



Contagious Bovine Pleuropneumonia: Sero-Prevalence and Associated Risk Factors in Cattle in Woliso, Dawo and Ameya Districts of Southwest Shewa Zone of Oromia, Ethiopia

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Abstract

Contagious bovine pleuropneumonia (CBPP) is a highly contagious disease of cattle caused by *Mycoplasma mycoides* subspecies *mycoides* small colony. It is one of the most important threats to cattle-raising and trade in Africa including Ethiopia. A cross sectional study was conducted on 384 randomly selected cattle from 96 cattle herds selected from herds of cattle managed under extensive production system in three selected districts (Woliso, Dawo and Ameya) of Southwest Shewa zone of Oromia Regional State with the aim of estimating the sero-prevalence of contagious bovine pleuropneumonia and to assess the risk factors associated with its occurrence. Competitive Enzyme Linked Immunosorbent Assay test was used to identify 384 cattle sera for *Mycoplasma mycoides* subspecies *mycoides* small colony specific antibodies. The sero-prevalence of CBPP was calculated as the number of sero-positive samples divided by the total number of samples tested. The association between risk factors and sero-prevalence of contagious bovine pleuropneumonia was computed using multivariable logistic regression analysis. Animal-level and herd-level overall sero-prevalence of contagious bovine pleuropneumonia were 9.4 % (95%CI: 6.5-12.3) and 32.3 % (95%CI: 22.9-41.6) respectively. Among predisposing risk factors assessed; age (OR = 4.3, 95%CI: 1.8-10.1, P < 0.05), body condition score (OR = 3.4, 95% CI: 1.3-9.0, P < 0.05) and herd size (OR = 7.5, 95% CI: 2.7-21.2, p < 0.05) were significantly associated with the sero-prevalence of contagious bovine pleuropneumonia. This study result indicates a need for intervening and implementing control measures to prevent further spread of the disease in the study area. Therefore, removing or culling older animal, animal with poor body condition and technical support for larger herd owners may help in reducing the prevalence of the disease in the study area until other control stage designed.

Keywords: Ameya; Cattle; Contagious Bovine Pleuropneumonia; Dawo; Ethiopia; Seroprevalence; Woliso

Introduction

Ethiopia is a leading country in the number of livestock population in the African continent with an estimated 59.5 million cattle, 30.7 million sheep, 30.2 million goats and 56.53 million poultry [1]. The Livestock sector plays important role in socioeconomic activities of the country and contributes much to the national economy [2]. However, the output of this sector in terms of its contributions to the improvement of the livelihood of animal owners and for the growth of national economy is at a lower stage compared to the

vast resource on hand [3,4]. The major constraints contributing to low productivity include low genetic potential of the animals, poor nutrition and prevailing animal diseases [5,6]. Among the health constraints, transboundary animal diseases such as Contagious bovine pleuropneumonia (CBPP) cause the major limitation to the livestock sector development in the country and affect livelihood through their impact on animal health, animal food production, availability and quality [6].

Contagious bovine pleuropneumonia (CBPP) is highly contagious disease of cattle caused by *Mycoplasma mycoides subspecies mycoides* Small Colony (*MmmSC*). Under natural conditions, it affects only domestic ruminants of the genus *Bos* [7], mainly *Bos taurus* and *Bos indicus* [8]. The disease is manifested by anorexia, fever and respiratory signs such as dyspnea, polypnea, cough and nasal discharges in cattle [9]. The principal route of infection is inhalation of infective droplets of diseased animals. Factors such as age, stress and concurrent infections may predispose to tissue invasion [10] and other risk factors for its spread include high-density confinement in night housings and mixing of herds at common grasslands and watering places [11]. Due to its high economic impact, OIE declared as one of the most serious contagious animal disease and listed on the group of notifiable animal diseases of high socio-economic impact and regarded as major animal disease [6].

Contagious bovine pleuropneumonia (CBPP) has a world-wide distribution because of increased international trade in live cattle [12]. Although CBPP was once found worldwide, it was eradicated from many countries through the application of restrictions to the movement of cattle, as well as test and slaughter policies combined with compensation for livestock owners [13]. However, the disease remains endemic in Africa particularly in tropical and subtropical regions (West, central, east and parts of southern Africa) of the continent [11,12,14] due to the economic and financial difficulties to prevent and control and becomes one of the major constraints to cattle-raising and trade in African countries [15].

Ethiopia is one of east African countries in which CBPP is endemically maintained in various parts of the country [2]. After rinderpest was eradicated, CBPP has become the most important cattle disease that hinders livestock development in the country. This is mainly caused due to the interruption of the consecutive yearly blanket vaccinations with combined rinderpest [10]. It is now re-emerging as one of the most economically important diseases that impede livestock production and remain a threat to livestock export potential in the country [16].

Previously CBPP was thought to be a problem of lowland pastoral areas with incursion to the adjacent highland but in recent years its incidence in the highland areas has raised [16] and reported by different researchers as one of the major threats to cattle production in mixed farming communities [9,17]. In Southwest Shewa zone, where mixed farming is the mainstay of the communities, animal disease is one of the major limiting factors to livestock production and field report showed that various livestock diseases like pasteurellosis and foot and mouth disease (FMD) were con-

sidered to be the major diseases affecting cattle production in this areas [18]. These diseases share closely similar clinical signs with contagious bovine pleuropneumonia and it might be confused with this disease when diagnosed tentatively.

Despite of the above facts; no previous investigation has been carried out to determine the status of CBPP in Southwest Shewa zone in general and in Woliso, Dawo and Ameya districts in particular. Therefore, this study was designed with the objective of investigating the sero-prevalence of contagious bovine pleuropneumonia and its associated risk factors in these selected districts of the zone.

Materials and Methods

Description of the study area

The study was conducted from November 2018 until August 2019 in Woliso, Dawo and Ameya districts which were conveniently selected from Southwest Shewa zone of Oromia regional state, central Ethiopia (Figure 1).

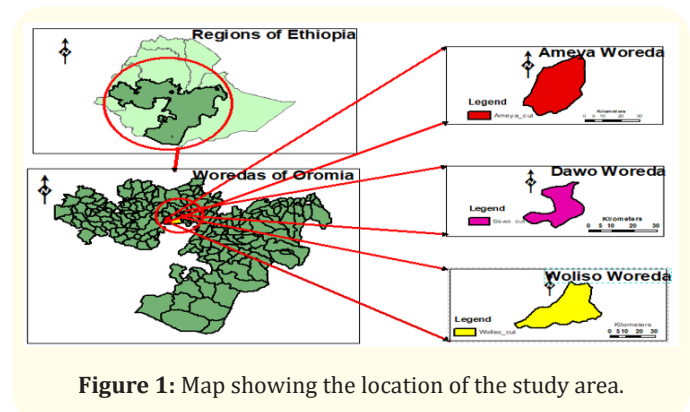


Figure 1: Map showing the location of the study area.

Woliso district is one of the districts of Southwest Shewa zone of Oromia regional state which is located at about 114 km from Finfinne (Addis Ababa) in South-west direction. The area is located at a latitude of 8° 32' 3.01" N and longitude of 37° 57' 54.54" E along Finfinne to Jimma main road with an elevation ranging from 1600 - 2880m above sea level. Agro-ecologically, it is classified into Midland (70%) and Highland (30%) zones. It has bimodal rain seasons. Long rainy season occurs from June to September and short rainy season extended from March to April. The minimum and maximum annual rain fall and daily temperature ranges from 1250 to 1450 mm and 8 to 25 respectively. The farming system of the area is mixed crop livestock production system and livestock population of the area is estimated to be about 224,334 heads of cattle, 2,101mules, 16,320 donkeys, 39,543 sheep, 51,042 goats and 127,679 poultry [19].

Dawo is one of the districts of Southwest Shewa zone of Oromia regional state which is located at about 96 km from Addis Ababa (Finfinne) in South-west direction (80 kilometers paved with asphalt and 16 kilometers gravel). The area is located at a latitude of 8° 44' 59.99" N and longitude of 38° 09' 60.00" E with an elevation ranging from 1650-2750m above sea level. Agro-ecologically, it is classified into Midland (69%) and Highland (31%) zones. Long rainy season occurs from June to September and short rainy season extended from March to April. The annual rainfall varies from 900 to 1100 mm; while the annual mean temperature vary from 17 to 20 with mean value of 18. The total land area of the woreda is 64,116.25 ha, of which 48,337 ha (75%) are considered suitable for agriculture. Grazing and forest lands accounts for 6.73% and 7% respectively. The livestock population of the area is estimated to be about 80,965 heads of cattle, 207 mules, 10,932 donkeys, 39,014 sheep, 10,942 goats and 65,944 poultry [20].

Ameya is one of the districts of Southwest Shewa zone of Oromia regional state which is located at about 144 km from Addis Ababa (Finfinne) in South-west direction and 30 kilometers far away from the administrative center of Southwest Showa zone -Woliso town. The area is located at a latitude and longitude of 8° 29' 59.99" N and 37° 44' 59.99" E with an elevation ranging from 1600 - 3200m above sea level. Agro-ecologically it is classified into Midland (64%) and Highland (36%) zones. Long rainy season occurs from June to September and short rainy season extended from March to April. The minimum and maximum annual rain fall ranges from 900 to 1800 mm and the mean annual temperature ranges from 12 to 32. The livestock population of the area is estimated to be about 160,500 heads of cattle, 3,300mules, 15,200 donkeys, 32,600 sheep, 35,400 goats and 101,000 poultry [21].

Study herds and animals

The target population comprises all cattle above six months of ages with no records of CBPP vaccination in Woliso, Dawo and Ameya districts of Southwest Shewa zone of Oromia Regional State. Herds of cattle managed under extensive production system were included in this study.

Study design

A cross sectional study was conducted in Woliso, Dawo and Ameya districts of Southwest Shewa zone of Oromia Regional State and proportional sample size were considered from these districts according to their cattle population. Body condition scores of animal was scored and body condition scoring 1 and 2 were recorded as poor body condition and body condition scoring 3, 4 and 5 were recorded as Good body condition.

Sample size determination and sampling technique.

Sample size determination

Since there was no previous report 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 [22] formula and a total of 384 animals were selected.

Where n = required sample size, P_{exp} = expected prevalence, and d = desired absolute precision (0.05).

An estimated sample size for households from which animals were selected for blood sample collection was calculated by dividing the total sample size ($n = 384$) to the number of animals sampled from each herd (six) given an estimated of 64 households for inclusion. However, due to the inclusions of households that having less than six cattle, the total sample size of herds was inflated to 96. Therefore, in this study a total of 96 herds were selected.

Sampling methods

Southwest Shewa zone was purposively selected because the status of CBPP has not been known yet in the area. Three districts were selected conveniently from the zone based on cattle population and accessibility. By assuming the population is homogeneous, equal numbers of peasant associations were considered from these three districts and from each selected district, three peasant associations were selected conveniently based on cattle population, accessibility and availability of infrastructure.

Household that have at least two cattle in each selected PA and defined as herd was considered as primary sampling unit. Lists of households were identified from Peasant association agricultural office. Then households were selected using systematic random sampling and the selected households were informed by animal health workers to provide their cattle for sampling purpose. From Woliso, Dawo and Ameya districts 150, 102 and 132 animals were selected respectively according to their cattle population. Equal numbers of animals were considered from three selected peasant association within each district. Animals from each household were tethered separately by object. The number was assigned to animal by counting tethered animals either from right to left or from left to right. The maximum sample size of six cattle (i.e. the average number of cattle per household of the area) was sampled from each selected cattle herd (household that having animal) for serum sample collection. Only six cattle were sampled randomly from households those having greater than six cattle. However, in case of households those having ≤ 6 cattle, all animals were sampled.

Data collection

Blood sample

About 10 milliliters of blood samples were collected from the jugular vein of each cattle using sterile vacutainer tubes and needles by following aseptic procedure after animal was restrained by owner, and sample was properly labeled. The samples were kept protected from sun light in a slanting position for 6-8 hours. Then the serum samples were separated and transferred to a sterile tube (cyrovial) and stored at -20°C at Woliso district veterinary clinic until required for analysis using Competitive ELISA at Bedelle Veterinary Regional Laboratory.

Animal and herd information

Animal and herd level data/information were collected from selected animals and herds to see the risk factors associated with occurrence of CBPP. Accordingly data on location (both districts and peasant associations), age, sex, body condition score, origin of the animal, history of respiratory related clinical signs, herd size, contact with other herds and introduction of new animal into herds were collected [23,24].

Diagnostic methods

Serological test

Sera were tested using the CBPP c-ELISA [25] following the manufacturer's instructions. The tests were interpreted as negative for results below 50% and positive above 50%.

Data storage and analysis

Data generated from laboratory investigations with herds and animals level information were recorded and coded using Microsoft Excel spreadsheet (Microsoft Corporation). It was transferred to Stata version 13 and analysis was performed. The seroprevalence of CBPP was calculated as the number of sero-positive samples divided by the total number of samples tested. The statistical significance difference of seroprevalence of CBPP across study districts and among peasant associations was tested by chi-square (χ^2) test. At the beginning, association of sero-positivity with risk factors age category into Young ≤ 5 , Adult > 5 , Sex (female, male), body condition categorized into poor (1, 2), Good (3, 4, 5), origin of animal (Born, Bought), herd size categorized into Large ≥ 5 , and small < 5 , history of herd mixing with other herd (yes, no) and introduction of new animal into herd (yes, no) were screened using univariable logistic regression analysis. Variables that found with $p \leq 0.25$ in univariable were subjected to multivariable logistic regression analysis with generalized linear model logit link. The presence multicollinearity was checked using tolerance and variance inflation factor. Spearman's rho was used to see the correla-

tion between the CBPP occurrence and history of previous respiratory related clinical signs. In all the analysis significance level 0.05 ($p < 0.05$) was considered to be statistically significant.

Results

Overall seroprevalence of CBPP

Out of 384 selected animals, 36 were sero-positive for *Mycoplasma mycoides subspecies mycoides* Small Colony (MmmSC) specific antibody. The overall animal level seroprevalence of CBPP in the study area was 9.4% (95%CI: 6.5-12.3). From 96 cattle herds, 31 were seropositive for CBPP specific antibody. The overall herd level sero-prevalence of CBPP was 32.3% (95% CI: 22.9-41.6). In this study different seroprevalence was recorded across the study locations in which the highest sero-prevalence (12%, 95%CI: 6.8 - 17.2) was observed in Woliso districts while the lowest sero-prevalence (7.6%, 95%CI: 3.1 - 12.1) was recorded in Ameya district. Similarly, the highest sero-prevalence (20%, 95%CI: 8.9 - 31.1) was observed in Gurura peasant association while the lowest seroprevalence (2%, 95%CI: 0.1- 5.9) was recorded in Obbi peasant association. However, chi-square (χ^2) test showed that there was no statistically significance difference in seroprevalence of CBPP across study districts and among peasant associations (Table 1).

Risk factors analysis

Animal level risk factors

In this study various animal-level sero-prevalence were recorded across different potential risk factors like age, sex, body condition and origin of the animal. Initially, these risk factors were screened at ($p \leq 0.25$) using univariable logistic regression analysis and this analysis revealed that age, sex, body conditions and origin of animal were entered into multivariable logistic regression analysis (Table 2).

Multivariable logistic regression analysis of potential risk factors with seroprevalence of CBPP found that age and body condition had statistically significant ($p < 0.05$) association with seroprevalence of CBPP. Cattle in adult age group (> 5 years) (OR = 4.3, 95%CI: 1.8-10.1, $P < 0.05$) were found more likely to be sero-positive of CBPP than cattle with young age group (≤ 5 years). Cattle with poor body condition (OR = 3.4, 95%CI: 1.3-9, $P < 0.05$) were found more likely to be affected by CBPP than cattle with good body condition score (Table 3).

Variables	Categories	No of cattle examined	No tested positive	Prevalence % (95%CI)	X ² (p value)
Districts	Woliso	150	18	12 (6.8 - 17.2)	2.00 (0.368)
	Dawo	102	8	7.8 (2.6 - 13.1)	
	Ameya	132	10	7.6 (3.1 - 12.1)	
Pas	Badesa Koricha	50	7	14 (4.4 - 23.6)	14.75 (0.064)
	Gurura	50	10	20 (8.9 - 31.1)	
	Obbi	50	1	2 (0.1 - 5.9)	
	KarsaAdai	34	3	8.8 (0.7 - 18.0)	
	Karsa Bombi	34	1	2.9 (0.1 - 8.6)	
	Nano Gabriel	34	4	11.8 (0.9 - 22.6)	
	Udad2	44	2	4.5 (0.1 - 10.7)	
	Kota	44	5	11.4 (2.0 - 20.7)	
	GomboroAliye	44	3	6.8 (0.6 - 14.3)	
Total		384	36	9.4 (6.5-12.3)	

Table 1: Sero-prevalence of CBPP across study districts and peasant associations.

CI: Confidence Interval; Pas: Peasant Associations

Variables	Categories	No of cattle examined	No tested positive	Prevalence % (95%CI)	OR (95%CI)	p-value
Age	Young (≤5yrs)	178	7	3.9 (1.1-6.8)	*	*
	Adult (>5yrs)	206	29	14.1 (9.3-18.8)	4.0 (1.7-9.4)	0.001
Sex	Female	188	14	7.4 (3.7-11.2)	*	*
	Male	196	22	11.2 (6.8-15.6)	1.5 (0.8-3.2)	0.207
Body condition score	Good (≥3bcs)	119	5	4.2 (0.6-7.8)	*	*
	Poor (<3bcs)	265	31	11.7 (8.4-16.1)	3.0 (1.1-8.0)	0.026
Origin of animal	Born	275	18	6.5 (3.6-9.5)	*	*
	Bought	109	18	16.5 (9.5-23.5)	2.8 (1.4-5.7)	0.003

Table 2: Univariable logistic regression analysis of risk factors with animal level seroprevalence of CBPP (n = 384) Southwest Shewa zone, Ethiopia.

*=Reference group; CI: Confidence Interval; OR: Odd ratio

Risk factors	Categories	Prevalence %(95%CI)	OR (95% CI)	P-value
Age	Young (≤5yrs)	3.9 (1.1-6.8)	*	*
	Adult (>5yrs)	14.1 (9.3-18.8)	4.3 (1.8-10.1)	0.001
Body condition	Good (≥3score)	4.2 (0.6-7.8)	*	*
	Poor (<3score)	11.7 (8.4-16.1)	3.4 (1.3-9)	0.015

Table 3: Multivariable logistic regression analysis of risk factors with animal level sero-prevalence of CBPP (n = 384) Southwest Shewa zone, Ethiopia.

*= Reference group; CI: Confidence Interval; OR: Odd Ratio; yrs: Years

Herd level risk factors analysis

In this study the risk factors that considered at herd level seroprevalence were herd size, contact (herd mix), and introduction of new animal into the herd. Results of risk factors analysis with herd-level seroprevalence showed that among herd-level risk factors

that considered, only herd size had statistically significant effect on seropositivity of CBPP ($p < 0.05$). Larger herds (OR = 7.5, 95%CI: 2.7-21.2, $p = 0.000$) were more likely to be affected by CBPP than smaller herds (Table 4).

Variables	Categories	No of cattle herds examined	No tested positive	Prevalence % (95%CI)	OR (95%CI)	p-value
Herd size	Small size (<5cattle)	48	6	12.5 (3.1-21.9)	*	*
	Large size (≥ 5 cattle)	48	25	52.1 (37.9-66.2)	7.5 (2.7-21.2)	0.000

Table 4: Final herd level risk factors analysis to CBPP seroprevalence (n = 96) Southwest Shewa zone, Ethiopia.

*=Reference group; CI: Confidence Interval; OR: Odd Ratio

Spearman correlation analysis

Correlation analysis using Spearman's rho was conducted to test correlation between history of previous respiratory problems and seroprevalence of CBPP which revealed that there was statistically significant association between history of respiratory problems and seroprevalence of CBPP ($r_s = 0.2016$, $P = 0.001$).

Discussion

In this finding, an overall seroprevalence of 9.4% (95%CI: 6.5-12.3) was recorded. This result is closely in agreement with the results of various researchers as reported by Mamo., *et al.* [4] from Gimbo District, Southwest Ethiopia (8.1%), (8) from Bishoftu and Export Oriented Feedlots around Adama using c-ELISA (8.7%), Schnier., *et al.* [23] from Southwestern Kenya (9.7%), Kassaye and Molla [24] from Export quarantine center of Adama (9.5%), Molla and Delil [25] from Dassenech district of South Omo zone (10%) and Teklue., *et al.* [26] from Southern part of Tigray using CFT test (11.9%). However, the overall seroprevalence is by far much lower than the previous findings reported by Ebisa., *et al.* [16] from Amaro special districts, southern part of Ethiopia (31.8%) and Daniel., *et al.* [17] from three districts of Western Oromia (28.5%). On the other hand, the findings of this study was higher than the results reported by Alemayehu., *et al.* [15] from bulls originated from Borena pastoral area of Southern Ethiopia (0.4 %) and Dele., *et al.* [27] from Export quarantine center of Adama (0.4%).

The overall herd level seroprevalence of CBPP was 32.3% (95%CI: 22.9-41.6) which is closely similar with the finding of Gizaw [28] who reported seroprevalence of 30.4% in Somali Regional State of Ethiopia. However, this result was higher than the previous report of Bonnet., *et al.* [11] with 4.6% in the Ethiopian highlands. On the other hand the finding was lower than the finding reported by Suleiman., *et al.* [29] from agro-pastoral areas of Nigeria (54.7%)

and Mersha [10] from selected districts of East Wollega and West Shewa zone of Oromia region (54%).

This variation in seroprevalence reported from various researchers might be due to variation in temporal and spatial distribution of the disease, differences in agro ecological systems, cattle management and production systems, population density, number of examined animals, and the types of tests used to determine seroprevalence of the disease Ebisa., *et al.* [16].

From risk factors that were assessed with seroprevalence of CBPP; age ($P < 0.05$, body condition score ($P = 0.015$), and herd size ($P = 0.000$) were significantly associated with the seroprevalence of contagious bovine pleuro-pneumonia. The seroprevalence of the disease was higher in animals within adult age group than cattle within young age group (≤ 5 yrs). On the contrary, Masiga., *et al.* [30] reported that adult cattle were more resistant to CBPP infection than younger animals. In addition, Masiga., *et al.* [31] reported that young animals are more susceptible to acute forms of CBPP infection than adult cattle and thus acutely infected young animals may die of CBPP and may not be available for testing. However, in contrast to Masiga., *et al.* [30], this study revealed that the seroprevalence observed in adult cattle was higher than seroprevalence observed in young cattle which is in line with the finding reported by Kassaye and Molla [24]. This may be associated to the fact that chronic stages of the disease are usually seen in adult cattle as the age progresses [32]. This might be due to long time exposure and persistence of sequestrum for a long period of time in CBPP recovered animals.

Body condition score was significantly ($P < 0.05$) associated with the seroprevalence of CBPP in which higher seroprevalence was recorded in cattle with poor body condition (< 3 bcs) as com-

pared to cattle with good body condition (≥ 3). This finding is in agreement with the finding reported by Biruhtesfa, *et al.* [33] from Bishoftu abattoir. This statistically significant association may be due to the fact that animals with poor body conditions are more susceptible to the disease and they may have low immunity to resist the disease. Besides, animals with poor body conditions are more susceptible to the disease than animals with good body condition [10], this findings was also in agreement with an assumption that CBPP sero-positive animals had poor body conditions than sero-negative animals; thus, this result may also indirectly describes the impact of the disease associated with loss of productivity.

Herd size had statistically significant association ($P < 0.05$) with sero-prevalence of CBPP. The sero-prevalence of the disease was higher in large herd size compared to small herd size. The significant difference in the sero-prevalence of the disease between the herd sizes was in agreement with the findings of the study conducted in bulls originated from Borena pastoral area of Southern Ethiopia [15]. This significance difference may be due to the fact that probability of getting the disease increase as herd size increase due to crowding of animal and the disease spread within the large herd size easily because of the contagious nature of disease.

Correlation analysis using Spearman's rho revealed that there was statistically significant association between history of previous respiratory problems and sero-prevalence of CBPP. Among the clinical signs of CBPP, the respiratory signs such as dyspnea, polypnea, cough and nasal discharges in cattle are the common to be mentioned [9,34]. The reason for this statistically significant association between history of previous respiratory problems and sero-prevalence of CBPP might be due to the clinical characteristics of the disease. Hence, based on the present study result, if animals were exhibited any respiratory health problem in the study area; it could be an indication of CBPP infection.

Conclusion and Recommendations

The present study revealed that animal and herd level overall sero-prevalence of CBPP in the study area was 9.4 % and 32.3 % respectively. This result showed that the disease is prevalent in cattle in selected (Woliso, Dawo and Ameya) districts of Southwest Shewa zone of Oromia region, suggesting the disease could cause considerable economic losses through morbidity and mortality. This study showed that age, body condition and herd sizes were the identified risk factors for CBPP sero-positivity in cattle. In conclusion, removing or culling older animal, animal with poor body condition and technical support for larger herd owners may help in

reducing the prevalence of the disease in the study area until other control stage designed.

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Conflicts of Interest

There is no conflict of interest.

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