



# Sero-Surveillance of Peste des Petits Ruminant (PPR) in Goats at Different Haor-Oriented Areas in Sylhet Division of Bangladesh

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# ABSTRACT

**Background and Objective:** Peste des Petits Ruminants (PPR) is a viral transboundary disease of small ruminants with high morbidity and mortality. A study was conducted to investigate the presence of viral antibodies in goats in different Haor-Oriented areas in the Sylhet Division. **Materials and Methods:** For this, blood samples were collected and serum separated from randomly selected eighty (20 goats from each Upazila) unvaccinated goats (devoid of history of PPR/Rinderpest vaccination and natural infection) from and Tahirpur and Dharamapasha Upazila of Sunamgonj District and Jaintapur and Bishwanath Upazila of Sylhet District of Sylhet Division. Serum antibody titer (competition percentage, CP value) was determined by a commercially available cELISA kit (ID Vet. PPR competition cELISA). **Results:** The overall seroprevalence of PPR in goats was 26.66%. Upazila wise seroprevalence was found 27.5%. District-wise seroprevalence is higher in goats of Sunamgonj District (32.5%) than Sylhet District (22.5%). **Conclusion:** It is concluded that high seropositive goats indicate a high prevalence of naturally circulating PPR virus in Haor (Howl) oriented areas, therefore a large-scale study would be conducted in the study area to formulate a strategic plan concerning the PPR control programme for Haor areas in Bangladesh.

# **KEYWORDS**

Seroprevalence, antibody, PPR disease, goats, Haor areas

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# INTRODUCTION

Peste des Petits Ruminants (PPR) is a viral disease of small ruminants caused by the PPR virus belonging to the family Paramyxoviridae, manifested by fever, oculonasal discharges, anorexia, necrotic stomatitis, fetid diarrhoea, enteritis and bronchopneumonia followed by either death or recovery<sup>1</sup>. Since the first



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report of PPR from the Ivory Coast, the disease has been reported in the Middle East, the Arabian Peninsula and most parts of Africa and Asia<sup>1</sup>. Among the South Asian subcontinent, PPRV under lineage IV was first reported in the southern part of India in 1987<sup>2</sup>. Subsequently, frequent outbreaks of PPR have been recorded in South Asian countries like Pakistan, Bhutan, Nepal, Afghanistan, India and Bangladesh. The total goat population of Bangladesh is approximately 23 M. In Bangladesh, the PPRV was first identified in 1993 during a severe PPR outbreak<sup>3</sup>. Now a day, the outbreak of PPR in goats occurs every year and is considered an endemic disease in Bangladesh. The country faces a huge economic loss due to PPR as morbidity and mortality are high in goats.

The livelihood of the peoples of the Haor (Howl) areas of Sunamgonj and Sylhet Districts has been facing different natural calamities such as flooding, flash rain etc even though these two districts lie in the bordered area of the country. Their economy moves around agriculture. Particularly during natural calamities (flash floods, heavy rain), crop production is hampered severely, that's why goat rearing become an important tool for exchanging currency. The goat population of the remote area did get veterinary services as a vaccination programme, due to natural calamities and poor communication. However, PPR is one of the main constrain of goat rearing in those areas. Therefore, knowing the status of the PPR disease in that particular area is important to take necessary action for the prevention and control of PPR disease as well as to the motivation of the farmers.

For the prevention and control of PPR, proper diagnosis as well as sero-monitoring, is crucial. For sero-monitoring virus neutralization test, virus-specific monoclonal antibodies in an immuno-capture Enzyme-linked Immuno-Sorbent Assay (ELISA) and sandwich ELISA are generally used<sup>4,5</sup>. But more specifically, PPRV antibodies can be identified by competitive ELISA (cELISA). Due to its circulating in nature, the PPRV can easily be spread from one place to another. Sometimes genetic mutation occurs in its genomic structure. Thus, in Bangladesh, to control this disease outbreak in small ruminants, regular sero-monitoring studies are needed to evaluate the status of any endemic disease, the vaccine efficacy and genomic changes of PPRV in different regions of Bangladesh. Few studies were conducted concerning sero-monitoring of PPR in different districts in Bangladesh but sero-monitoring of PPR of goats in Haor-Oriented District as Sunamgonj and Sylhet Districts has not been done previously<sup>6</sup>. Therefore, this study was designed for the detection of PPRV-specific antibodies in the serum of goats in the Haor area of Sunamgonj and Sylhet district of Sylhet Division by using cELISA.

#### MATERIALS AND METHODS

**Study area:** Four Haor-Oriented Upazila from two bordered districts of the country (Sylhet and Sunamgonj) were selected for this study based on zoogeography (flood prone), the density of animal population and PPR prone area by emphasizing the bordered and Haor (howl) areas. Dharamapasha is located at 24.9000°N 91.0167°E on the banks of the Kangsha River close to the Haor areas. Tānguār Hāor, the largest Haor of Bangladesh, lies between the Upazila of Dharamapasha and Tahirpur. Tahirpur is located at 25.0917°N 91.1750°E. Other haors, rivers and beels in Dharamapasha and Tahirpur include Surma River, Tagār Hāor, Dhārām Hāor, Ghāglājur River, Sārdā Bil, Kaimer Dair, Dhānkuniyā Hāor, etc. The study was conducted from July, 2020 to June, 2021.

**Selection of farmers:** A survey was conducted using a pre-structured questionnaire to collect data concerning farmers' mass awareness about PPR vaccination and PPR disease in goats to facilitate easy identification of unvaccinated and natural infection-free goats. Farmers belonging to remote areas, far away from local Veterinary hospitals are unaware of PPR vaccination and disease were selected for this study under the direct supervision of respective Upazila livestock officers, based on the information

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concerning natural PPR outbreaks and PPR vaccination programme conducted by Upazila livestock office. Selection of experimental animals for blood collection and serum separation: Unvaccinated animals (goats) that have no history of getting natural PPR infection throughout their life were selected and used for the collection of blood and serum separation.

**cELISA test for detection of antibody:** A total of 80 blood samples (20 from each Upazilla, irrespective of age and sex) were collected by jugular venipuncture without anticoagulant, kept for an hour at room temperature and then serum was separated and stored at -20°C until use. A competitive ELISA kit was used for the detection of antibodies against the PPR virus by competitive screening ELISA (ID screen PPR competition-ID. Vet. Innovative Diagnostics). Briefly, 40  $\mu$ L of dilution buffer-13 (supplied kit) was added to each well. Positive control (10  $\mu$ L) was poured into wells A1 and B1 and negative control in C1 and D1 and 10  $\mu$ L samples were poured into the remaining wells. ELISA plate was then incubated at 37°C for 45 min. Each well was washed manually 3 times with approximately 300  $\mu$ L of wash solution, prepared by diluting the wash concentrate (20X) with double distilled water (supplied kit). About 100  $\mu$ L of previously prepared single-strength conjugate was added to each well and incubated at room temperature for 30 min. Further wash was done and 100  $\mu$ L substrate solutions were added to each well and incubated for 15 min in a dark room. Stop solution (100  $\mu$ L) was added to stop the reaction. Optical Density (OD) values were recorded at 450 nm with an ELISA plate reader (BioTek, Inc.). The absorbance was converted to Competition Percentage (CP) using the following formula:

Competition (CP) (%) = 
$$\frac{\text{OD sample}}{\text{ODNC}} \times 100$$

where, NC is negative control, PC is positive control Test serum samples showing a CP value less than or equal to 35% were considered positive, greater than 35% and less than or equal to 45% were considered doubtful and greater than 45% were considered negative. The test was validated if the mean value of the negative control OD was greater than 0.7 and the mean value of the positive control (ODpc) was less than 30% of the ODNC.

**Data analysis and interpretation:** Data were analyzed and interpretation was made as per kit instruction. The average mean (%) of seropositive serum was calculated by descriptive statistics.

#### RESULTS

Four Haor-Oriented Upazila from two districts were visited and the farmers/owners of the non-vaccinated goats were motivated with the help of relevant Upazila livestock officers and supporting staff. Average 27.5% of the seropositive sample was found where, Upazila wise percentages of seropositive serum were 30, 35, 25 and 20 at Tahirpur, Dharamapasha, Jaintapur and Bishwanath Upazila of Sylhet Division of Bangladesh in Table 1.

| Name of the District and Upazila |                    | Total    | Number of    | Number of    |              |                 |         |
|----------------------------------|--------------------|----------|--------------|--------------|--------------|-----------------|---------|
|                                  |                    | number   | seropositive | seronegative | Seropositive | Average (%)     | Mean/   |
| Sunamgonj                        | Sylhet             | of serum | serum        | serum        | serum (%)    | (district-wise) | average |
| Tahirpur Upazila                 |                    | 20       | 06           | 14           | 30           | 32.5            |         |
| Dhārām-pasha Upazila             |                    |          | 07           | 13           | 35           |                 | 2.75    |
|                                  | Jayanta Upazila    |          | 05           | 15           | 25           | 22.5            |         |
|                                  | Bishwanath Upazila |          | 04           | 16           | 20           |                 |         |

Table 1: Upazila wise seropositive serum

### DISCUSSION

Despite no history of PPR or Rinderpest vaccination or Rinderpest outbreak in the study area and population, the overall seroprevalence was 27.5%. In sheep and goats in Bangladesh, overall seroprevalence of PPR in goats of 21% has been reported but varied greatly in different districts. Prevalence was highest in Jessore (49.4%) South-West district and lowest (6.3%) in Chittagong South-East district. Sero-prevalence was 26.7% in Rajshahi (North West), 12.5% in Mymensingh (North East), 10.5% in Dhaka (Central) and 20.0% in Sylhet (North East) which is very close to the findings of this research by Siddique et al<sup>7</sup>. In goats, 37.5% seroprevalence was reported in Saint Martin's Island of Bangladesh in another study by Bhanuprakash et  $al^8$ . In this study, seroprevalence was found at 32.5% in Sunamoni District, which is somewhat more than Sylhet district (22.5%). The reasons are not clear because both districts belong to hill tract and bordered area of Bangladesh, even more, prone to flash floods. As both Sunamgonj and Sylhet are Haor (Howl-water reservoir) oriented areas of the country, People are always facing poverty and natural calamity, so geomicrobiology, the density of goats population, poor communication and socioeconomic status might be the causes. Another factor may be the illegal dispatch of animals through India-Bangladesh border. The presence of seropositive antibodies against PPRV in goats indicates that PPR viruses were circulating in these animals in Bangladesh. The virus might be circulated to the study area from India as the study area (four Upazila belongs to two districts) are the bordered Upazila, close to India and highly connected to the neighbour country India through Tamabil land port, the prevalence of antibodies to PPRV in goats of 2.1, 51.8, 40.1, 47.5, 62.7 and 41.5%, respectively, at Meghalaya, Tamilnadu, Andhra Pradesh, Karnataka, Maharashtra and Gujarat of India. In another study, overall PPR antibody was in 82.60% of goats in Syangja district of Nepal<sup>9,10</sup> Seroprevalence of 21.8% against PPRV in five states (Andhra Pradesh, Gujarat, Jammu and Kashmir, Maharashtra and Rajasthan) of India. The antibody prevalence of PPRV in small ruminants in India was 33% in another study which also supports the findings of this study by Singh *et al.*<sup>11</sup>.

In this study, the presence of PPRV antibodies indicated that the PPR virus was circulating in the population, therefore, considering the geography of the Sylhet and Sunamganj District (as the hilly and bordered district with poor veterinary services), its lack of border security and the lack of PPR vaccination, it is suggested that as part of PPR control strategy the goat population should be vaccinated and incoming goats should be kept in quarantine.

## CONCLUSION

It is concluded that high seropositive goats indicate a high prevalence of naturally circulating PPR virus, therefore a large-scale study including sheep and even large ruminants would be conducted in Haor areas to formulate a strategy concerning PPR control programme.

#### SIGNIFICANCE STATEMENT

This study discovered the linkage between natural transmission of the PPR virus and the different predisposing factors of virus transmission such as close rearing of animals due to heavy rain, flash flood and poor veterinary services as the areas are Haor-Oriented with poor communication. This study will help the researchers to uncover the critical areas of natural circulation of the PPR virus, particularly in bordered flood-prone Haor areas, which may contribute to formulating a new strategy concerning the PPR control programme.

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