



Study on The Prevalence of Bovine Babesiosis and its Associated Risk Factors in Cattle in and Around Dera Districts of Amhara Region, Northwest Ethiopia

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Abstract

Tick-borne hemoparasites are causing devastating losses to the livestock industry and thus pose major constraints to the livestock production throughout the world. A cross-sectional study using simple random sampling was conducted from January 2023 to March 2024 in Dera district of Amhara region of Northwest Ethiopia with the objectives to estimate prevalence and associated risk factors of bovine babesiosis in cattle in Dera districts. A total of 450 blood samples were collected from randomly selected cattle to assess the presence of *Babesia* species by using thin smear techniques in the study area. The overall prevalence of bovine Babesiosis was found to be 7.6%. In this study, only *Babesia bigemina* (7.6%) was encountered. The highest prevalence of bovine babesiosis was found in Goha and Dewol kebeles with a prevalence of 11.3% and 10.12% respectively. According to multivariable logistic regression analysis, sex, Body condition score, tick infestation, communal grazing land and communal watering points were identified as potential risk factors. In conclusion, currently low awareness of farmers about the diseases transmitted by ticks and resulting significant economic loss and increases occurrence of the diseases. In order to minimize losses attributed to bovine babesiosis in the area strategic tick control measures should be designed and implemented.

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Introduction

Background

Tick-borne diseases are economically important diseases of tropical and subtropical parts of the world [1]. They have a serious economic impact on livestock sector due to decreased productivity, lowered working efficiency, increased cost for control measures and limiting introduction of genetically improved cattle in the area and death of livestock [2].

Bovine babesiosis is a disease that commonly infects cattle, sheep, goats, horses, pigs, dogs and cats and occasionally man. *Babesia bovis* and *B. bigemina* are the main species affecting cattle widely distributed in tropical and subtropical countries which are responsible for high mortality rates up to 50% in susceptible herds and it's known to be transmitted in this country by *Rhipicephalus* [3].



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Animals suffering from acute babesiosis shows symptoms such as fever, oculo-nasal discharge, increased heart rate, increased respiratory rate, abnormal mucous membrane color, and low Packed Cell Volume (PCV) values. Although these symptoms are very typical, they are not pathognomonic and animals with chronic infections can be asymptomatic carriers [4].

Disease signs vary in severity from silent infection to acute circulatory shock with anemia, depending on susceptibility, immunity, and age of the host and *Babesia* species and parasite load and bovine *Babesia* is principally maintained by sub-clinically infected cattle that have recovered from disease [5].

Ticks are considered to be most important to the health of domestic animal in Ethiopia. Ticks comprise various types of genera, including *Amblyomma*, *Rhipicephalus*, *Haemaphysalis*, *Hyalomma* and *Rhipicephalus* (*Boophilus*). The genus *Amblyomma* and *Rhipicephalus* (*Boophilus*) are predominating in many parts of country [6].

The challenges of Tick-borne hemoparasites are growing steadily due to establishment of the tick vector in various geographical locations. In Ethiopia, anaplasmosis, babesiosis and theileriosis have been reported as major tick-borne diseases affecting domestic animals [7]. Environmental conditions and vegetation coverages determine the epidemiology for ticks and TBDs perpetuation [8]. The presence of tick-borne hemoparasitic disease broadly related to the presence and distribution of their vectors (ticks) in cattle [9].

Ticks are more prevalent in the warmer climates, especially in tropical and sub-tropical areas [10]. The seasonal variations within a bioclimatic zone may favour or hinder the development or activity of a tick species during certain periods [11]. Dry environmental conditions are a serious danger to ticks, particularly to the questing larvae, which are very susceptible to drying out fatally [12].

The importance of ticks is principally due to the ability to transmit a wide spectrum of pathogenic microorganisms, such as protozoa, Rickettsial, spirochetes, and viruses [13]. The main effect of tick infestation in cattle is mild to severe anemia, loss of appetite, leading to a reduction in growth rate and decreased productivity and tick infestation also results in increased calf mortality [14] (Mohsen et al., 2013). The seasonal variations within a bioclimatic zone may favor or hinder the development or activity of a tick species during certain periods [15]. Dry environmental conditions are a serious danger to ticks, particularly to the questing larvae, which are very susceptible to drying out fatally [12].

Although quite a lot of similar studies on bovine babesiosis in cattle have been conducted in different areas of Ethiopia, it is worth nothing that Ethiopia is a large country with a huge number of livestock populations, mostly cattle. Information regarding bovine babesiosis and its vectors (tick) in cattle in the study area is scarce. A study is required in the area to generate baseline information on bovine babesiosis and its vectors (ticks) for developing disease control and prevention programs. Therefore, the objectives of this study were to quantify the Prevalence of bovine babesiosis, associated risk factors and species identification of tick in cattle in the study area.

Statement of the problem

Babesia infections in cattle are economically important diseases of tropical and subtropical parts of the world. This dis-

ease causes economic loss towards livestock production and has a negative impact on food security, animal product and by products [16]. In Ethiopia, ticks and tick-borne diseases in cattle population cause serious economic loss to smallholder farmers, the tanning industry and the country as a whole through the mortality of infected animals, decreased production, down grading and rejection of hide [17].

Despite their important role in disease transmission, no research on these diseases has been conducted, and none have been enabled to analyze the association of distinct tick species with disease occurrence. Thus, there is paucity of information on cattle babesiosis in the districts of Dera. In order to address the above problems, a study is required to fill the gaps in knowledge about bovine babesiosis and its vectors in order to create baseline information that can be used to develop efficient disease control and prevention program. So, because of these problems the following research questions are formulated.

Research questions

This research work will attempt to answer the following research questions.

- ✓ What is the prevalence of bovine babesiosis in cattle in the study districts?
- ✓ What are the associated risk factors for the occurrence of bovine babesiosis in cattle?

Objectives of the study

General objectives:

The overall aim of this study is to quantify the Epidemiology of babesiosis in cattle in Dera districts.

Specific objectives:

The specific objectives of this study are:

- To estimate the prevalence of bovine babesiosis in cattle in the study area.
- To assess the potential factors associated with the occurrence of bovine babesiosis

Significance of the study

This study would be used to estimate the prevalence of bovine babesiosis and its associated risk factors in cattle in the study area. To update the required bodies about the important risk factors responsible for the occurrence of bovine babesiosis in cattle. The study would be promoted to future researchers to use the gap for further investigating the occurrence of this disease in cattle. This study would provide baseline information for those investigating the disease of cattle in Dera districts. Therefore, this study would facilitate zonal and regional animal health sectors used to designing and implementing effective control and prevention strategies of bovine babesiosis in cattle.

Materials and methods

Study area

The study was conducted in Dera districts in South Gondar administrative zone in Amhara regional state, North western Ethiopia from January 2023 to March 2024. It is located at latitude of 11°23' 15" - 11°53' 30" North and longitude of 37°25' 45" E - 37°54' 10" East with an area of 1525.24 square kilometers [18]. The district is bordered on the south by the Abbay

River which separates it from the West Gojjam Zone, on the west by Lake Tana, on the north by Fogera, on the northeast by East Este, and on the east by West Este. It is located 588 kilometers away from Addis Ababa, the capital city of the country, and 42 kilometers away from Bahir Dar, the capital city of the region [19].

The administrative center of the district is Ambesamie. The livestock populations of the district were 659276 (cattle 267063, sheep 97840, goat 54701, horse 1471, mule 2685, donkey 39434 and poultry 196082). While cattle serve as sources of drought power and milk, small ruminants (sheep and goats) are important cash sources. The altitude of the district ranges from 1560 - 2600 meters above sea level (m.a.s.l), with the district is characterized under Weyna Dega agro-ecological zone with an average rainfall ranging from 1000 - 1500 mm and its annual temperature ranges from 13 - 30 °C with flat land accounts for 51% and mountain and hills accounts for 49% [20].

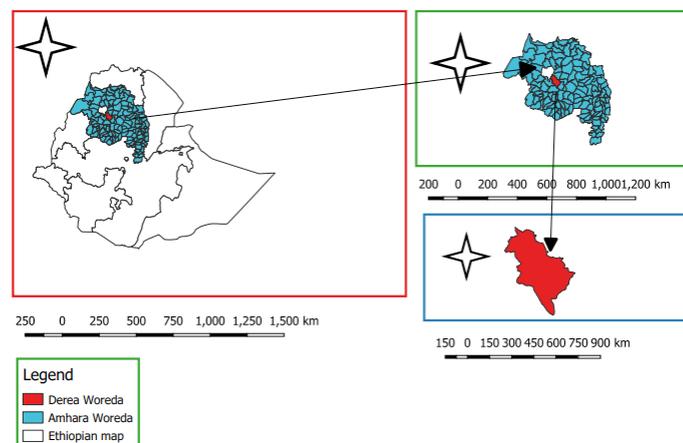


Figure 1: Maps of the study area.

Source: QGIS software version 3.22.6

Study design

A cross-sectional study was conducted from November 2023 to February 2024 to estimate the prevalence of bovine babesiosis, its associated risk factors and species identification of ticks in cattle in Dera district. Body condition scores of each cattle was evaluated during sample collection and the cattle was classified as emaciated (poor), moderate (medium) and good based on anatomical parts and the flesh and fat cover at different body parts [21] (Annex 1). Animals were conveniently classified as young (<3 years) and Adult (>3 years) age categories as described by [22] De-lahunta and Habel (1986).

Sampling method and sample size determination

The study districts were selected purposively based on their livestock population, agro ecology representation and accessibility. Simple random sampling techniques were used to select study kebeles, villages and animals.

Sample size was calculated according to the formula given by [23] with 95% confidence level and 5% absolute precision.

$$n = \frac{1.96^2 (P_{exp} (1 - P_{exp}))}{d^2} = \frac{1.96^2 (0.5 (1 - 0.5))}{0.05^2} = 384$$

Where, n = required sample size

P_{exp} = expected prevalence

d = desired absolute precision

However, to increase the absolute precision, 450 samples were taken throughout the study period.

Study population

The study was conducted on local and cross breed of cattle with different age, sex and body condition scores (BSCs).

Sample collection and examination

Blood sample collection and laboratory analysis:

A total of 450 blood samples will be collected from ear vein of simple randomly selected cattle from Dera districts following the standard protocol described by [24]. Briefly, after proper restraining of the animal, ear vein will be disinfected with alcohol (70%) and the hair around the intended area was shaved with scalpel blade followed by a slight tearing of the vein with lancet. Take drop of blood on a grease free clean slide and spread the blood by another clean slide at angle of 45° then dry it quickly and labeled. After labeling, it was transported to Dera district, Veterinary laboratory and to fix the slide with methyl alcohol for 2 minutes, dry and stain the slide with 0.76% Giemsa for 30 minutes. After staining wash the slide with distilled water till it assumes a bluish-purple color. Finally allow it to dry by standing upright on rack and examine under the microscope (X100).

Table 1: Proportional sampling allocations from the selected kebeles in the study district.

Districts	Kebeles (Villages)	Cattle population (S)	Sample size (n)
Dera	Huletu wogedami	11945	125
	Goha	8974	79
	Dewol	7259	71
	Ema shenkoro	6705	65
	Tebabari	6032	59
	Arbayitu	5235	51
Total		46150	450

Sources: (Dera district livestock resources and development of-fice, 2024); S: Cattle population, : sample size.

Blood sample examination:

Thin blood smear:

A thin blood smear was prepared by taking drops of blood and placed on frosted microscopic slides and spread by using other clean slides at an angle of 45°, air dried and fixed with methyl alcohol for 2 minute. Giemsa staining procedures and microscopic examination of the slides were conducted according to [25]. The slides were immersed in 0.76% Giemsa stain solution for 30 minutes according to [26]. The excess Giemsa solution were drained and washed using distilled water, allowed to dry by standing up right on the rack and examined under the microscope with oil immersion objective lens [27]. Fields from each stained slides were examined for identification of blood parasite at genus and species level [24].

Data analysis

The collected data was entered into Microsoft Excel, coded and summarized using descriptive statistics. The prevalence was calculated for all data by dividing positive samples over the total number of examined samples and multiplied by hundred. All statistical analyses was done using Stata 17 statistical software. Kebeles, breed, sex, age, body condition score, agroecology, tick infestation, health status, tick season occurrence, communal grazing land and communal watering point were the predictor variables used for statistical analysis. Univariable logistic

regression was used to assess if there is a statistically significant association between the occurrence of bovine babesiosis and potential risk factors. Statistical significance was considered to exist if p-value less than or equal 0.25. Correlation, confounding and interaction tests was checked. In the multivariable mixed effect logistic regression, P-value < 0.05 was considered as cut off for statistical significance and Odds Ratio (OR) and 95% CI were also calculated.

Results

Prevalence of bovine babesiosis at kebele level

A total of 450 blood samples were collected from ear vein and examined using a thin blood smear and an overall Prevalence of bovine babesiosis 32(7.62%) was recorded at 95% confidence interval in the study areas. Out of the total animals exposed to bovine babesiosis 10(8%), 8(10.12%), 8(11.3%), 3(4.6%), 3(5.08%) and 2(3.9%) were from Huletu wogedami, Goha, Dewol, Ema shenkoro, Tebabari and Arbayitu kebeles respectively.

Risk Factors for bovine babesiosis in cattle

In univariable logistic regression, the risk factors such as kebeles, breed, sex, age, body condition score; season, health sta-

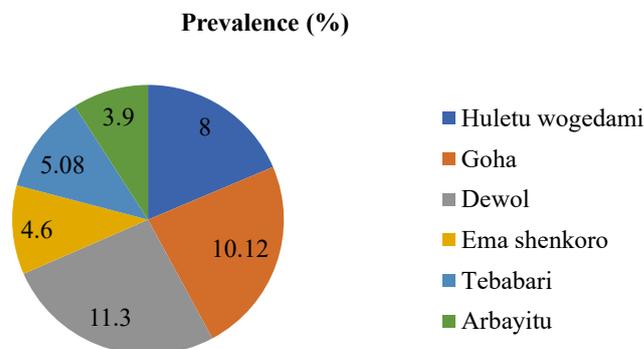


Figure 2: Prevalence of bovine babesiosis at kebele level in cattle.

tus, tick infestation, communal grazing land and communal watering points were analyzed. Among these factors sex, season, body condition score, tick infestation, communal grazing land and communal watering points were found to be statistically significant association with bovine babesiosis infection (P<0.25) effects on the occurrence of these infections. However, the factors considered in the initial univariable logistic regression analysis kebele, breed, age and health status were removed for multivariable logistic analysis in which p-value greater than 0.25.

Table 2: Univariable logistic regression analysis of risk factors associated with bovine babesiosis in cattle.

Variables	Categories	Number examined	Number positive (%)	OR (95%CI)	P-value
Kebeles	Huletu wogedami	125	10 (8)	Ref.	-
	Goha	79	8 (10.12)	1.30 (0.49 - 3.44)	0.603
	Dewol	71	8 (11.3)	1.46 (0.55 - 3.99)	0.45
	Ema shenkoro	65	3 (4.6)	0.56 (0.15 - 2.09)	0.386
	Tebabari	59	3 (5.08)	0.62 (0.16 - 2.33)	0.475
	Arbayitu	51	2 (3.9)	0.47 (0.09 - 2.22)	0.34
Breed	Cross	111	7(6.31)	Ref.	-
	Local	339	27 (7.96)	1.08 (0.48 - 2.47)	0.85
Sex	Male	158	10 (6.3)	Ref.	-
	Female	292	24 (8.22)	0.41 (0.204 - 0.84)	0.014
Age	Young	72	5 (6.9)	Ref.	-
	Adult	378	29 (7.7)	0.71 (0.30 - 1.71)	0.45
Season	Dry	213	10 (4.7)	Ref.	-
	Wet	237	24 (10.13)	1.70 (0.82-3.52)	0.15
BCS	Good	162	5 (3.08)	Ref.	-
	Medium	182	15 (8.24)	3.68 (1.34 - 10.10)	0.011
	Poor	107	14 (9.35)	3.24 (1.07 - 9.75)	0.037
Agro ecology	High land	151	8 (5.3)	Ref.	-
	Mid land	144	12 (8.33)	1.52 (0.65 - 3.54)	0.33
	Low land	155	14 (9.03)	0.96 (0.37 - 2.52)	0.94
Tick infestation	None infested	236	14 (5.93)	Ref.	-
	Infested	214	20 (9.35)	2.14 (1.03 - 4.44)	0.041
Health status	Sick	69	2 (2.9)	Ref.	-
	App healthy	381	32 (8.4)	1.95 (0.58 - 6.56)	0.281
Communal grazing land	Absent	167	7 (4.2)	Ref.	-
	Present	283	27 (9.54)	2.49 (1.01 - 6.16)	0.047
Communal watering points	Absent	160	4 (2.5)	Ref.	-
	Present	290	30 (10.3)	3.70 (1.40 - 9.75)	0.008

BCS: Body condition score; OR: Odd ratio; CI: Confidence interval; Ref.: Reference

Table 3: Final multivariable logistic regression model of factors associated with bovine babesiosis in cattle.

Variables	Categories	Number examined	Number positive (%)	OR (95%CI)	P-value
Sex	Male	158	10 (6.3)	Ref.	-
	Female	292	24 (8.22)	0.53 (0.16 - 0.74)	0.006
BCS	Good	162	5 (3.08)	Ref.	
	Medium	182	15 (8.24)	3.63 (1.35- 10.80)	0.012
	Poor	107	14 (9.35)	3.82 (1.16 - 11.36)	0.027
Tick infestation	None infested	236	14 (5.93)	Ref.	
	Infested	214	20 (9.35)	2.35 (1.02 - 5.40)	0.044
Communal grazing land	Absent	167	7 (4.2)	Ref.	
	Present	283	27 (9.54)	4.89 (1.82-13.09)	0.002
Communal watering points	Absent	160	4 (2.5)	Ref.	
	Present	290	30 (10.3)	3.95 (1.40-11.26)	0.010

For final mixed effect logistic regression model, factors with p-value <0.05 in the multivariable logistic regression analysis were included to fit the model. In the final multivariable logistic regression analysis; sex, body condition score, tick infestation, communal grazing land and communal watering points were found to be the potential risk factors for the occurrence bovine babesiosis in the study area.

The sex of study animals, the prevalence of bovine babesiosis in cattle was estimated to be higher in female animals (8.22%) compared to male animals (6.3%). The odd of occurrence of bovine babesiosis in female animals were 0.53 times more likely than in male animals and there was statistically significant difference between the two groups female and male animals (OR=0.53; CI=0.16 – 0.74; P=0.006) (table 3).

The prevalence of bovine babesiosis was significant (P=0.012) based on the body condition score of the study animals. The highest prevalence was recorded in animals with a poor body condition (9.35%) followed by medium body condition animals were (8.24%) and the lowest in animals with good body condition scores were (3.08%). According to multivariable logistic regression analysis, the odd of bovine babesiosis occurrence in poor and medium body condition score of animals were 3.82 and 3.63 times more likely than good body condition score of animals positive for bovine babesiosis respectively.

Regarding the prevalence of bovine babesiosis in tick-infested and non-tick-infested cattle the higher prevalence was found in tick-infested (9.35%) cattle than non-tick infested (5.93%) cattle. The risk of occurrence of bovine babesiosis in tick-infested cattle was 2.35 times more likely than non-tick infested cattle. This difference was statistically significant (OR=2.35; 95% CI=1.02- 5.40; P=0.044) (Table 3).

Animals on the communal grazing land (9.54%) had the highest prevalence of bovine babesiosis than animals no grazing on communal land (4.2%) animals, which were 4.89 times more likely to be positive for bovine babesiosis than animals no grazing on communal land. This difference was statistically significant (OR=4.89; 95% CI=1.82-13.09; P=0.002) (Table 3).

According to the prevalence of bovine babesiosis in communal grazing land (10.3%) had the highest prevalence of bovine babesiosis than animals no grazing on communal land (2.5%) animals, which were 3.95 times more likely to be positive for bovine babesiosis than animals no grazing on communal land. This difference was statistically significant (OR= 3.95; 95% CI=1.40-11.26; P = 0.010) (Table 3).

Discussion

In the present study, the overall prevalence of bovine babesiosis was found to be 7.6%. The present result of bovine babesiosis is in line with the finding of [7,28] who indicated 6.1%, 6.51% and 7% prevalence of bovine babesiosis in Southwestern Ethiopia and higher than [29,5,30] who reported 0.6%, 1.5% and 3.64% prevalence of bovine babesiosis in Central Ethiopia and Western Ethiopia respectively. This result was found to be low when compared to the result of [9,31,32,33] who reported 11.2%, 16.9%, 21.7% and 23% respectively in Ethiopia. This variation in the prevalence of bovine babesiosis could be due to the climatic factors which directly influence the vector distribution, animal husbandry practices and time of study.

In the present study, slightly higher infection was recorded in female animals 8.22% as compared to male animals 6.3%. This sex difference might be due to the fact that female cattle were under high level of hormonal disturbances, milk production, draught power and breeding system which pose it to weakened immune system [34].

In the present study, highest prevalence was recorded in poor body condition animals 9.35% followed by medium body condition animals 8.24% and least in good body condition animals (3.08%). This could be due to the fact that cattle with poor body condition have lower immunity which encourages infection of animals by *Babesia* infections. In addition, during this survey it was very common to see high burden of ectoparasite (ticks) on cattle with poor body condition and this can increase rate of infection from *Babesia*. With regard to the body condition, poor body condition cattle were highly infected with *Babesia*. Similar result had been reported by [35]. According to the reports of [36], poor body condition cattle harbor higher number of tick infestation. Because poor body conditioned animals had reduced resistance to tick infestation and they exposed tick infestation during grazing on the field than medium and good body conditioned animals.

The present study showed that higher prevalence was recorded in tick-infested (9.35%) cattle than non-infested (5.93%) cattle. This variation is due to the fact that ticks are very important vectors that transmit tick-borne hemoparasites in cattle during blood suckling from the infested host.

Animals having access of grazing on communal land was significantly affected and higher occurrence of *Babesia* infections (9.54%) than animals having no access of grazing on communal land (4.2%). This could be due to the reason that cattle having

access of grazing on communal land have high chance of contact with tick infected animals and tick vectors that helps the transmission of tick-borne hemoparasites from tick infested to tick non-infested animals.

In the present study, higher prevalence was recorded in animals having access of communal watering points (10.3%) than animals having no access of communal watering points (2.5%). This variation could be due to the reason that cattle having access of communal watering points having high chance of contact with tick infected animals and tick vectors that helps the transmission of tick-borne hemoparasites from tick infested to tick non-infested animals.

Conclusion and recommendations

The present findings indicated that bovine babesiosis had 7.6% of prevalence in the study area that might be due to abundance of tick infestation and presence of communal grazing land. Only *Babesia bigemina* was identified. The main risk factors found to be significantly associated with bovine babesiosis were sex, body condition score, tick infestation, communal grazing land, and communal watering points. In order to minimize losses attributed to bovine babesiosis in cattle in the area were strategic tick control measures should be implemented, as it is a level of control that prevents ticks from becoming a nuisance. In conclusion, bovine babesiosis are one of the most important cattle diseases in the study area.

Therefore, based on the above conclusion the following recommendations are forwarded.

- Further research works on molecular level should be carried out.
- The livestock sector should be played an important role in designing and implementing tick and babesiosis control and prevention programs.
- Strategic tick control measures should be carried out mainly during season of high tick occurrence in order to minimize losses attributed to babesiosis in cattle in the study area.
- The economic impact and effects of ticks and TBD on cattle productivity should be further studied at zonal and regional level.

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