two sites, 12 cm apart, horizontally on the mid neck of the animal were shaved and the skin thickness was measured with a caliper. Aliquots of 0.1 ml of 30000 IU/ml bovine purified protein derivative (PPD) (Bovitubal, strain AN5, Bioveta, Czech Republic) and 0.1 ml of 2500 IU/ml avian PPD (Avitubal, strain D 4 ER Bioveta Czech Republic) were injected separately into the respective shaved site. The thickness of the skin at each injection site was measured again 72 hours later. Results were interpreted according to [19] at cut-off ≥ 2 mm, if the difference between bovine PPD injection site and avian PPD injection site was greater or equal to 2 mm, the animal was considered as positive, while if the difference is less than 2 mm, the animal was considered as negative.

Data collection related risk factors on each tested cattle (such as sex, age, breed and body condition were collected and recorded at the time of the test. The body condition of each of the study animal was scored using the guidelines established by [20]. Accordingly, on the basis of observation of anatomical parts such as vertebral column, ribs, and spines, the study animals were classified as lean (score, 1 to 2), medium (3 to 4), or fat (greater than or equal to 5).

2.5 Data Analysis

All collected field data was entered into Microsoft-excel version 2010. To analyze the entered data STATA 11 software was used. Chi-square test ($\chi 2$) was computed to see the association of risk factors with that of test-positivity to tuberculosis.

3. Results

In this study out of 413 animals tested for bovine tuberculosis in Ambo and Debre Berhan dairy farms with CIDT, 29 animals were found to be positive giving an overall prevalence of 7.02%. The prevalence of BTB in Ambo and Debre Berhan were 11.90% (20/168) and 3.67% (9/245) respectively. This difference in prevalence of BTB in these two sites is statistically significant (P<0.05) (Table 1).

Table 1: Prevalence of Bovine tuberculosis in Ambo and Debre Berhan at individual animal level

Study sites	No. examined	No. positive	Prevalence (%)	P-value
Ambo	168	20	11.90	
Debre Berhan	245	9	3.67	0.001
Total	413	29	7.02	

Out of 42 herds examined in this study 11 were positive for BTB yielding a herd prevalence of 26.19% in Ambo and Debre Berhan. However the herd prevalence of 100% (4/4) and 18.42% (7/38) were recorded in Ambo and Debre Berhan dairy farms respectively (Table 2).

Table 2: Herd level prevalence of BTB in Ambo and Debre Berhan dairy farms

Study site	No. of herds	No. of positive	Prevalence (%)
Ambo	4	4	100.00
Debre Berhan	38	7	18.42
Total	42	11	26.19

Among risk factors studied in this study only age category was found to be statistically significant (P<0.05) and the prevalence was lower in animals less than two years of age and higher prevalence was recorded in animals from six to 10 years of age. However, there was no statistically significant difference among risk factors like sex, breed, body condition and farm size (Table 3).

Table 3: Prevalence of BTB and its associated risk factors in Ambo & Debre Berhan

Risk factors	Category	No. examined	No. positive	Prevalence (%)	P-value
Sex	Female	323	25	7.74	0.279
	Male	90	4	4.44	
Age	<2 years	75	1	1.33	0.030
	2≤x<6	221	23	10.41	
	6≤x<10	90	4	4.44	
	≥ 10 years	27	1	3.7	
Breed	Cross	371	27	7.28	0.545
	Local	42	2	4.76	
Body condition	Lean	60	3	5.00	0.738
	Medium	351	26	7.41	
	Fat	2	0	0	
Herd size	Small	144	5	3.47	0.086
	Medium	48	3	6.25	
	Large	221	21	9.50	
Total		413	29	7.02	

4. Discussion

In the current study, prevalence of BTB at individual animal level in Ambo and Debre Berhan dairy farms was 11.90% (20/168) and 3.67% (9/245) respectively. Higher prevalence of the disease was observed in Ambo (Table 1). The possible explanation for the higher prevalence in this study might be due to differences in ecological zones. Prevalence lower than the present study was reported by different workers 2% in Somali [21], 0.9% in Tanzania [22], 1.4% in Uganda [23] and 0.9% in Hamer [13]. Higher prevalence than the present study also reported by [24] in Eritrea (14.5%), by [25] in Wuchale-Jida, Ethiopia (46.8%), by [10] in Hawassa (11.6%) and by [26] in Addis Ababa and surrounding intensive dairy farms (30%). The herd level prevalence for Ambo and Debre Berhan was 100% (4/4) and 18.42% (7/38) respectively. The herd prevalence recorded in Ambo was higher when compared to Debre Berhan (Table 2). Herd level prevalence lower than Ambo were reported by other researchers, 48.7% in and surrounding Hawassa [10], 15% in Adama Town [27], 45.9% to 62.1% in central Ethiopia [28], 50% in intensive dairy farms in Addis Ababa and surrounding [26] and 43.4% in Addis Ababa [29]. High herd prevalence in Ambo could be due to intensive production system in the area. Several studies have also indicated that as herd size increases, the risk of cattle within the herd showing a positive reaction also increases [30, 41].

In current finding statistically significant association (P<0.05) was observed in the prevalence of BTB in relation to age group of study animals (Table 3). The prevalence of BTB increase with age and this finding agrees with previous reports [10, 32, 33] who reported that older animals had longer and repeated chance of exposure to mycobacterial infection during their life time.

Statistical analysis BTB on basis of body condition indicated no significant difference (p > 0.05) among body condition (Table 3). This fact agrees with the studies conducted by [13, 33, 34]; who reported that there was no association between body condition score and tuberculin skin test positivity.

There was no statistically significant difference (p>0.05) observed in the prevalence BTB with in the breed of the study animals (Table 3). This might be due to low proportion of local breeds in the study population. This finding is inconsistent with the result of [28, 35, 36, 37]; who reported that genetically improved cattle may suffer more severely from deficient housing and malnutrition and thus be more prone to infection than local breeds. There was no statistically significant difference (P>0.05) observed in the prevalence BTB in regard to the sex of study cattle, however [35, 38], reported that cows were more positive reactor than bulls.

In the present study there was no statistically significant difference (P>0.05) observed in the prevalence BTB in regard to the herd size (Table 3). This finding is inconsistent with the studies reported by [29] and other researchers [41, 39] who reported that the as herd size increases, the risk of cattle within the herd showing a positive reaction also increases.

5. Conclusion

This study has shown the occurrence and prevalence of BTB in these study areas even though it was higher in Ambo as compared to Debre Berhan at individual animal level as well as herd level. The prevalence of this disease was affected by age groups. Therefore emphasis should be given on controlling and preventing this disease especially in dairy intensification area through provision of separate barn for animals to reduce human exposure to the disease, and milk from positive cows should be use after boiling.

6. Conflict of interests

The authors declare that they have no conflict of interests.

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