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Research article

Seroprevalence and associated risk factors of pox infection among sheep and goats in selected districts of Afar region, Ethiopia

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ABSTRACT

Background: Sheep and goat pox virus infection is highly devastating viral disease of small ruminants that cause severe production losses in sheep and goats in Ethiopia and also limits international trade. A cross-sectional study was employed with the objective to estimate the seroprevalence of pox infection and to assess associated risk factors during the study period, February to April, 2020. A total of 384 serum samples were collected from apparently healthy sheep and goats. Serum neutralization test was used to detect the presence of antibodies against pox virus at national veterinary institute. Descriptive statistics, univariable and Multivariable logistic analyses were used in this study.

Results: The overall animal level seroprevalence of sheep and goat pox virus was found to be 15.36% (n = 59/384) and species level prevalence rate was found to be 14.5% (n = 16/110) in sheep and 15.69% (n = 43/274) in goats in the study areas. Among the associated factors considered in this study, sex (P = 0.010), age (P = 0.012) and herd size (P = 0.029) were found to be statistically associated with seropositivity of pox infection in multivariable logistic regression. The odds of seropositivity in female animals were 3.9 times more likely to develop pox infection than male animals (AOR = 2.2; 95% CI = 1.203–4.015%) and the odds of young animals were 2.14 times more likely to be seropositive to pox infection than old aged animals (AOR = 2.14; 95% CI = 1.169–3.633%). Moreover, the odds of large-sized flocks of animals were 3.10 times more likely to be seropositive to pox disease than small-sized flocks (AOR = 3.10; 95% CI = 1.30–4.42%).

Conclusion: This study finding revealed that sheep and goat pox virus is prevalent and widespread diseases of small ruminant in afar region. Therefore, further study should be carried out to estimate region wise magnitude of the disease and control measures should be put in place to minimize the economic losses associated with this disease.

1. Introduction

Ethiopia is the first in Africa in terms of livestock population, with an estimated 65.35 million cattle, 39.89 million sheep, 50.50 million goats, and 48.96 million chickens [1]. In Ethiopia, the livestock sector is a significant contributor to the majority of the population's livelihood as a source of meat, milk, drought power, and income, particularly in pastoral and agro-pastoral areas [2]. Small ruminants are important livestock assets because they grow faster, require less investment, have higher fertility (shorter production cycles), and are more adaptable to adverse settings than giant ruminants [3, 4]. Despite having huge small ruminant resources, the country is unable to exploit the sector due to incredibly widespread livestock diseases, a lack of an appropriate disease control strategy, and a lack of government attention [2, 5].

Infectious diseases are one of the most serious challenges affecting small ruminant production and productivity, particularly sheep pox (SP) and goat pox (GP) and peste des petits ruminants (PPR) are major problems of small ruminants and widely distributed in all regions of the country and placed at the top of the list and widespread across the country [6]. It is a devastating systemic viral disease that affects domestic animals. Widespread skin eruptions, fever, generalized papules or nodules, vesicles (rarely) on non-wool skin, internal lesions in the lungs, pulmonary, and gastrointestinal mucosa, and mortality are the most common symptoms of the disease [7]. Because the diseases have a significant role in the agricultural economy, these are classified as OIE notifiable diseases [8, 9]. Ethiopia is the first of the top three African countries to have experienced the most small ruminant pox outbreaks in 2011 [10]. The viruses causing these diseases are members of the genus

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Capripoxvirus, subfamily *Chordopoxvirinae* and family *Poxviridae*. These *Capripoxvirus* isolates have a close genetic relationship. Clinically and serologically, including virus neutralization tests, the diseases caused by strains of SP virus, GP virus, and lumpy skin disease (LSD) virus cannot be distinguished. However, most strains of SP and GP virus have unique host preferences, with homologous hosts suffering from more severe disease [11, 12].

Sheep and goats pox (SGP) viruses cause significant production losses in endemic areas due to decreased milk yield, decreased weight gain, increased abortion rates, damage to wool and hides, and increased susceptibility to pneumonia and fly strike, as well as being a direct cause of mortality [13]. Morbidity is 75–100% in these vulnerable herds, with mortality rate ranging from 10 to 85 percent depending on the virus's virulence nature [14]. Despite its significant economic importance and trade threats, data on pox infection sero-prevalence and associated risk factors in the study districts in particular and the afar region in general is lacking. Appropriate disease control strategies would result from a better understanding of its seroprevalence and associated factors. As a result, the current study aimed to estimate the seroprevalence of sheep and goat pox viruses in small ruminants from the Asayita and Mille districts of Afar region as well as assess the associated factors.

2. Materials and methods

2.1. Description of study area

The study was conducted from February to April, 2020 in two districts namely (Asayita and Mille), which are located in the administrative zone one of afar region, Ethiopia. The afar pastoral region is located in northeast of Ethiopia between 39°34′ to 42°28′E longitude and 8°49′ to 14° 30′ N latitude (Figure 1). The region shares common international boundaries with Eritrea in the northeast and Djibouti in the east and it is characterized by an arid and semi-arid climate with low and erratic rainfall. Rainfall is bi-modal throughout the region, with a mean annual rainfall below 500 mm in the semi-arid western escarpments and decreasing to 150 mm in the arid zones to the east. The altitude of the Region ranges from 120 m below sea level in Danakil depression to 1500 m above sea level. Temperatures vary from 20 °C in higher elevations to 48 °C in lower elevations. Majority of human population of Afar region are pastoralists who largely depend on livestock production for their livelihood. The study populations were managed under pastoral husbandry which allows high mobility of animals and these animals are usually mixed with other animal species. In the study areas, sheep and goats were kept under the extensive farming system [1].

2.2. Study population

Sheep and goats older than six months were used to estimate the seroprevalence of pox virus infection in the study areas. This is due to the possibility that the young animals' passive immunity from the dam may have some influence on the outcome. Even while some studies indicated that maternal antibodies transferred on to the offspring provide protection predominantly for three months, there is a chance that maternal antibodies will still be present by the time the test is performed at six months, which could result in a false interpretation. Blood samples were drawn from non-vaccinated sheep and goats, and the study population were categorized into three age groups: young (6 months–1.5 year), adult (1.5–2.5 years), and old (>2.5 years) [15].

2.3. Study design and sampling strategy

A cross-sectional study design was used to estimate pox seroprevalence and assess associated risk factors from February to April 2020, in study areas across the Afar region. In Ethiopia, there is no serological test to differentiate between animals treated with the pox vaccine and animals who have recovered from a natural pox infection. As a result, a questionnaire was identified as the best source of information on sheep and goat vaccination status to support in sampling. Since the study districts were purposefully selected based on higher study population, access to transportation, history of no vaccination for the previous 6 months, absence of outbreak cases, and willingness of pastoralists to participate, the sampling method was three stage random sampling to collect the samples.

2.4. Sample size determination

Although sheep and goats are two species, they can be considered as one study population due to the management practices and the similar course of the diseases in both species. So, the sample size was determined according to the formula given by Thrusfield [16], using 50% expected prevalence (since there is no previous seroprevalence report of PPR infection in the study areas), 5% desired absolute precision and 95% confidence interval as below:

$$n = \frac{Z^2 x\ Pexp(1 - Pexp)}{d^2}$$

Where: n = required sampling units.

Z = Multiplier from normal distribution at 95% Confidence interval (1.96).

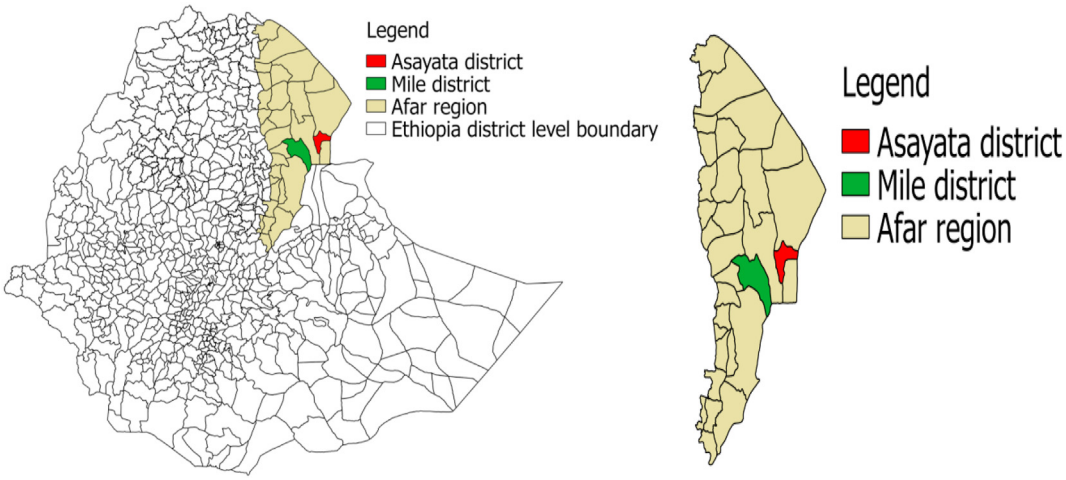

Figure 1. Map of the study districts.

Pexp = Estimated (expected) prevalence 50% (0.5).
(1-P) = Probability of having no disease 50% (0.5).
D = Desired absolute precision 5% (0.05).

Sampling was proportionally distributed based on the total small ruminants' population in the study districts' kebeles. The number of sheep and goats sampled is proportional to the herd sizes in each study district. Accordingly, a total of 384 study population (n = 110 sheep and n = 274 goats) from twenty four herds and eight kebeles or peasant associations and two districts were included in this study.

2.5. Sample collection and transportation

Whole blood samples approximately 6–8ml was collected from the jugular vein of non-vaccinated sheep and goats using plain 10 ml vacutainer tubes and 19 gauge sterile needles. The samples were labelled to allow identification of each animal. The potential risk factors (such as species, age, sex, herd size and study areas) were recorded during sampling. Collected samples were kept in slant position overnight at room temperature to allow serum separation. Then, serum was decanted and aliquoted into cryovials and stored in a freezer (−20 °C) at microbiology laboratory of Samara University, and transported to National Veterinary Institute (NVI) in order to test for antibodies against natural pox infection exposure using serological analysis. All sera samples were transported to NVI laboratory in icebox and stored at −20 °C until processed.

3. Serological analysis

3.1. Antibody detection against sheep and goat pox infection

The serum samples were tested for the presence of antibodies against sheep and goat infection using virus neutralization test (VNT) in NVI virology laboratory following the procedures described by Boshra et al. [17]. Sheep and goat pox antibodies cannot be distinguished by Viral Neutralization Test (VNT), thus, it is called 'sheep and goat pox antibody' when referring to the serological result. In brief, VNT cannot differentiate between antibodies made against the sheep pox virus and the goat pox virus. It is therefore only referred to as "sheep and goat pox antibodies."

3.2. Administration of questionnaire survey

A structured questionnaire format was prepared to interview individual sheep and goat owners. Respondents from each district were randomly selected and interviewed to assess associated factors of sheep and goat pox disease such as; species, sex, age, herd size and study areas. All necessary epidemiological information was collected on individual animal bases.

3.3. Data management and statistical analysis

All collected data generated from field and laboratory analysis was entered in to the Microsoft excel sheet data management and analysis Window® 2007 and then it was analyzed using Stata version 14 software. Descriptive statistics was employed to quantify the results of seroprevalence of antibodies against sheep and goat natural pox virus infection. The seroprevalence of pox virus infection was calculated as the number of pox positive animals divide to the total population at risk of acquiring pox disease[18]The association of associated factors such as different location, species, sex, herd size and age to the results of seroprevalence of pox infection was analyzed using Univariable and multivariable logistic regression model. A statistically significant association between variables was said to exist if the calculated P-value is less than 0.05 at 95% confidence interval (CI).

4. Results

In the current study, out of 384 sera collected from the study population and tested using serum neutralization test (SNT), The overall seroprevalence of pox virus infection was 15.36% (95% CI; 12.07–19.34) of which 14.5% in sheep and 15.69% in goats. The demographic characteristics of study population were presented in (Table 1). Majority of study population, 81.51% (n = 313) were females while about 18.49 % (n = 71) of them were males.

4.1. Seroprevalence and associated factors of pox infection in sheep and goats

Out of 384 sheep and goats sampled and sera tested, a total of 59 sheep and goats were found to be positive against pox antibodies as depicted in (Figure 2) and the overall seroprevalence was found to be 15.36% with 95% CI (12.07–19.34). In this study, out of 110 sheep sampled, a total number of 16 samples of sheep were found to be positive and the seroprevalence of sheep pox virus infection was 14.5% in the study districts of Afar region. Similarly, among 274 goats sampled, a total of 43 samples were found to be positive and the seroprevalence of goats' pox virus infection was 15.69% in the study areas. However, the antibody prevalence variation was not statistically significant between sheep and goats (P > 0.05). Out of the total sera samples tested, 35 samples were from females and 24 serum samples were from males with a prevalence of 19.44% (n = 35/180) and 11.76% (n = 24/204), respectively.

Animal and herd level associated factors of pox disease such as; species, sex, age, study area, herd size and body conditions were also analyzed using univariable and multivariable logistic regression. The result revealed that the associated risk factors that had a significant association with c-ELISA sero-positivity were sex (P = 0.047), age group (P = 0.033) and herd size (P = 0.039). Whereas, Species (P = 0.47), study areas (P = 0.91) and body condition score (P = 0.76) had no significant association with seropositivity of the disease as depicted (Table 2). In the current study, the seroprevalence of pox virus infection between sex groups showed that it was 13.3% in males and 17.16% in females and this seroprevalence result was found to be statistically significant variation between sex groups (P = 0.047; COR = 2.3; CI; 1.306–4.352). When the current study result of pox virus infection were compared among different age groups of the study populations, an increasing seroprevalence trend was observed with increasing age, and the difference was statistically significant (P = 0.033; OR = 2.06; CI; 1.306–4.352). Moreover, the study population's seroprevalence of antibodies against the pox virus increased as herd size increased, as shown in Table 2, and this difference was statistically significant (P = 0.039; OR = 1.81; CI; 1.02–3.20), revealing a direct positive association between herd size and pox virus seroprevalence.

Table 1. The demographic characteristics of the tested animals.

Variable	Category	Frequency (%)
Sex	Female	313 (81.51)
	Male	71 (18.49)
Species	Sheep	110 (28.60)
	Goats	274 (71.40)
Age	Young (*6months* −1*year*)	55 (65.4)
	Adult (1 year < x ≤ 2.5 years)	141 (47.8)
	Old (2.5 < x ≤ 4 years)	193 (67.9)
Herd size	Small (< 50 *animals*)	86 (53.4)
	Medium (51–100animals)	116 (51.7)
	Large (>100 *animals*)	182 (69.2)
District	Asayita	179 (46.61)
	Mille	206 (53.64)
Total		384 (100%)

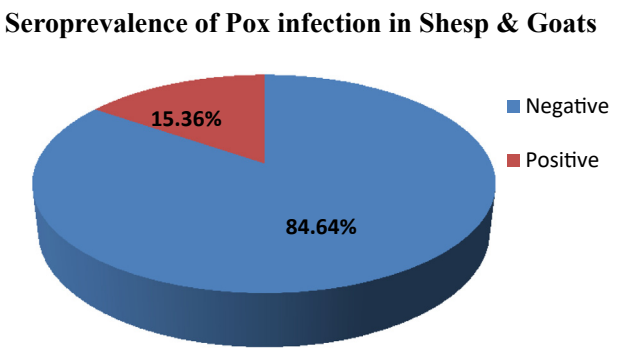

Figure 2. Seroprevalence of sheep and goat pox infection in the study districts of afar region.

Accordingly, among associated risk factors considered in this study; sex, age and herd size were found to be statistically significant variables in univariable logistic regression and were fitted to the final multivariable logistic regression model to check the real significant contribution of these associated risk factors without compounding effect on the others as depicted in (Table 3) with adjusted odds ratio (AOR) and hence, sex, age and herd size of animals were identified as associated factors for the occurrence of sheep and goat pox infection. The odds of seropositivity in female animals were 3.9 times higher than male animals (AOR = 2.2; 95% CI = 1.203–4.015%). The odds of young animals were 2.14 times more likely to be seropositive to pox virus infection than old aged animals (AOR = 2.14; 95% CI = 1.169–3.633%). In addition, the odds of large-sized flocks of sheep and goats were more likely to be seropositive to pox disease than small-sized flocks (AOR = 3.10; 95% CI = 1.30–4.42%), which means the study population that were found in large herd size were 3.10 times more likely to develop pox virus infection as compared to animals found in small herd size (<50 animals) as shown in (Table 3).

5. Discussion

Sheep and goat pox viruses' infection are highly devastating viral diseases of small ruminants that cause severe production losses in sheep and goats in the diseases' respective endemic areas in Ethiopia and also limits international trade and cause other economic losses. Because of the pox infection's rapid transboundary nature and substantial financial impact on the livestock industry, the World Organization for Animal Health (OIE) has categorized it as a notifiable disease [8, 9]. The present study revealed that the overall seroprevalence of 15.36% of which 14.5% of sheep and 15.69% of goats were exposed to pox viruses' infection, indicated that the disease is prevailing in small ruminants and circulating in the study areas. According to the seroprevalence report of the current study result, sheep and goats in the study area were both equally exposed to sheep and goat pox infection in the study districts of Afar region. The current finding was in agreement with previous studies of Fentie et al. [19], who reports 15.5% in western part of the Amhara region. However, the present study was lower than the reports of Elshafie and Ali [20] who reported 63.55% in Sudan and Masoud et al. [21] who reported 17.24% in Pakistan. According to a number of study findings, differences in animal movement and the introduction of new animals to the herd may be to blame for the differences in seroprevalence by region.

Among the associated risk factors considered in the current study; sex (P = 0.010), age (P = 0.012) and herd size (P = 0.029) were found to be statistically associated with the diseases in multivariate logistic regression analysis model. In this study the seroprevalence of pox infection among sex groups was 17.16% in females and 13.3% in males. This relative seroprevalence variation was statistically significant, which means female animals are more likely to develop pox virus infection as compared to male animals. The odds of pox infection seropositivity in female animals were 2.2 times higher than male animals (OR = 2.2; 95% CI = 1.203–4.015%). The results of the current study is in agreement with those from previous studies in Ethiopia (Amhara region), Pakistan, Sudan and Iran published by Fentie et al. [19], Masoud et al. [21], Elshafie and Ali [20] and Sadri [22], respectively. When the seroprevalence of sheep and goat pox was compared in different age groups of sheep and goats, it was found that young animals were more likely to be seropositive than older ones (P = 0.012), and this difference was significantly associated (OR = 2.06; 95% CI: 1.169–3.633). The lower immunity in young and female sheep and goats induced by lambing/-kidding phase or deprived physiological state may also contribute to the higher seroprevalence in these groups and maternal immunity provides protection only for up to 3 months. However, the current study result disagrees with previous study Sari [22], who reported higher seroprevalence in older animals than young ones.

In the present study finding, seroprevalence of sheep and goats pox infection was also significantly affected by herd size. In this study, multivariable logistic regression analysis showed that herd size and seropositivity of pox virus infection were statistically significant (P = 0.029), which means large herd sized of sheep and goats were 3.10 times more likely to develop pox infection as compared to those animals from small herd size keeping the other factors constant (OR = 3.10; 95% CI = 1.30–4.42%). This direct association might be an indication of the contagious nature of the disease and mode of transmission, which is

Table 2. Univariable analysis of associated factors with pox infection in the study districts.

Associated factors		Positive samples	COR	95% CI	P-value
Species	Ovine	16	Ref	-	-
	Caprine	43	1.27	0.66–2.43	0.47
Sex	Males	24	Ref	-	-
	Females	35	2.3	1.306–5.351	0.047
District	Asayita	33	Ref	-	-
	Mille	26	0.91	0.45–1.85	0.811
Age	Young (6m–1 year)	11	Ref	-	-
	Adult (1–2.5 years)	34	0.83	0.37–1.84	0.64
	Old (2.5 < X < 4 years)	14	2.06	1.26–5.94	0.033
BCS	Poor BCS	20	Ref	-	-
	Good BCS	39	0.76	0.41–1.41	0.39
Herd size	Small herd size	8	Ref	-	-
	Medium herd size	16	1.87	0.65–5.35	0.24
	Large Herd Size	35	1.81	1.02–3.20	0.039

NB: COR = Crude odds ration; BCS = Body condition score; CI = Confidence interval.

Table 3. Multivariable logistic regression analysis of associated risk factors of pox seropositivity in sheep and goats.

Associated risk factors		Positive samples	Prevalence	AOR	95% CI	P-value
Sex	Male	24	11.8%	Ref	-	-
	Female	35	19.4%	2.2	1.203–4.015	0.010
Age	Old (2.5 < X < 4 years)	14	10.20%	Ref	-	-
	Young (6m–1 year)	11	20%	2.14	1.169–3.633	0.012
Herd size	Small herd size	8	9.3%	Ref	-	-
	Medium herd size	16	13.8%	1.87	0.58–2.42	0.83
	Large herd size	35	19.2%	3.10	1.30–4.42	0.029

NB: AOR = Adjusted odds ration; BCS = Body condition score; CI = Confidence interval.

attributed to crowding of animals that could facilitate the frequency of direct contact and hence accelerating the likelihoods of transmission. The only factors that were not shown to have a significant association with seropositivity to pox virus infection in the current study are study areas and body condition score. According to the current study's multivariate regression analysis, there was no statistically significant variation in seropositivity among the study districts. This could be probably because of pox virus infection is persistent and evenly distributed in the study areas. These results may indicate that pox virus infection is endemic and extensively circulating within these study districts. Moreover, the study areas may have similar agro-ecological conditions.

6. Conclusions

The current study finding indicated that the overall pox virus infection seroprevalence among the study populations was 15.36%. Hence, this study confirmed the pox virus is widely prevalent in small ruminants and circulating in Afar region, Ethiopia. Among the risk factors assessed in this study; only sex, age and herd size were found to be statistically associated with the seropositivity of pox virus infection in sheep and goats. Further studies that cover entire region and nationwide to determine the seroprevalence and titer level of antibodies against pox virus are highly recommended which help to formulate appropriate control measures of pox disease in sheep and goats. Annual mass vaccination is recommended as the best feasible, cost-effective and viable method for the control of sheep and goat pox diseases, where animal movement is unrestricted.

Declarations

Author contribution statement

Teshager Dubie and Betelhem Dagnew: Conceived and designed the experiments; Analyzed and interpreted the data; Wrote the paper.

Fanuel Bizuayehu and Muhammed Hamid: Performed the experiments; Wrote the paper.

Gizachew Fentahun: Contributed reagents, materials, analysis tools or data.

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Data availability statement

Data will be made available on request.

Declaration of interest's statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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