

# Haemato- Biochemical Analysis of Goats Naturally Infected with Peste Des Petits Ruminants

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## ABSTRACT

The study was aimed to examine the haematological and biochemical profile of goats naturally infected with Peste des petits ruminant (PPR). Severe disease was recorded in few villages of Kamrup and Nalbari districts of Assam affecting large population of goats causing death in many animals due to diarrhoea. Serum samples were taken from 15 animals showing clinical signs of PPRV infection (infected group) for haematological and biochemical analysis. In addition, 10 numbers of sera samples were also examined from apparently healthy animals (control group). Presence of antibodies against PPR virus was determined using competitive enzyme linked immunosorbent assay (c-ELISA). Haematological and biochemical analysis of blood profile of PPR positive serum samples (n=15) as well as control (n=10) were performed. Among the haematological parameters Hb, RBC and PCV showed a significant ( $P<0.05$ ) decrease while WBC, both Absolute and Relative Monocyte and relative lymphocyte showed a significantly ( $P<0.05$ ) increase in PPR infected group than those of control group. Among the biochemical parameters, it was observed that there was a statistically significant ( $P<0.01$ ) increase in creatinine and bilirubin and a significant decrease ( $P<0.01$ ) in glucose, protein, albumin, A-G ratio in infected goats compared to those of control group. Results of the present study indicated that infection with PPR in goats provide valuable data about the haematological and biochemical findings and also clarifies the pathogenesis of PPR infection.

**Keywords** —Blood Chemical Analysis, Antibodies, Peste Des Petits Ruminants, Competitive Enzyme Linked Immunosorbent Assay, Goats.

## 1. INTRODUCTION

Peste des petits ruminant (PPR) is currently an emerging, economically important transboundary and notifiable World Organization for Animal Health (OIE) viral disease of sheep

and goats. The disease is highly contagious and characterized by pyrexia, ocular and nasal discharges, necrotizing and erosive stomatitis, enteritis, diarrhoea and bronchopneumonia, followed by either death or recovery from the disease [1].

PPR was first reported in the Ivory Coast, West Africa and later from other parts of the world namely sub-Saharan Africa, the Middle East and Southern Asia ([2] and [3]). In India, PPR was first recorded in 1987 from Tamil Nadu [4] and it continues to be present in the Southern peninsular India until 1994. Later, a number of PPR outbreaks were reported from the northern states of India [5]. Now, PPR is enzootic in India as outbreaks occur in small ruminants regularly throughout the country ([5] and [6]) and is a major constraint in small ruminant production incurring huge economic losses (estimated to be INR 1,800 million (US\$ 39 million) annually in terms of morbidity, mortality, productivity losses with trade restriction ([7] and [6]). In India, severity of the disease is more pronounced in goats than in sheep with a combined susceptible population of about 200 million and thus it is one of the major threats to the small ruminant population of the country.

The etiological agent, Peste des petits ruminants virus (PPRV) has been classified under family Paramyxoviridae, Order Mononegavirales and Genus Morbillivirus. There is a single serotype of PPRV, but genetically grouped into four distinct lineages (I, II, III, and IV) based on partial sequence analysis of Fusion (F) gene [2].

The mortality and morbidity rates in small ruminants affected with PPR may vary. It has been reported that morbidity and mortality ranges between 10% to 80% and 0% to 90% in sheep and goats respectively ([8] and [9]).

Attention should be given to this disease because it has very high morbidity and mortality rate and causes severe economic losses. To describe the disease better and to make a fast and accurate diagnosis so that accurate symptomatic treatment and protection may be given, clinical biochemical and haematological findings need to be examined. Therefore, the present study was conducted since the biochemical and haematological parameters of PPR infected goats were not documented from this part of the country. Moreover, these parameters are needed to clarify the pathogenesis of PPR infection.

**1.1 Ethical Approval:** The prior approval from the Institutional Animal Ethical Committee was obtained for collection of blood from the animals in this study.

## 2. MATERIALS AND METHODS

A series of PPR outbreaks were observed in the Kamrup district of Assam affecting goat populations between February 2013 and April 2013. The outbreaks were investigated and according to the history taken from the owner, the goats suffered from nasal and ocular discharges and bloody diarrhoea. Many goats also died due to this disease. Fifteen goats naturally infected by PPRV were categorized infected group and 10 clinically healthy goats from the same locality were included as a control group.

### 2.1 Blood Collection

Approximately, 5ml of blood per goats was drawn aseptically from jugular vein, of which 1 ml of blood was transferred to a sterile vial containing EDTA (1mg/ml of blood) in order to estimate routine blood parameters. Remaining 4 ml of blood sample was transferred to vacutainer tube for serum separation. Serum samples obtained by centrifugation were properly labelled and preserved at -20°C for further use.

### 2.2 Serological Analysis.

Serum samples were analyzed for the presence of PPR antibodies using an approved competitive ELISA kit (ID Screen PPR Competition, Montpellier, France).

### 2.3 Haematological Analysis

Blood samples for haematological and serum biochemical changes in PPR infected goats were collected only once i.e. on the day of observation/visit. Blood was collected from jugular vein into EDTA containing tubes for hematological analysis. Blood samples with EDTA were used to determine red blood cell (RBCs), peak cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cell (WBCs), Lymphocyte (Absolute), Monocyte (Absolute), Granulocyte (Absolute), Lymphocyte (Relative), Monocyte (Relative), Granulocyte (Relative), Hb and Thrombocyte using Automatic hematology cell counter, HD-Consortorium.

### 2.4 Biochemical Analysis

The serum biochemical studies were performed on serum harvested from 4 ml blood in sterilized tubes. The biochemical parameters studied included total protein, total albumin, globulin, blood glucose, cholesterol, creatinine, uric acid,

serum bilirubin, blood urea nitrogen (BUN), conjugated bilirubin and unconjugated bilirubin were estimated spectrometrically in Sysmotic 20 using commercially available kits. Globulin and indirect bilirubin concentrations were obtained by subtracting the concentrations of albumin and direct bilirubin from total protein and total bilirubin concentrations, respectively.

### 2.5 Statistical Analysis

The student's t-test was used to compare the results of different haemato-biochemical parameters and statistical significance for a given parameter was determined between infected and apparently healthy goats. The results were presented as mean with the standard error and p-value <0.05 was considered significant.

## 3.RESULTS and DISCUSSION

For a quick diagnosis of PPR in endemic areas clinical and macroscopic findings may be helpful, but histo- pathological and serological examination are also need to be applied to definitive diagnosis of PPRV infection in outbreaks [10].

### 3.1 Clinical Signs

Animals were found to have mild to moderate mucopurulent, oculonasal discharges, and mild erosions within the nasal cavity. Evidence of increased faecal soiling of the hindquarters suggesting mild diarrhoea was also seen (Figure 1).



(A)



(B)

**Figure 1: Infected Goats showing typical symptoms of PPR.**

(A) Ocular and Nasal Discharge (B) Diarrhoea

### 3.2 Serological Findings

All the serum samples collected from infected lambs were tested by competitive ELISA and found to be positive for antibodies specific to PPRV. Contrastingly; there was no detectible antibody against PPRV in samples collected from control goats. Different workers have also used competitive ELISA for screening of sheep for presence of antibodies against PPRV ([11], [12] and [13]).

### 3.3 Haematological Findings

Hematological values obtained from both groups are given as mean and mean of the standard error (mean  $\pm$  SE) (Table 1). Significant differences in some of the parameters were obtained between infected and control groups. Hb, RBC, PCV, MCV, MCHC, MCH counts were low in infected group compared to that of control group, but only Hb, RBC and PCV showed a significant ( $P < 0.05$ ) decrease. WBC, both Absolute and Relative Monocyte and relative lymphocyte values were significantly ( $P < 0.05$ ) higher in infected group than those of control group (Table 1).

### 3.4 Biochemical Findings

Biochemical parameters of control and infected groups are presented in Table. II. According to the biochemical analysis, there were statistically significant ( $P < 0.05$ ) decrease in glucose, protein, albumin and A: G ratio in samples obtained from infected goats compared to those of control group while serum creatinine and bilirubin showed a significant ( $P < 0.05$ ) increase in the infected goats. The rest of the parameters did not show much variation.

Reference [11] reported that multifocal areas of coagulative necrosis and vacuolation of hepatocytes in sheep naturally infected with PPRV. Ours data showed that the infected sheep also had significant ( $P < 0.05$ ) decreased protein and albumin level. This finding is in agreement with the findings of Reference [11] and [14].

**Table-1:** Haematological Parameters in Control and PPRV Infected Goats.

Variable	Control Group		PPR Group	
	N	Mean± S.E	N	Mean± S.E
Hb(gm/dl)	1 0	9.00±1.054 <sup>a</sup>	1 5	6.3±0.76 <sup>b</sup>
Thrombocyte(M/m <sup>3</sup> )	1 0	377.66±87.4 5 <sup>a</sup>	1 5	341.331±43.8 7 <sup>a</sup>
Erythrocyte indices				
RBC (x10 <sup>12</sup> /l)	1 0	15.91±0.27 <sup>a</sup>	1 5	13.55±0.67 <sup>b</sup>
MCV(fl)	1 0	13.74±0.81 <sup>a</sup>	1 5	10.94± 1.20 <sup>a</sup>
MCH(pg)	1 0	5.69±0.09 <sup>a</sup>	1 5	4.84±0.43 <sup>a</sup>
MCHC(g/dl)	1 0	42.63±2.46 <sup>a</sup>	1 5	49.76±4.66 <sup>a</sup>
PCV(%)	1 0	21.78±1.14 <sup>a</sup>	1 5	14.51±1.73 <sup>b</sup>
Leucocyte indices				
WBC (M/mm <sup>3</sup> )	1 0	8.29±0.19 <sup>a</sup>	1 5	14.51±0.42 <sup>b</sup>
Lymphocyte (Absolute)	1 0	54.29±0.92 <sup>a</sup>	1 5	55.46±2.14 <sup>a</sup>
Monocyte (Absolute)	1 0	3.74±0.19 <sup>a</sup>	1 5	5.17±0.23 <sup>b</sup>
Granulocyte (Absolute)	1 0	41.97±0.98 <sup>a</sup>	1 5	39.36±2.32 <sup>a</sup>
Lymphocyte (Relative)	1 0	4.27±0.13 <sup>a</sup>	1 5	7.089±0.39 <sup>b</sup>
Monocyte (Relative)	1 0	0.27±0.01 <sup>a</sup>	1 5	0.69±0.04 <sup>b</sup>
Granulocyte (Relative)	1 0	3.75±0.24 <sup>a</sup>	1 5	6.73±0.50 <sup>b</sup>

Mean with different superscripts indicates significant difference between rows (P<0.05)

**Table-2:** Biochemical Parameters in Control and PPRV Infected Goats.

Variable	Control Group		PPR Group	
	N	Mean±Std	N	Mean ±Std
Glucose(mg/dl)	10	53.74±3.09 <sup>a</sup>	15	44.30±1.82 <sup>b</sup>
Protein(g/dl)	10	7.41±0.23 <sup>a</sup>	15	5.65±0.25 <sup>b</sup>

Albumin(g/dl)	10	4.49±0.13 <sup>a</sup>	15	2.76±0.89 <sup>b</sup>
Globulin(g/dl)	10	2.92±0.25 <sup>a</sup>	15	2.64±0.18 <sup>a</sup>
AG Ratio	10	1.46±0.13 <sup>a</sup>	15	1.12±0.08 <sup>b</sup>
Cholesterol(mg/dl)	10	67.70±4.36 <sup>a</sup>	15	71.92±3.37 <sup>a</sup>
BUN(mg/dl)	10	13.43±1.03 <sup>a</sup>	15	15.73±1.39 <sup>a</sup>
Urea(mg/dl)	10	30.12±2.30 <sup>a</sup>	15	35.17±3.16 <sup>a</sup>
Creatinine(mg/dl)	10	1.18±0.18 <sup>a</sup>	15	2.52±0.26 <sup>b</sup>
Uricacid(mg/dl)	10	2.60±0.42 <sup>a</sup>	15	3.17±0.34 <sup>a</sup>
Bilirubin(mg/dl)	10	0.19±0.04 <sup>a</sup>	15	0.38±0.05 <sup>b</sup>
Conbilirubin(mg/dl)	10	0.09±0.02 <sup>a</sup>	15	0.14±0.02 <sup>a</sup>
Unconjugated bilirubin(mg/dl)	10	0.10±0.03 <sup>a</sup>	15	0.21±0.04 <sup>a</sup>

Mean with different superscripts indicates significant difference between rows (P<0.05) Peste-des-petits-ruminants is an important disease of small ruminants causing great economic losses due to their high morbidity and mortality [15] and threats national live- stock industries [16]. PPR infection is still a major problem as sporadic outbreaks occur among sheep, goats and wild small ruminants [17]. The disease is characterized by digestive and respiratory system disorders ([18] and [19]). In present study, the infected goats also showed the same clinical signs and are parallel with the reports of [11],[13] and [14] also reported a significant (P<0.05) decrease in RBC and PCV that correlates with our study. The WBC, lymphocytes, monocytes, granulocytes have increased due to infection with PPR virus. Conversely, [11] and [20] determined low WBC and lymphocyte value in infected lambs and sheep.. The differences between these hematological parameters may be due to the different phase of the disease , presence of secondary infections and nutrition or dehydration degree [12] . Similarly, results were also reported byreference [11] where they also reported that total bilirubin increased significantly (P<0.05) during PPR . The total serum protein, albumin values decreased significantly (P<0.05) which is similar with the findings of [11]who also reported that during PPR, the liver function test showed abnormal result which may be due to dysfunction of the liver during PPR. This may be the reason for decrease in protein and albumin concentration in our study.

In conclusion, in the present study, PPRV infection cause considerable erosive-ulcerative lesions on various organs and alter haematological and biochemical parameters in goats. Therefore, it is suggestive that the infection impairs the

function of many organs or systems such as liver, skin, oral cavity and digestive and respiratory systems. These valuable data obtained in the present study may be helpful in the diagnosis and treatment (symptomatic) of the disease. In addition, these clinical, haematological and biochemical parameters obtained should be kept in mind by the veterinary practitioners for the possible occurrence of new outbreaks in the region and for its diagnosis.

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