American Journal of Bioscience and Bioengineering
2024, Vol. 12, No. 6, pp. 116-127
https://doi.org/10.11648/j.bio.20241206.13

Research Article

Seroprevalence and Associated Risk Factors of Contagious Bovine Pleuropneumonia in Selected Districts of South West Shoa Zone, Oromia, Ethiopia

Gemechis Biratu[1, *], Motuma Debelo[1], Tadale Tolosa[1], Walde Abdisa [1], Moti Wagari[2], Dasalegn Mardasa[2], Dagne Guta[2], Walde Abdisa[3]

[1]School of Veterinary Medicine, Jimma University, Jimma, Ethiopia
[2]Bedele Veterinary Regional Laboratory, Bedele, Ethiopia
[3]Veterinary Public Health, Jimma University, Jimma, Ethiopia

Abstract

Contagious bovine pleuropneumonia is highly contagious transboundary disease of cattle caused by *Mycoplasma mycoides* subspecies *mycoides Small Colony.* In Ethiopia, this disease possesses a significant threat to cattle production and impacts on export markets. Therefore, this study was designed to estimate the seroprevalence and assess its associated risk factors in selected districts of the Southwest Shoa zone. A cross-sectional study design with a simple random sampling technique was carried out from December 2022 to August 2023. For this study, a total of 384 sera samples were taken and tested using a competitive Enzyme-Linked Immunosorbent Assay. Information on risk factors influencing the occurrence of disease was collected using structured questionnaire survey. SPSS versions 26 for data analysis were used. The overall animal and herd level seroprevalence of contagious bovine pleuropneumonia in the study area was 13.5% (95% CI: 10.5–17.3) and 40.6% (95% CI: 31.3–50.6) respectively. Regarding risk factors, at the individual animal level; age (OR = 5.0, 95% CI: 2.2–11, P = 0.001), animal origin (OR = 4.3, 95% CI: 2.1–8.6, P = 0.001), and body condition (OR = 11.12, 95% CI: 4.5–27.4, P = 0.001); and at the herd level, only herd size (OR=38, 95% CI: 9.6-148.7, P= 0.001) was statistically significant at P = 0.001 with contagious bovine pleuropneumonia seroprevalence. In the present study, the result showed that the disease is prevalent and appears to be common in the study area. This suggests the disease could cause considerable economic losses through morbidity and mortality. Therefore, control measures should be implemented to prevent further spread of the disease through the use of better and more coordinated vaccination programs.

Keywords

Bovine, Competitive Enzyme-Linked Immunosorbent Assay, CBPP, Risk Factors, Seroprevalence, Southwest Shoa and Ethiopia

[*]Corresponding author: gemebira37@gmail.com (Gemechis Biratu)

Received: 6 January 2024; **Accepted:** 6 November 2024; **Published:** 28 November 2024

1. Introduction

Ethiopia has an estimated human population of over 114 million [32] and is an agricultural country employing almost 85% of the workforce and generating 40% of GDP [9]. Livestock is an integral part of agriculture, contributing about 45% of the total value of agricultural production [25]. Ethiopia ranked first largest in Africa and tenth in the world by livestock population [15]. Among livestock population cattle are used as the most source of draught power for the rural farming population [49]. However, due to different factors, this huge resource makes a disproportionate contribution to national income, Poor nutrition, low animal genetic potential for production traits, and widespread diseases are the main barriers contributing to the low productivity of local breeds [20, 21].

Diseases such as contagious bovine pleuropneumonia (CBPP) cause tremendous disruption to the country's livestock industry and livelihoods by affecting animal health and the production, quality, and availability of animal food [23,11]. According to Alhaji et al. [5], CBPP is one of the most important infectious diseases of cattle in Africa. The Pan African Program for the control of epizootics has already identified CBPP as the second most common transboundary disease in Africa after rinderpest [14].

Contagious bovine pleuropneumonia (CBPP) is an infectious and contagious respiratory disease of bovidae caused by Mycoplasma mycoides subsp. mycoides small colony (MmmSC) with a major impact on livestock production and a potential for rapid spread. As a result, CBPP-infected countries are excluded from international trade of live animals [40,43]. Infected animals show signs like anorexia, fever, dyspnea, polypnoea, cough, and nasal discharge [24, 50]. The main way of transmission is the inhalation of contaminated aerosol by agents from diseased animals. Outbreaks tend to be more widespread in confined and in those transported by train and on foot [41]. Issues such as age, stress, and coexisting disease may predispose to tissue invasion [36].

According to the World Animal Health Organization report in 20 African countries the highest number of cases of outbreaks of CBPP reported in Ethiopia and become one of the African countries where CBPP is causing a significant economic impact on cattle owners [1, 14]. The occurrence of such diseases impacts both extensive and intensive livestock producers by marginalizing them from higher-priced livestock markets and restricting their capacity for value-added trade [24].

In Ethiopia, field studies have shown that CBPP poses a major threat to cattle production in different parts of the country [26, 35]. Moreover, reports from various export quarantine centers in the country [10] seem to indicate a huge threat to the livestock export markets. The disease also affects investments in the livestock sector through direct losses (in terms of mortality and reduced milk, live weight, fertility, and traction) and indirectly through the cost of control measures and the resulting trade ban [2, 18].

As a result, this current study focused on estimating seroprevalence and identifying potential risk factors of CBPP, which are not well documented yet in the current study area. This study was conducted in selected districts of the Southwest Shoa zone (Goro, Wonchi, and Ilu). To prevent huge economic losses resulting from this infectious and transboundary disease, a laboratory-based study was followed, identifying its associated risk factors that can bring about a great change in economic loss. Therefore, based on these key statements stated above, the study was conducted with the following general and specific objectives:

1.1. General Objective

To study on status of CBPP infection in selected districts of Southwest Shoa zone of Oromia regional state, Ethiopia.

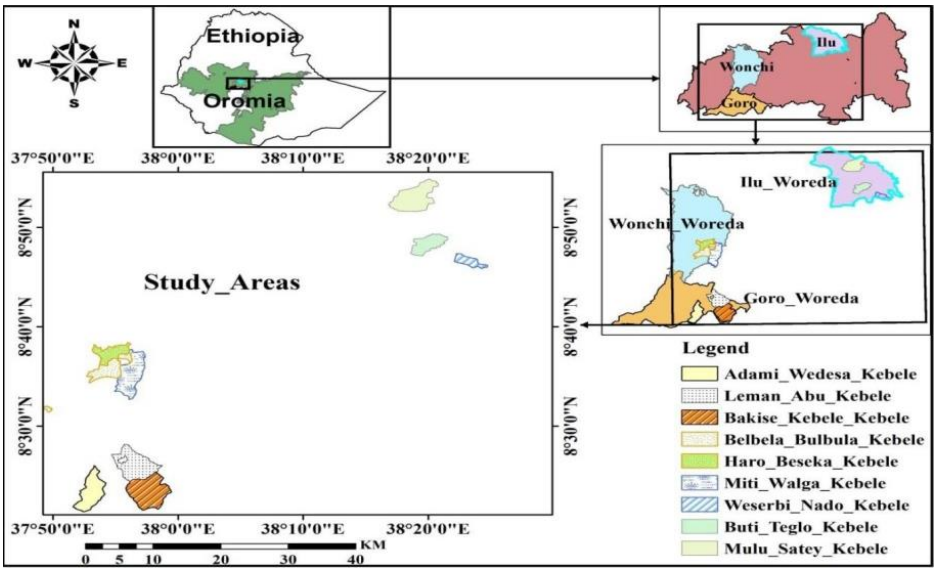
1.2. Specific Objectives

1) To estimate the seroprevalence of contagious bovine pleuropneumonia in the study area.
2) To assess the associated risk factors responsible for the occurrence of contagious bovine pleuropneumonia in the study area.

2. Materials and Methods

2.1. Description of the Study Area

Southwest Shoa zone is among the zonal administrations located in Oromia regional state, 114 km from the capital city of Ethiopia, Addis Ababa. The zone has a total population of 2,058,676, of whom 1,028,501 are men and 1,030,175 women; with an area of 14,788.78 square kilometers, while 242,352 or 6.10% are urban inhabitants, a further 53 individuals are pastoralists.



Source: GIS (Arc map)

Figure 1. *Map of the study area.*

Southwest Shoa is astronomically located at latitude and longitude of 08° 25' 56" N and 034°33'41" E respectively. The zone has two towns; Weliso and Dilala and it comprises 21 districts and 558 rural and urban kebeles. The elevation of the zone ranges from 880 to 3360 m. a. s. l. The mean annual rainfall of the area is estimated to be 1350 mm. The temperature of the zone varies between 18.7 ℃ and 27 ℃ [7].

2.2. Study Population

The target populations in this study included all local and cross-breed, cattle above six months of age of both sexes with no history of vaccination in selected peasant associations (PAs) of selected districts. Herds of cattle (a number of animals which are greater than or equal to two animals kept, fed, or traveling together) managed under the traditional extensive production system were included in this study and the study populations in this study were all cattle selected for this study.

2.3. Study Design

A cross-sectional study was conducted from December 2022 to August 2023 in three selected districts (Goro, Wonchi, and Illu) of the Southwest Shoa Zone. The study was supported by a questionnaire survey, and a serological test was carried out to determine the seroprevalence of contagious bovine pleuropneumonia and to assess potential risk factors that played a role in the existence of the disease in the cattle population in selected districts. A proportional sample size was considered according to their cattle population, and a simple random sampling technique was used to select the households from the list of households available at the respective districts and PA agricultural offices, individual study animals were selected randomly from the selected herds.

2.4. Sample Size Determination

Because of the unavailability of well-documented previous reports of seroprevalence of CBPP in the study area, an expected prevalence of 50% was considered to determine the sample size required for blood sample collection according to Thrusfield [47] formula, and a total of 384 animals were selected.

$$n = \frac{(1.96)^2 * Pexp\ (1-Pexp)}{d^2}$$

Where n = required sample size, Pexp = expected prevalence, and d = desired absolute precision (0.05). The estimated sample size for households from which animals were selected for blood sample collection was calculated by dividing the total sample size (n=384) by the number of animals sampled from each herd (6) given an estimated 64 households. However, due to the inclusion of households that have less than six cattle, the total sample size of herds was inflated to 96. Thus, depending on the population of cattle, 96 herds (35 from Goro, 33 from Wonchi, and 28 from Ilu districts) were chosen for this study.

Questionnaire Survey

During the study cattle sampling process, interviews were conducted with 96 households that were selected for sample collection. The questionnaire covered the owner's name, address, and gender identity as well as details about their animals, like herd contact, contact area between herds at watering points or grazing areas, livestock markets, the introduction of new animals, encountering diseases in their herds as in Annex 1. Afaan Oromo was used during a face-to-face

interview to deliver this questionnaire to animal owners.

Inclusion criteria:

Cattle above six months of age with no vaccination history of CBPP and volunteer owners to collect samples and also for the questionnaire survey.

Exclusion criteria:

Cattle less than six months of age those with a vaccination history of CBPP, and non-volunteer owners to collect samples and also for the questionnaire survey.

2.5. Sampling Methods

The Southwest Shoa Zone was selected because the status of CBPP has not been well documented in the area. Three districts were selected conveniently from the zone based on cattle population and accessibility. Since the population is homogeneous, equal numbers of peasant associations were considered from these three districts, and in each selected district, three peasant associations were selected randomly. Accordingly, Adami Wedesa, Leman Abu, and Bakise were selected from the Goro district; Balbala Bulbulo, Haro Basaka, and Meti Walga were selected from the Wonchi district; and Wasarbi Nado, Buti Talgo, and Mulo Satayi were selected from the Ilu district.

Households that have two animals in each selected PA were defined as herds, and lists of households were taken from the respective districts and PA agricultural offices. Then households were selected using a simple random sampling method, and the selected households were informed by animal health workers to provide their cattle for sampling purposes early in the morning before their animals were left in the field. Finally, from selected households or herds, individual animals were selected randomly.

From Goro, Wonchi, and Ilu districts, 140, 130, and 114 animals were selected respectively, as described in Annex 2 proportionally, and selected herds or households were described in Annex 6. In order to select animals from selected household, first tethered separately by objects and after that number was assigned to an animal by counting tethered animals either from right to left or from left to right and selected through a simple random sampling method, and animal information was recorded as described in (Annex 3). The maximum sample size of six cattle (i.e., the average number of cattle per household in the area) was sampled from each selected cattle herd (household that has animals) for the serum sample collection. Only six cattle were sampled randomly from households with more than six cattle. However, in the case of households with ≤ 6 cattle, two animals were sampled. Accordingly, proportional sample size producers for selected districts and selected PAs are described in (Annex 2).

Similarly, questionnaire surveys were carried out immediately during sample collection. Households or animal owners selected for the questionnaire survey were those households selected from nine PAs. Those selected simply randomly initially to select individual animals for blood sample collection were interviewed. After they were selected, they were interviewed during sampling by face-to-face interviews (with total participants of 96 animal owners).

2.6. Sample Collection

A pre-tested, structured questionnaire was used to collect information on factors influencing the occurrence of CBPP within or between herds by face-to-face interview. Data on sex, age, origin of animal, herd size, previous infection history, body condition scores of animals, and introduction of new animals and herd contacts were collected. Body condition scores of animals were scored according to Yosef et al. [51], and body condition scoring 1 was recorded as good body condition, and body condition scoring 2 was recorded as poor body condition (Annex 4). Age was categorized as young (<3 years old) and adult (3 years) [34].

After individual animal selection was carried out, following aseptic protocol, about 5–10 milliliters of blood were collected from each cattle's jugular vein using sterile vacutainer tubes and needles after the cattle had been restrained, and each sample was appropriately labeled, including all necessary information like owner name and potential risk factors (species of animal, sex, age, breed, and body condition [50].

Blood samples were collected once from the jugular veins of individual cattle and kept protected from sunlight in a slanting position for 6–8 hours. The serum was separated by centrifuge and manually, transferred to a sterile Criovial tube, stored at -20^{o}C, and tested using competitive ELISA at Bedele Veterinary Regional Laboratory to detect MmmSC antibodies based on the manufacturer's instructions for the c-ELISA kit CIRAD-UMR$_{15}$ (France). Competitive ELISA is a WOAH-prescribed test and can be used for official CBPP testing [42].

2.7. Laboratory Test

2.7.1. Competitive Enzyme-Linked Immunosorbent Assay (cELISA)

In this current study, competitive enzyme-linked immunosorbent assay (cELISA) tests were used based on a monoclonal anti-MmmSC antibody named Mab [50] and carried out as described in Annex 5. Microplates were coated with MmmSC purified lysate. Blood samples were tested by premixing with the specific monoclonal antibody Mab in a separate plate (“preplate”) and the content of the preplates was transferred into the coated microplate. Any MmmSC-specific antibodies present in the sample form an immune complex with MmmSC antigen coated on the microplate competing with Mab for the specific epitope. After washing away unbounded material, anti-mouse antibody-enzyme conjugates were added.

In the presence of an immune complex between MmmSC

antigen and antibodies from the sample, Mab cannot bind to its specific epitope, and the conjugate is blocked from binding to Mab. Conversely in the absence of MmmSC antibodies in the test sample, MAb can bind to its specific epitope and the conjugate is free to bind to Mab. Unbound conjugates were washed away and the enzyme-substrate Tetra methyl Benzedrine (TMB) was added. In the presence of an enzyme, the substrate is oxidized and develops a blue color becoming yellow after adding a stop solution. Subsequent color development is inversely proportional to the amount of anti-MmmSC antibodies in the test sample. The underside of the plate was wiped and the optical density (OD) of individual reactions was measured at 450 nm using a plate reader. The percentage inhibition (PI) value for each sample was calculated by the following formula.

$$PI = ((OD\ Mab - OD\ test\ serum)\ x100\%)/\ (OD\ Mab - OD\ conjugate)$$

Where OD Mab is the optical density of the monoclonal antibody; OD test serum is the optical density of the test serum; and OD conjugate is the optical density of the conjugate [50].

2.7.2. Interpretation of Result

Samples with a percentage of inhibition less than or equal to 40% were considered negative for the presence of MmmSC antibodies. Samples with a percentage of inhibition greater than 40% and less than 50% were considered doubtful, whereas samples with a percentage of inhibition greater than or equal to 50% were considered positive for the presence of MmmSC antibodies. The specificity and sensitivity of cELISA were 99.9% and 63.8%, respectively [50].

2.8. Data Analysis

Data obtained from both serological tests and questionnaire surveys were entered and stored in the Microsoft Excel spreadsheet program and analyzed using SPSS software version 26. The total seroprevalence of individual animals was calculated by dividing the number of c-ELISA-positive animals by the total number of animals tested, and herd prevalence was also calculated by dividing the number of herds that were positive by the total number of herds tested. The statistical significance of the difference in seroprevalence of CBPP across study districts and among peasant associations was tested by the chi-square (χ^2) test.

Initially, associations of seropositivity with risk factors (sex, age, breed, body condition, origin of the animal, and herd size) were screened using univariable logistic regression analysis, and variables with a p-value of less than or equal to 0.25 ($p \leq 0.25$) were subjected to multivariable logistic regression analysis. In all the analyses, a significance level of 0.05 ($p<0.05$) was considered to be statistically significant. The strength of the association between the risk factors and the occurrence of the disease was assessed using the odds ratio (OR).

2.9. Ethics Statement

Ethical approval was obtained from Jimma University College of Agriculture and Veterinary Medicine and also from the Animal Research Ethics and Review Committee. Permission was sought from the South West Shoa Zone, and informed consent was obtained from the individual's animal owner to take a sample from their cattle.

3. Results

3.1. Overall Sero-prevalence of CBPP

Out of 384 animals sampled, 52 (13.5%) were seropositive for the Mycoplasma mycoides subspecies Small Colony (MmmSC)-specific antibody. The overall animal-level seroprevalence of CBPP in the study area was 13.5% (95% CI: 10.5–17.3). From 96 cattle herds, 39 were seropositive for CBPP-specific antibodies. The overall herd-level seroprevalence of CBPP was 40.6% (95% CI: 31.3–50.6). In this study, different seroprevalence was recorded across the study locations, with the highest seroprevalence (16.7%, 95% CI: 10.9–24.6) observed in Ilu districts and the lowest seroprevalence (11.5%) was recorded in Wonchi district. Similarly, the highest seroprevalence (26.7%, 95% CI: 14.2–44.4) was observed in the Mulo Satayi peasant association, while the lowest seroprevalence (0%) was recorded in the Meti Welga peasant association. The Chi-square (χ^2) test showed that there was no statistically significant difference in the seroprevalence of CBPP among study districts; however, there was a statistically significant difference in the seroprevalence of CBPP among peasant associations (Table 1).

Table 1. *Seroprevalence of CBPP across study districts and Pas.*

Variables	Categories	No of cattle examined	No of tested positive	Prevalence % (95%CI)	χ^2 (p-value)
Districts	Goro	140	18	12.9(8.3– 19.4)	1.45 (0.484)
	Ilu	114	19	16.7(10.9-24.6)	

Variables	Categories	No of cattle examined	No of tested positive	Prevalence % (95%CI)	χ^2 (p-value)
	Wonchi	130	15	11.5(7.1 – 18.2)	
	Total	384	52		
	Adami Wadesa	43	2	4.7(1.3– 15.5)	
	Leman Abu	47	10	21.3(12 – 34.9)	
	Bakise	50	6	12(5.6 – 23.8)	
	Wasarbi Nado	42	9	21.4(11.7–35.9)	
PAs	Buti Telgo	42	2	4.8(1.3– 15.8)	25.27(0.001)
	Molo Satayi	30	8	26.7(14.2–44.4)	
	Balbala Bulbulo	48	11	22.9(13.3–36.5)	
	Haro Basaka	42	4	9.5(3.8 – 22.1)	
	Meti Welga	40	0	-	
Total		384	52	13.5(10.5-17.3)	

PAs = Peasant Associations

3.2. Risk Factors Analysis

3.2.1. Animal-Level Risk Factors Analysis

In this study, various animal-level seroprevalences were recorded across different potential risk factors like sex, age, breed, body condition, and origin of the animal. The findings of the present study showed that higher seroprevalence was observed in male animals (17.0%, 95%CI: 11-25.3) than it was observed in female animals (12.2%, 95%CI: 8.9-16.6). The higher seroprevalence was observed in adult age group (>3years) (20.5%, 95%CI: 15.5-26.5) than it was in young age group (<3years) (5.6%, 95%CI: 3.1-10) with P = 0.001. The seroprevalence observed in cross breed animals (33.3%, 95%CI: 12.1-64.6) was higher than it was in local breed animals (2.4%, 95%CI: 1.3-4.5). The seroprevalence observed in cattle with poor body condition score (<3) (24.7%, 95%CI: 19-31.5) was higher than seroprevalence observed in cattle with good body condition score (≥3) (3.5%, 95CI: 1.7-7) with P = 0.001 and the seroprevalence observed in animals from outside source (22.2%, 95%CI: 16.4-29.2) was higher than seroprevalence observed in animals within their herds (7.5%, 95%CI: 4.7-11.7) with P = 0.001.

Initially, these risk factors were screened at (p≤0.25) using univariable logistic regression analysis and this analysis revealed that age, breed, body condition, and origin of the animal were entered into multivariable logistic regression analysis (Table 2).

Table 2. *Univariable logistic regression analysis of risk factors with animal-level seroprevalence of CBPP.*

Varaibles	Categories	No of cattle examined	No tested positive	Prevalence % (95%CI)	OR (95%CI)	p-value
Sex	Female	278	34	12.2% (8.9-16.6)	1	
	Male	106	18	17.0% (11-25.3)	1.5 (0.8-2.7)	.226
Age	Young (<3yrs)	179	10	5.6% (3.1-10)	1	
	Adult (≥3yrs)	205	42	20.5% (15.5-26.5)	4.4 (2.1-9.0)	.001*
Breed	Local	375	49	13.1% (1.3-4.5)	1	
	Cross	9	3	33.3% (12.1-64.6)	0.3 (0.1- 0.2)	.097
Body Condition	Good (Score 1)	202	7	3.5% (1.7-7)	1	
	Poor (Score 2)	182	45	24.7% (19-31.5)	9.2 (4.0- 21)	.001*

Varaibles	Categories	No of cattle examined	No tested positive	Prevalence % (95%CI)	OR (95%CI)	p-value
Origin of the animal	Own herd	226	17	7.5% (4.7-11.7)	1	
	Outside source	158	35	22.2% (16.4-29.2)	3.5 (1.9- 6.5)	.001*

1 = Reference group; * = statically significant, CI = Confidence Interval; OR = Odd ratio; yrs = years

Finally, further analysis of risk factors was performed using multivariable logistic regression analysis. A multivariable logistic regression analysis of potential risk factors with seroprevalence of CBPP found that age, body condition, and animal origin had a statistically significant ($p<0.05$) association with seroprevalence of CBPP (Table 3).

Cattle in the adult age group (≥ 3 years) (OR = 5.0, 95% CI: 2.2–11, P = 0.001) were more than five times more likely to be seropositive for CBPP than cattle in the young age group (<3 years). Cattle with poor body condition score (OR = 11.12, 95% CI: 4.5–27.4, P = 0.001) were eleven times more likely to be affected by CBPP than cattle with a good body condition score, and animals comes from outside source (OR = 4.3, 95% CI: 2.1–8.6, P = 0.001) were four times more likely to be affected by CBPP than cattle within their herd of the study area (Table 3).

Table 3. *Multivariable logistic regression analysis of risk factors with animal-level seroprevalence of CBPP.*

Risk factors	Categories	Prevalence % (95%CI)	Adjusted OR (95% CI)	P-value
Age	Young (<3yrs)	5.6 % (3.1-10)	1	
	Adult (≥3yrs)	20.5 % (15.5-26.5)	5.0 (2.2- 11)	0.001*
Body condition	Good	3.5% (1.7-7)	1	
	Poor	24.7% (19-31.5)	11.12 (4.5- 27.4)	0.001*
Animal origin	Their own	7.5% (4.7-11.7)	1	
	From outside Source	22.2% (16.4-29.2)	4.3 (2.1- 8.6)	0.001*

1 = Reference group; * = statically significant, CI = Confidence Interval; OR = Odd ratio; yrs = years

3.2.2. Herd-Level Risk Factors Analysis

In this study, the risk factors that were considered at the herd level were herd size, contact (herd mix), contact areas, the introduction of new animals, the presence of livestock market activities, and the presence of disease in the herd. Herds with a history of contact with other herds (51.6%), CI: 39.4-63.6, have a higher prevalence than herds with no history of contact with other herds (20.6%, 95% CI: 10.3-36.8. The prevalence of CBPP in animals with history of new animal introduction (50%) (33/66) was higher than in herds with no history of new animal introduction (20%) (6/30). The prevalence of CBPP in animals in the areas where there were livestock market activities was 46.9% (31/66), which was higher than in herds or animals in areas where there were no livestock market activities (26.7%) (8/30).

Similarly, there was higher CBPP seroprevalence in the herd in which other diseases were more common than in the (45.7%) (32/70) herd in which no other diseases were common. Finally, there was a higher seroprevalence of CBPP in animals or herds contacted at both the watering and grazing points (41.4%) (29/70) than animals contacted at the watering point only (38.5%) (10/26). Results of risk factor analysis with herd-level seroprevalence showed that among the herd-level risk factors that were considered, only herd size had a statistically significant effect on the seropositivity of CBPP ($p< 0.05$). Large herds (OR = 38, 95% CI: 9.6–148.7, p = 0.001) were more than thirty-eight times more likely to be affected by CBPP than smaller herds (Table 4).

Table 4. Final herd-level risk factors analysis to CBPP seroprevalence (n=96).

Varaibles	Categories	No of herds examined	No tested positive	Prevalence % (95%CI)	Crude OR (95%CI)	Adjusted OR (95%CI)	p-value
Herd size	Small herd (≤ 6)	48	5	10.4% (4.5-22.2)	1		
	Large herd (>6)	48	34	70.8% (56.8-81.8)	20.9 (6.8-63.7)	38 (9.6-148.7)	0.001*
Herd contact	No	34	7	20.6% (10.3-36.8)	1		
	Yes	62	32	51.6% (39.4-63.6)	4.1 (1.6-10.8)	1.7 (0.07-39.9)	0.7
Introduction of new animals.	No	30	6	20% (9.5-37.3)	1		
	Yes	66	33	50% (38.3-61.7)	4 (1.4-11)	5.4 (0.18-166.9)	0.34
Livestock market activity	No	30	8	26.7% (24.6-57.7)	1		
	Yes	66	31	46.9% (29.9-53)	2.4 (0.9-6.3)	1.8 (0.4-7.7)	0.4
Encounter disease in the herd	No	26	7	26.9% (13.7-46.1)	1		
	Yes	70	32	45.7% (34.6-57.3)	2.3 (0.9-6)	2 (0.4-9.5)	0.4
Contact area	Watering	26	10	38.5% (22.4-57.5)	1		
	Water and grazing	70	29	41.4% (30.6-53.1)	1.1 (0.5-2.8)	1.3 (0.3-5.7)	0.7

1= Reference group; * = statically significant, CI = Confidence Interval; OR = Odd ratio

4. Discussion

The serological-based results of the present study showed that CBPP was a major cattle health problem in the study areas. The current finding (13.5%) is nearly similar to the result of various researchers, who reported a prevalence of 16.14% in selected districts of west Arsi zone [6], 13% in selected districts of the north Gondar zone [38], 12% in the Borana pastoral area using the CFT test and c-ELISA [3], 12% in the southern zone of the Tigray region of Ethiopia [46], and 14.3% in the Horo Guduru zone of the western Oromia region of Ethiopia [48], 14.6% in a selected district of East Wollega and West Showa zones, western Ethiopia [36]; 11.9% in southern part of Tigray tested using the CFT test conducted by Teklue et al. [46] and 11.6% in the Somali region [29].

However, the overall seroprevalence is by far lower than the previous findings reported by Ebisa et al. [22] from Amaro special districts in the southern part of Ethiopia (31.8%), Daniel et al. [16] from three districts of Western Oromia (28.5%), 39% in the Somali region [27], and 28% in the west Wollega zone [42].

On the other hand, the findings of this study were higher than the results reported by Alemayehu et al. [4] from bulls originated from Borena pastoral area of Southern Ethiopia (0.4%), Dele et al. [17] from the Export Quarantine Center of Adama (0.4%), Dereje and Shawul [19] in Bale zone (1.4%), Kassaye and Molla [31] in and around Adama (4%), Asmamaw [8] in Southern Ethiopia (6.14%), Mamo et al. [35] from Gimbo District, Southwest Ethiopia (8.1%), Biruhtesfa et al. [12] from Bishoftu and Export Oriented Feedlots around Adama using c-ELISA (8.7%), Schnier et al. [44] from Southwestern Kenya (9.7%), and Kassaye and Molla [30] from the Export Quarantine Center of Adama (9.5%).

The overall herd-level seroprevalence of CBPP in the current study was 40.6% (95% CI: 31.3–50.6), which was similar to the finding of Amenu *et al*. [6], who reported a seroprevalence of 43.24% in the selected districts of west Arsi zone. However, this result was higher than the previous report of Bonnet et al. [13], with 4.6% in the Ethiopian highlands. On the other hand, the finding was lower than the finding reported by Suleiman et al. [45] from agro-pastoral areas of Nigeria (54.7%), Mersha [36] from selected districts of East Wollega and West Shewa zones of the Oromia region (54%), and Molla et al. [38] from selected districts of North Gondar (66%).

The variation in these observed prevalence levels reported by various researchers might be due to the differences in the types of tests used, time of sample collection, and differences in agro ecological systems, herd size, breed susceptibility, production systems and contact patterns. In the current study, only the c-ELISA test with CIRAD-UMR15 (France) kit instruction was used to categorize cattle as CBPP seropositive or negative. This test was more sensitive for detecting cattle at the chronic stage, but it is less sensitive for detecting animals at the early stage of CBPP infection [39, 22].

Out of the predisposing risk factors at the animal level were; age, body condition, and animal origin with statistically significant (P<0.05) difference with seroprevalence of CBPP. The outcome of this research revealed that there was

significant variation in CBPP disease among the age groups. Higher seroprevalence was recorded in adult animals (20.5%) than in young animals (5.6%) (P = 0.001). This result agreed with Tola *et al*. [48] and Geresu *et al*. [28] who reported in adult animals (17.3%), in young animals (10.8%); and in adults (8.19%) and in young (1.9%) age categories with significant associations, respectively. In contrast, different studies reported insignificant associations, such as Yosef *et al*. [51], who reported an insignificant association between age groups in the Gimbo district of southwest Ethiopia, and Mersha [37] in young (25.5%) and adult (30.3%) age categories in selected districts of western Oromia.

Young animals are relatively more resistant than adult animals to Mmm SC infection [33], which may be explained by the fact that as age increases, the chance of exposure also increases. Additionally, the low prevalence of infection in young animals could be due to the decreased contact between them and other animals because young animals don't move long distances, and higher seroprevalence in adults might be associated with the fact that chronic stages of the disease are usually seen in adult cattle as age progresses (Olabode et al., 2013). In addition to the test used, which is more sensitive in detecting cattle with chronic stages than any other test, and it is more likely to miss individual animals at the early stage of infection [39].

Body condition is a potential risk factor assessed that had a statistically significant (P = 0.001) association with the seroprevalence of CBPP. There was a higher CBPP seroprevalence in cattle with poor body condition score (24.7%) than in cattle with good condition score (3.5%), with an overall seroprevalence. The present result agrees with the finding of Mamo *et al*. [35], which found higher seroprevalence in poor BCS (11.5%) than in good BCS (5.6%) but no significant difference. This result of the current study showed that animal body condition has an association with seropositive CBPP. This could be due to the weak immune response in poor body-conditioned cattle compared to good ones. Emaciation is one of the indications of the presence of an infection in the animal. Mostly, chronic carriers of CBPP animals became emaciated because of the clinical characteristics of the disease. Besides, animals with good body condition have relatively better immunological responses to the infectious agent than animals with poor body condition scores [41].

Animal origin also had a statistically significant (P<0.05) association with the seroprevalence of CBPP. The findings of this study disagreed with a study reported from the Bishoftu abattoir and export-oriented feedlots around Adama [12], in which a statistically significant association was absent in the occurrence of CBPP between origins. The reason for the significant difference in the present study may be due to the animals comes from outside of study area was transmitted and spread CBPP, in the study area than their own herds origions of animal. This current study result showed that animals that entered into their herds were play a great role in the transmission of CBPP, which is prevalent over time as infection is aerosally transmitted.

Finally, herd size had a statistically significant association (P<0.05) with the seroprevalence of CBPP in this study. The seroprevalence of the disease was higher in large herd sizes (70.8%, 95% CI: 56.8–81.8) than in smaller herd sizes (10.4%, 95% CI: 4.5–22.2). Large herd groups (OR = 38, 95% CI: 9.6–148.7, p = 0.001) were more than thirty-eight times more likely to be affected by CBPP compared to the small herd groups. This significant variation in the seroprevalence of the disease between the herd sizes was in agreement with the findings of the study conducted on bulls originating from the Borena pastoral area [4]. This significant difference in this study might be related to the health management of large herds and the increased risks of an individual animal becoming infected with the disease as herd size increases due to overcrowding of animals and contagious nature of the disease.

5. Conclusion and Recommendations

The seroprevalence of CBPP out of 384 sampled animals, 52 cattle were seropositive and this study indicated that the disease was endemic in the study area. An overall 13.5% seroprevalence was recorded using c-ELISA test and of which, 16.7% recorded from Ilu, 12.9% from Goro district and 11.5% fromWonchi district. The potential risk factors like location (PAs), age, body condition score, Animals origion and herd sizes were statistically significant effect on seroprevalence of the CBPP. Similarly, significantly the highest seroprevalence was observed in the Mulo Satayi (26.7%), while the lowest seroprevalence (0%) was recorded in the Meti Welga peasant association. Animals kept in these study areas are always at risk of exposed to CBPP because of the uncontrolled replacement of animals from outside origin and body condition-related problems. The presence of statistically significant differences in the seroprevalence of CBPP among the above mentioned predisposing risk factors at individual animal and herd level were favor the occurrence and spread of the disease in the current study area. The serological test used in this study was can resulted in misleading in case of individual animal. Thus, the use of an additional test should be considered in future studies.

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

1) Further investigation in wide geographical areas and with a large sample size using reliable tools like molecular techniques and biochemical tests is needed to know the true picture of the disease.
2) All stakeholders, including animal owners, veterinarians, and the government should work together to successfully implement control measures in this area.
3) It need to develop schemes and implement control measures directed at preventing further spread and lowering the prevalence of the disease in the zone through

the use of better and coordinated therapeutic and vaccination programs.

4) Special attention for management of animals with poor body condition and also for aged animals should be adapted since they were more affected compared to others.

Abbreviations

CBPP	Contagious Bovine Pleuropneumonia
N	Required Sample size
Pexp	Expected Prevalence
D	Desired Absolute Precision

Conflicts of Interest

The authors declare no conflicts of interest.

References

[1] Abdela, N. and Yune, N., 2017. Seroprevalence and distribution of contagious bovine pleuropneumonia in Ethiopia: update and critical analysis of 20 years (1996–2016) reports. *Frontiers in veterinary science*, *4*, 100.

[2] Abera, Z., Mengistu, D. Batu, G. and Wakgari, M., 2016. Review on contagious bovine pleuropneumonia and its economic impacts. *Acad. J. Anim. Dis., 5*(1), 01-15.

[3] Ahmed, I., 2004. Epidemiological study of contagious bovine pleuropneumonia in Borana pastoral areas using complement fixation test and competitive enzyme-linked immunosorbent assay. *Unpublished MSc thesis, Addis Ababa University*.

[4] Alemayehu, G., Leta, S. and Hailu, B., 2014. Low seroprevalence of Contagious Bovine Pleuropneumonia (CBPP) in bulls originated from the Borena pastoral area of Southern Ethiopia. *Tropical animal health and production*, *47*(5): 983-987.

[5] Alhaji, N. B., Ankeli, P. I., Ikpa, L. T. and Babalobi, O., 2020. Contagious Bovine Pleuropneumonia: Challenges and prospects regarding diagnosis and control strategies in Africa. *Veterinary Medicine: Research and Reports*, pp. 71-85.

[6] Amenu, M., Bedu, H., Neggasa, T. and Benti, T., 2022. Sero Prevalence, Associated Risk Factors and Knowledge, Attitude, and Practice of Farmers towards the Contagious Bovine Pleuro Pneumonia in Selected Districts of West Arsi Zone of Oromia Regional State.

[7] Ararsa Derara, K. D. and Molla, M., 2018. Assessment of Livestock Feed Resources in Weliso District South West Shoa Zone Central Ethiopia. *American-Eurasian Journal of Agriculture and Environment Science*, *18*(3), 145-167.

[8] Asmamaw, M., 2003. The situation of CBPP in the selected district of Southern Ethiopia. MSc thesis, Faculty of Veterinary Medicine, Addis Ababa University, Debrezeit, Ethiopia.

[9] Asresie, A., and Zemedu, L., 2015. Contribution of livestock sector in Ethiopian economy: A review. *Adv. Life. Sci. Technol.*, *29*, 79-90.

[10] Atnafie, B., Goba, H., Sorri, H., and Kasaye, S., 2015. Seroprevalence of contagious bovine pleuropneumonia in abattoirs at Bishoftu and export-oriented feedlots around Adama. *Glob. Vet*., *15*, 321-324.

[11] Behnke, R., 2010. The contribution of livestock to the economies of IGAD member states study findings, application of the methodology in Ethiopia, and recommendations for further Work. In: *IGAD LPI working*, Great Wolford, UK: Odessa Centre, IGAD Livestock Policy Initiative, Pp. 2-10.

[12] Biruhtesfa, A., Henok, G., Hundera, S. and Surafel, K., 2015. Seroprevalence of contagious bovine pleuropneumonia in abattoirs at Bishoftu and export-oriented feedlots around Adama, Ethiopia. *Global Veterinaria*, *15*(3): 321-324.

[13] Bonnet, P., Lesnoff, M., Thiaucourt, F., Workalemahu, A. and Kifle, D., 2005. Seroprevalence of contagious bovine pleuropneumonia in Ethiopian highlands (West Wollega zone, Boji District. *Ethiopian Veterinary Journal*, *9*(2): 85-93.

[14] Constable, P. D., Hinchcliff, K. W., Done, S. H., and Grunberg, W., 2017. Veterinary Medicine: a textbook of the diseases of cattle, horses, sheep, pigs, and goats. 11th ed. Elsevier Ltd.

[15] CSA, 2021. Agricultural Sample Survey. Report on Livestock and Livestock Characteristics (Volume II). *Statistical Bulletin 589*. Central Statistical Agency (CSA). Addis Ababa, Ethiopia.

[16] Daniel, G., Abdurahaman, M., Tuli, G., and Deresa, B., 2016. Contagious bovine pleuropneumonia: seroprevalence and risk factors in Western Oromia, Ethiopia. *J. Vet. Res*., *83*, 1-5.

[17] Dele, E., Afera, B., Kebede, E., Awol, N., and Hadush, B., 2014. Seroprevalence of trade hampering livestock diseases in animals originated from Borana at export quarantine centers in Adama, Central Ethiopia. *Afr. J. Bas. Appl. Sci*., *6*, 30-36.

[18] Demil, E., 2017. Review on the economic impact of contagious bovine pleuropneumonia (CBPP). *Acad. J. Anim. Dis., 6*(2), 51-56.

[19] Dereje, L., and Shawul, W., 2017. A Sero-prevalence study of contagious bovine pleuropneumonia (CBPP) in Bale zone, Asella Regional Veterinary Laboratory, Asella, Ethiopia. *Acad. J. Anim. Dis.*, 6(3): 83-87.

[20] Duguma, B., Kechero, Y., and Janssens, G. P. J., 2012a. Productive and reproductive performance of Zebu × Holstein-Friesian crossbred dairy cows in Jimma town, Oromia, Ethiopia. *Glob. Vet.*, *8*, 67-72.

[21] Duguma, B., Kechero, Y., Janssens, G. P. J., 2012b. Survey of major diseases affecting dairy cattle in Jimma town, Oromia, Ethiopia. *Glob Vet*., *8*, 62-66.

[22] Ebisa, T., Hirpa, H., and Aklilu, F., 2015. Study on seroprevalence and risk factors CBPP in SNNPRS in Amaro special district. *Sci. Technol. Arts Res. J*., *4*, 106-112.

[23] FAO, 2002. "Improved animal health for poverty reduction and sustainable livelihoods," *FAO, Animal Production and Health*, *153*.

[24] FAO, 2004. Animal production and health Proceedings. FAO, 00100 Rome, Italy. Gebremedhin, E. Z., Agonafir, A., Tessema, T. S., Tilahun, G., Medhin, G., Vitale, M.

[25] FAO, 2019. The future of livestock in Ethiopia. Opportunities and challenges in the face of uncertainty. Rome. 48 pp. License: CC BY-NC-SA 3.0 IGO.

[26] Fulasa, A., Teshome, I., Bulto, A. O., Lakew, M., Tadesse, B., Benti, T., Tamiru, M. and Dessalegn, E., 2020. Seroprevalence, isolation, and associated risk factors of Contagious Bovine Pleuropneumonia at Bako Tibe and Ilu Galan Districts of West Shoa Zone, Western Ethiopia. *J. Anim. Res. Vet. Sci., 4*, 028. https://doi.org/10.24966/ARVS- 3751/100028

[27] Gedlu, M. G., 2004. Serological, clinical, and participatory epidemiological survey of CBPP in Somali Region. *Unpublished MSc thesis, Addis Ababa University, Addis Ababa University, Debrezeit Ethiopia. Pp.*, *75*.

[28] Geresu, M. A., Kedir, K., Birhanu, D., and Teshome, A., 2017. Seroepidemiological investigation and risk factors for contagious bovine pleuropneumonia infection of cattle in Dello Mena and Sawena Districts of Bale Zone, South Eastern Ethiopia. *J. Public Health Epidemiol*., *9*, 122-132.

[29] Gizaw, G. M., 2004. Serological, clinical, and participatory epidemiological survey of contagious bovine pleuropneumonia in the Somali region, Ethiopia. *MVE thesis. Faculty of Veterinary Medicine, University of Addis Ababa, Addis Ababa.*

[30] Kassaye, D. and, Molla, W., 2013. Seroprevalence of contagious bovine pleuropneumonia at export quarantine centers in Adama, Ethiopia. *Trop. Anim. Hlth. Prod*., *45*, 275-279.

[31] Kassaye, D., and Molla, W., 2012. Seroprevalence of contagious bovine pleuropneumonia at export quarantine centers in and around Adama, Ethiopia. *Trop. Anim. Hlth. Prod.*, *45*(1): 275-279.

[32] Kebede, W., Abebe, R. and Harito, J. B., 2022. Sero epidemiology of Contagious Bovine Pleuropneumonia in the Bench-Maji Zone, southwest Ethiopia. *Ethiopian Veterinary Journal*, *26*(1), 30-48.

[33] Lesnoff, M., Laval, G., Bonnet, P., Chalvet-Monfray, K., Lancelot, R. and Thiaucourt, F., 2004. A mathematical model of the effects of chronic carriers on the within-herd spread of contagious bovine pleuropneumonia in an African mixed crop-livestock system. Prev. Vet. Med., *62*: 101–170.

[34] Malicha, G., Alemu, S., Aklilu, F. and Abraha, A., 2017. Study of seroprevalence and associated risk factors of contagious bovine pleuropneumonia in Sidama Zone, Southern Ethiopia. *J. Vet. Sci. Technol*, *8*(5).

[35] Mamo, Y., Bitew, M., Teklemariam, T., Soma, M., Gebre, D., Abera, T., Benti, T. and Deneke, Y., 2018. Contagious Bovine Pleuropneumonia: Seroprevalence and Risk Factors in Gimbo District, Southwest Ethiopia. Hind. Vet. Med. Int., 1-6.

[36] Mersha T., 2017. Epidemiological Study on Contagious Bovine Pleuropneumonia and Farmers Knowledge, Attitude And Practice Towards The Disease In Selected District of East Wollega And West Showa Zones, Western Ethiopia (MVSc. Thesis), Bishoftu, Ethiopia.

[37] Mersha, T., 2016. Seroprevalence of CBPP and its potential risk factors in selected sites of Western Oromia, Ethiopia. *Ethiopian Veterinary Journal*, *20*(2), pp. 31-41.

[38] Molla, W., Jemberu, W. T., Mekonnen, S. A., Tuli, G. and Almaw, G., 2021. Seroprevalence and Risk Factors of Contagious Bovine Pleuropneumonia in Selected Districts of North Gondar Zone, Ethiopia. *Frontiers in Veterinary Science*, *8*, p. 626253.

[39] Muuka, G., Hangombe, B. M., Nalubamba, K. S., Kabilika, S., Mwambazi, L. and Muma, J. B., 2011. Comparison of complement fixation test, competitive ELISA and LppQ ELISA with postmortem findings in the diagnosis of contagious bovine pleuropneumonia (CBPP). *Tropical Animal Health and Production*, *43*, pp. 1057-1062.

[40] Olabode, H. O. K., Mailafia, S., Adah, B. M. J., Nafarnda, W. D., Ikpa, L. T., Jambalang, A. R. and Bello, R. H., 2013. Serological Evidence of Contagious Bovine Pleuro-Pneumonia antibodies in trade cattle (*Bos indicus*) sold in Kwara state-Nigeria. *Online International Journal of Microbiology Research*, *1*(1): 14-19.

[41] Radostits, O. M., Gay, C. C, Hinchcliff, K. W. and Constable, P. D., 2007. Veterinary Medicine, a Textbook of the Diseases of Cattle, Sheep, Pigs, Goats and Horses, 10th ed., (Sounders Elsevier, Spain), 1131-1135.

[42] Regassa, F., 2001. Herd prevalence of contagious bovine pleuropneumonia (CBPP), bovine tuberculosis, and dictyocaulosis in Bodji woreda, West Wellega. Addis Ababa University, Faculty of Veterinary Medicine, Debrezeit, Ethiopia, DVM thesis.

[43] Saxmose N., S., Álvarez, J., Bicout, D. J., Calistri, P., Canali, E., Drewe, J. A., Garin Bastuji, B., Gonzales Rojas, J. L., Gortázar, C., Herskin, M. and Michel, V., 2022. Assessment of the control measures for category diseases of Animal Health Law: Contagious Bovine Pleuropneumonia. *EFSA Journal*.

[44] Schnier, C., Mtui-Malamsha, N. J., Cleaveland, S., Kiara, H., Grace, D., McKeever, D. J. and Zadoks, R., 2006, August. CBPP seroprevalence and associated risk factors in the Maasai ecosystem of south-western Kenya. In *Proceedings of the 11th International Symposium on Veterinary Epidemiology and Economics* (pp. 150-165). Australia: Cairns.

[45] Suleiman, A., Bello, M., Dzikwi, A. A., Talba, A. M., Grema, H. A. and Geidam, Y. A., 2015. Serological prevalence of contagious bovine pleuropneumonia in agro-pastoral areas of Nigeria. *Tropical animal health and production*, *47*(6): 1033-1042.

[46] Teklue, T., Tesfaye, T., Nirayo, T., Hailu, B., Wayu, S., and Atsbha, T., 2015. Epidemiological status of CBPP in southern Zone of Tigray, Northern Ethiopia. *Anim. Vet. Sci*., *3*, 32-36.

[47] Thrusfield, 2007. "Veterinary epidemiology," in *Black well Science Ltd*, London, 3rd edition, pp. 46–65.

[48] Tola, E. H., Mosisa, T. and Kebede, A., 2021. Seroprevalence of Contagious Bovine Pleuropneumonia and Assessments of Community Knowledge, Attitudes & Practices in Western Oromia, Ethiopia.

[49] Tonamo, A., 2016. A review on cattle husbandry practices in Ethiopia. *Intern. J. Livestock Prod.*, *7*: 5-11.

[50] WOAH, 2021. "Manual of diagnostic tests and vaccines for terrestrial animals," in *Chapter 2.4.9. Contagious bovine Pleuropneumonia*.

[51] Yosef M., Molalegne B., Tsegaye T., Murga S., Debebe G., Temesgen A., Tefera B., and Yosef D., 2018. Contagious Bovine Pleuropneumonia: Seroprevalence and Risk Factors in Gimbo District, Southwest Ethiopia. Hindawi Veterinary Medicine International.