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Seroepidemiology of Peste des Petits Ruminants in Sheep in Hama Governorate, Syria

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ABSTRACT

Peste des Petits Ruminants (PPR) is a rapidly spreading and severe disease. A high mortality rate also characterizes the disease, which is caused by a virus of the genus Morbillivirus, the family Paramyxoviridae. This study aimed to detect peste des petits ruminants (PPRV) antibodies in sheep herds located in Hama governorate, Syria. The study was conducted on 400 blood samples collected from sheep with clinical signs of PPR in herds. The serological investigation was carried out using ELISA. Based on cELISA, 28% (112/400) samples were PPR-positive (95% CI: 23.59-32.41). The seropositivity of PPR was significantly (P<0.001) higher in the Alhamra district (60%; 42/70) as compared to other districts. Similarly, seropositivity was higher in less than 6 months age (50.59%; 43/85), followed by market sheep, farming as rearing system and install feeding (47.5%, 19/40), desert (32%; 103/350) than in other ecological zones, and in female sheep (31.53%, 99/314). It was concluded that PPR is an imminent threat to sheep herds and index to an upcoming epizootic in Syria.

Keywords: Sheep, Peste des Petits Ruminants, Seroepidemiology, Hama Governorate, Syria.

INTRODUCTION

Peste des petits ruminants (PPR) infects domesticated small ruminants, mainly sheep and goats, which include more than 90% of the total national livestock, excluding poultry in Syria (FAOSTAT 2023). In opposition-controlled areas in northwestern Syria, a survey was conducted under the supervision of a unit affiliated with FAO in 2019, which showed that Peste des Petits Ruminants (PPR) is widespread in those areas, with prevalence reaching 35% in the absence of vaccination against the disease (Benfield et al. 2023).

The first report on PPR was published in 1942 in Côte d'Ivoire (Gargadennec and Lalanne 1942). It was founded on observations that PPR was not transmitted to livestock that came into contact with it. Three decades later, the causal agent was identified as a different organism (Gibbs et al. 1979).

Peste des petits ruminants is primarily a disease of sheep and goats. Still, it has also been reported in camels (Govindarajan et al. 1997; Fakri et al. 2019), buffaloes (Govindarajan et al. 1997), cattle (Lembo et al. 2013), gazelles and dears (Elzein et al. 2004), etc. PPR, also known as goat plague and ovine Rinderpest, affects sheep, goats, and related species of small ruminants (Chowdhury et al. 2014) and is a highly contagious viral disease (Balamurugan et al. 2014; Khan et al. 2018). Pigs (Nawathe and Taylor 1979) suffer from subclinical infection but cannot shed the virus; thus, they are not considered to be of interest in virus epidemiology.

A virus of the genus *Morbillivirus* and the family *Paramyxoviridae* causes this disease (Gibbs et al. 1979; Barrett et al. 2006). This virus is faithfully related to the Rinderpest virus that infects cows and buffalo and is also associated with the measles virus, which infects humans; the distemper virus, which infects canines; and the measles virus that infects marine mammals (Barrett et al. 2006). It is a virus also known as "goat plague," "kata," "contagious pustular stomatitis," "pneumonia and enteritis syndrome," or "sheep plague" (Diallo 2000). PPR virus is easily transmitted by direct contact or through secretions of infected animals and is a highly contagious virus (Ezeibe et al. 2008; Khaliq et al. 2020).

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PPR is a rapidly spreading and severe disease characterized by a sudden onset of loss of appetite, eye, and nasal discharge, mouth ulcers, difficulty breathing, coughing, fever, and foul-smelling diarrhea (Khan et al. 2018; OIE 2020). The disease also has a high mortality rate (Dou et al. 2020; Abd-Elfatah et al. 2022; Alemu 2024). Morbidity and mortality could be 100% and 50–80%, respectively (Lefevre and Diallo 1990; Taylor et al. 1990; Parida et al. 2015; Abubakar et al. 2015; Khan et al. 2018; Zafar et al. 2024).

It was supposed to be a mutant of Rinderpest adapted for growth in sheep and goats because of the similarity in clinical signs and laboratory tests (Diallo 2000), and epidemiological and molecular research have established the difference between the causative agents of these diseases (Barrett et al. 2006; Esonu et al. 2022). This disease is a transboundary disease that is widespread in some developing countries. It causes significant economic losses (Khan et al. 2018). Therefore, it has been categorized as a disease that must be reported to the World Organization for Animal Health (Diallo 2000). The disease carries a severe threat to the lives of sheep and goat breeders in the endemic areas, and combating and eradicating it is important to alleviate insufficiency in those areas (FAO 2020).

Infected animals may transmit the agent to susceptible animals that come into contact with them through exhalation or clinical secretions (tears, nasal, saliva, feces) (Rossiter and Taylor 1994). Infected animals that get well from the disease acquire lifelong defensive immunity, and no carrier cases have been recognized (Hamdy et al. 1976). Also, the virus can occur in previously infected animals as mildly symptomatic, causing outbreaks in which previously uninfected disposed groups mingle with previously infected animals and show a slight form of the disease (Banyard et al. 2014). However, the virus's strain virulence and the infected animal's immune status cause clinical symptoms and death rates, which vary widely (WOAH 2022). For that, we decided to detect seroprevalence (antibodies) of PPRV in sheep herds located in Hama governorate, Syria.

MATERIALS AND METHODS

Ethical Approval

Those responsible for the work got blood samples from sheep while following ethical principles and considering animal rights. The animal owners agreed to collect samples from their animals.

Study Area and Epidemiological Data Collection

This study was conducted between February 2020 and August 2020. Fifteen (15) geographical areas were selected in Hama Governorate, Syria (Fig. 1), within two environmental regions in the current study (cross-sectional study). The study areas were chosen based on the location, environment, and locations of sheep herds in Hama Governorate, Syria.

Epidemiological data from sheep were collected using particular questionnaires based on previous studies, which involved information about animals such as farm name, geographical region, age, size of the herd, sex, breed, and disease status of the animals studied, as well as the climatic conditions of the area. Based on the information extracted, we categorized the data into various geographical regions (15 districts), ecological Zone (riverine/desert), sex (male/female), age (4 age groups: 0-6, 7-12, 13-24 and >24 months), source (market/born at the farm), rearing System (farming/non-farming), and Feeding System (stall feeding/grazing).

Fig. 1: Hama Governorate, a study area in Syria.

Sample Size

We collected 400 blood samples randomly from sheep herds located in 15 different geographical areas in Hama Governorate, Syria. Briefly, whole blood samples (5mL) without anticoagulant were collected aseptically from the jugular vein of each sheep and left to clot at room temperature. Then, these blood samples were placed in an ice pack until transported to the laboratory. Serum was collected in clean, sterile 1.5mL Eppendorf tubes and stored at -20°C until analysis.

Serological analysis

Serological analysis was performed at the Central Veterinary Laboratory at the Ministry of Agriculture and Agrarian Development, Damascus, Syria. The diagnostic kit (Biological Diagnostic Supplies Limited) was used to detect PPRV antibodies using a competitive enzyme-linked immunosorbent assay (ELISA). The cELISA and diagnostic kit were per protocol (Anderson et al. 1991). The optical density (OD) values of the finishing plates were at 450nm using an ELISA reader, and the results were measured according to the optical density, depending on the monoclonal antibody concentration. The results were taken to mean (depending on the manufacturer's instructions) by the PI (Percentage inhibition) value for every sample. PI value ≥50 was considered positive.

Statistical Analysis

Data thus collected from field and laboratory managed in a Microsoft Excel 2010 spreadsheet (Los Angeles, CA, USA) and analyzed using the statistical program SPSS version 22 (Statistical Package for Social Sciences (IBM) Inc., Chicago, IL, USA). Absolute and relative frequency were calculated for the study variables with a definite pattern. The seroprevalence of PPR was calculated based on cELISA test results using EACH categorical variable in the study. 95% confidence intervals (CI) were calculated for the recorded epidemic prevalence. Analytical statistics for each categorical variable included in the questionnaires, i.e., study areas (n=15), environmental zone (n=2), sex (n=2), source of sheep (n=2), age (n=4), rearing system (n=2), and feeding system (n=2) were carried out. The association between the PPR seroprevalence and the studied variables (risk factors) was investigated using chi-square, considering significance at $P<0.05$.

RESULTS

Distribution of the study sample

The relative frequency of variables was calculated. The most significant number of samples was from the Alhamra region, constituting (17.5%) of the total studied samples. Other samples comprised from the desert region (87.5%), female sheep (78.5%), sheep 7-12 months old (54.5%), sheep born in farm/home (90.0%), sheep raised in a nonagricultural system (90.0%), and sheep grazed (90.0%) of the total samples studied (Table 1).

Seroprevalence

The overall seroprevalence of PPR in sheep herds according to cELISA was 28% (112/400; 95% CI: 23.59-32.41%), with the highest seroprevalence in the Alhamra region (60.0%; 95% CI: 55.19-64.81%) followed by in sheep aged 0-6 months (50.59%; Fig. 2), market sheep, farming system and stall grazing system (47.50%), the desert region (32.0%) and female sheep (31.53%; 95% CI: 26.97-36.09%) (Table 1).

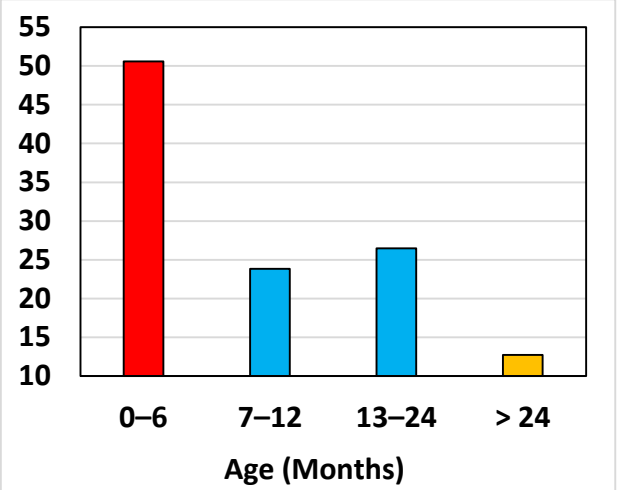

Fig. 2: Seropositivity of PPRV differs significantly ($P<0.001$) in various age groups of sheep.

Study of variables associated with PPR

The study showed a relationship between the seroprevalence of the disease in sheep herds, and several studied variables were considered risk factors for this disease occurrence (Table 1). Data related to study areas/districts, ecological regions, sex, age, source of sheep origin, rearing systems, and feeding system showed significant differences ($P<0.001$).

DISCUSSION

The present study is one of the first quantitative epidemiological studies recorded for the first time in Syria on the prevalence of PPR disease in sheep in different geographical areas in Hama Governorate, Syria, including the sheep herds in those areas. Fifteen different geographical areas were studied in Hama Governorate, Syria, where sheep are raised, and 400 blood samples were collected from sheep suffering from clinical signs and symptoms of PPR to examine the prevalence of PPR among sheep suffering from clinical signs and symptoms related to the disease. The study recorded that the prevalence of Peste des petits ruminants (PPR) was 28% of the total blood samples examined according to the scientific methodology.

The results of our study were consistent with a study on the seroprevalence and risk factors for PPR that was conducted in northern Jordan, which is located to the south of Syria. The seroprevalence for PPR in sheep herds was 29%; this percentage is similar to previous epidemiological studies conducted by researchers (Dhar et al. 2002; Ozkul et al. 2002) in separate areas of the western Asian continent. The results of this study were also consistent with the seroprevalence of PPR in sheep conducted by researchers (Banik et al. 2008) in Bangladesh, where the prevalence reached 27%.

Table 1: Seroprevalence depending on cELISA results and its association with categories of studied variables

Variable	Category	N	Positive		95% CI		P Value
			No.	%	Lower	Upper	
District	City Center	40	19	47.50	42.60	52.40	0.001
	Aqirbat	30	7	23.33	19.18	27.48	
	Aqarib	18	2	11.11	8.03	14.19	
	Alsein	26	4	15.38	11.84	18.92	
	Tal Altut	16	0	0.00	0.00	0.00	
	Alsabora	35	9	25.71	21.43	30.00	
	Casson	20	0	0.00	0.00	0.00	
	Alhamra	70	42	60.00	55.19	64.81	
	Taibat Alimam	20	0	0.00	0.00	0.00	
	Ma'radas	22	0	0.00	0.00	0.00	
	Shatha	21	8	38.10	33.33	42.86	
	Howiz	20	5	25.00	20.75	29.25	
	Algeed	9	0	0.00	0.00	0.00	
	Gedrin	20	4	20.00	16.08	23.92	
	Taksis	33	12	36.36	31.64	41.08	
Ecological Zone	Riverine	50	9	18.00	14.23	21.77	0.001
	Desert	350	103	32.00	27.42	36.58	
Sex	Male	76	13	17.11	13.41	20.80	0.001
	Female	314	99	31.53	26.97	36.09	
Age	0–6 Months	85	43	50.59	45.68	55.49	0.001
	7–12 Months	218	52	23.85	19.67	28.04	
	13–24 Months	34	9	26.47	22.14	30.80	
	> 24 Months	63	8	12.70	9.43	15.97	
Source	Animal Market	40	19	47.50	42.60	52.40	0.001
	Born In Farm	360	93	25.83	21.54	30.13	
Rearing System	Non Farming	360	93	25.83	21.54	30.13	0.001
	Farming	40	19	47.50	42.60	52.40	
Feeding System	Stall Grazing	40	19	47.50	42.60	52.40	0.001
	Grazing	360	93	25.83	21.54	30.13	

On the other hand, the seroprevalence of PPR in sheep was more than what was stated by researchers in Saudi Arabia, where the seroprevalence was 3.1% (AL-Afaleq et al. 2004) and higher than the seroprevalence reported in Yemen, which was 15% (Taylor 1997). Our results are higher than those reported in a study conducted in Pakistan, where the seroprevalence of PPR in sheep was 2.98% (Krishna et al. 2001). The reason for recording low seroprevalence in these countries could be due to the regular vaccination of sheep and goat herds. Vaccination provide protection from PPR infection (Zafar et al. 2024). Other reason could be that farmers are not allowing the entry of animals carrying the disease and illegally into those countries (Zhao 2021).

While the seroprevalence of PPR in sheep based on positive antibodies was lower in the present study was lower than those reported by Rahman et al. (2004) in Bangladesh (Ahaduzzaman 2020), separate areas of the Western Asian continent (Ali et al. 2023), and Central-Western Sudan (Balamurugan et al. 2020) in the northern region of India, which reported to 36, 38.63, 88, and 44.05% respectively, and lower than the percentage that was recorded by researchers (Rahman et al. 2023) which amounted to 44.3% in Afghanistan, and also lower than the percentage reported in (Hota et al. 2018) in India, in which the seroprevalence of PPR in sheep herds was 44.7%.

These varying seroprevalence of PPR in sheep might be attributed to many reasons, including the differences in the size of the study herds, breeding systems, methods of disease diagnosis, the presence or absence of preventive vaccination, as well as the spread of the disease in certain areas, and the presence of previous PPR infection usually creates immunity, the porous borders for the movement of animals and the entry of new animals into the herds lead an essential role in the spread of the disease (Taylor 1997; Rahman et al. 2021; WOAH 2022; Algezoli et al. 2024).

The present study recorded that the highest seroprevalence of the disease was in the Alhamra region in the Hama Governorate, Syria, compared to the other geographic regions in the governorate ($P<0.001$), which reached 60%. This high prevalence could be attributed to the Alhamra region containing more sheep than the other regions. Most of the samples were collected from this area because many sheep were suspected of being infected with the disease. The Alhamra area is an open area in the Syrian desert, which helps in the entry of illegal animals into it, in addition to the fact that veterinary services are insufficient in this area due to its distance from the city center.

This study showed that the seroprevalence of positive antibodies to PPR in herds of sheep belonging to areas with a desert environment is higher compared to riverine areas ($P<0.001$) and this is consistent with the findings of both researchers (Hota et al. 2018; Fentie et al. 2018) who linked difference reasons in seroprevalence between environmental regions with climatic features such as hotness, soil quality, rainfall, and climate humidity (Chauhan et al. 2012; Hota et al. 2018).

PPR infection was more common in female sheep compared to male sheep ($P<0.001$) in this study, which was consistent with the findings of Abdalla et al. (2012), Gari et al. (2017), Salih et al. (2014) and Ejigu et al. (2023) but was inconsistent with the findings of Mahajan et al. (2012) and Tajpara et al. (2022) who reported that PPR seroprevalence was significantly higher in male sheep. High seroprevalence in males could be due to

selling male sheep at young ages for slaughter for human consumption (Rony et al. 2017). Female sheep are kept by breeders for reproduction at older ages, which makes them more susceptible to infection with the disease (Tajpara et al. 2022).

The present study recorded a significantly ($P<0.001$) higher seroprevalence of PPR in sheep less than six months of age as compared to other age groups (Fig. 2). High seroprevalence of PPR in young age animals could be attributed to not having sufficient immunity yet to protect. In addition, passive immunity could be weak if the ewes are not vaccinated (Dubie et al. 2022; Zafar et al. 2024) or if they were not previously exposed to the disease. These findings are consistent with the findings of Ozkul et al. (2002) and Algezoli et al. (2024), who recorded an increase in positive antibodies in the serum of sheep for PPR with increasing age.

The current study showed that if the sheep source was from the local market, it had higher seroprevalence compared to sheep born on the farm ($P<0.001$). This finding confirms the role of the entry of new animals into the herd as a source of infection spread, which has also been reported by Almeshay et al. (2017). In addition, there is an increased risk of infection if new sheep are not quarantined or are purchased and added to the herd from an illegal source (Ejigu et al. 2023).

The current study also confirmed that PPR's seroprevalence is higher in farming rearing methods than in non-farming rearing systems ($P<0.001$) and higher in stall grazing feeding systems than grazing feeding systems. These findings contradict those of researchers (Bwihangane et al. 2016) in Congo, who reported that non-farming rearing systems are more susceptible to infection than farming-rearing systems. In fact, the density of breeding in farming-rearing systems is higher than in non-farming-rearing systems, which helps spread the disease.

Conclusion

The peste des petits ruminants in sheep in Hama Governorate, Syria, is considered an endemic disease. Because of its high seroprevalence, it may turn into a widespread epidemic in the region. Several factors also predispose to the occurrence of PPR, such as the geographical and environmental region, sex, age, and source of sheep, in addition to care and feeding systems. We propose Implementing preventive vaccination campaigns against PPR for sheep residing throughout Syria to limit the spread of the disease.

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Conflict of interest

The authors declare that they have no financial benefits or personal relationships that could have influenced the research for this article.

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