DETECTION OF *PESTE DES PETITS RUMINANTS* VIRUS ANTIBODIES IN SERA OF CATTLE *

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Abstract

A total of 120 serum samples of cattle collected from six districts in Tamil Nadu were screened for PPRV antibodies. Competitive enzyme linked immune sorbent assay and micro serum neutralization test were used for seromonitoring of PPRV antibodies. In areas with no previous history of PPR incidence and PPRV vaccination in goats and sheep under field/ farmers holdings, the serum samples collected from selected cattle herds showed 8.3% positivity for PPRV antibodies by both cELISA and micro SNT, whereas in areas with previous history of PPR incidence and PPRV vaccination in goats and sheep, the cattle herds showed 18.3% positivity.

Key words: PPR, Vaccination, Tamil Nadu, Cattle, Immune response.

INTRODUCTION

Peste des petits ruminants virus (PPRV) causes natural disease in goats and sheep. Gibbs *et al.* (1979) had first classified PPRV as the fourth member of the genus *Morbili virus*. The genus *Morbilli virus* comes under the family *Paramyxoviridae* of the order *Mononegavirales* (International Committee on Taxonomy of Viruses, 2017). The virus is closely related to Rinderpest virus (RPV), another member of *Morbilli virus* genus, which causes similar disease in large ruminants (Anderson *et al*.,1990 ; Couacy-Hymann *et al*., 1995). It is generally considered that cattle alone are naturally infected sub-clinically, although in the 1950s, disease and death were recorded in calves experimentally infected with PPRV-infected tissue (OIE, 2012).

Antibodies against PPRV in cattle can provide cross protection against RPV (Taylor, 1979). Because of its close relationship to RP, PPR was suggested to be taken into account in the rinderpest control programmes (Lefevre and Diallo, 1990). Infection of cattle with PPRV also interferes with immune response against RPV (Anderson and McKay, 1994). With the successful eradication of rinderpest, global strategy for the control and eradication of PPR is being contemplated (Singh *et al.,* 2009 / 2011; OIE–FAO, 2015; Banyard and Parida, 2016; FAO-OIE, 2016). The aim of the present study is to seromonitor PPRV antibodies in cattle sera in Tamil Nadu.

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MATERIALS AND METHODS

During the study period, the serum samples were collected from selected herds of cattle in Thiruvallur, Kancheepuram and Tiruvannamalai districts in those areas there was no previous history of PPR incidence and PPRV vaccination in goats and sheep. The details of the samples collected are given in table 1.

Table 1. Details of cattle sera collected under field / farmers holdings in areas with no previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	No. of serum samples		
	Thiruvallur District	Kancheepuram District	Tiruvannamalai District
Cattle (below 1 year)	10	10	10
Cattle (above 1 year)	10	10	10

During the study period, the serum samples were collected from Pudukottai, Thoothukudi and Tirunelveli districts in the selected herds of cattle from areas with previous history of PPR incidence and PPRV vaccination in goats and sheep. The details are given in table 2.

Table 2. Details of cattle sera collected under field / farmers holdings in areas with previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	No. of serum samples		
	Tirunelveli District	Thoothukudi District	Pudukottai District
Cattle (below 1 year)	10	10	10
Cattle (above 1 year)	10	10	10

The PPR virus competitive enzyme linked immunosorbant assay **(**cELISA) test kit was procured from Indian Veterinary Research Institute (IVRI), Mukteswar, India and the test was carried out as per the protocol. The principle of competitive ELISA test is based on the inhibition of binding of monoclonal antibody to antigen in the presence of PPR antibody in the test sera. This results in reduced colour development, when anti-mouse antibody conjugated to HRPO is used for tracing the binding of monoclonal antibody. This monoclonal antibody is directed against a neutralising epitope of Hemaggultinin (HA) protein of PPR virus. As this is a solid phase assay, washing is required between each step to ensure removal of unbound reagents.

Plates were read in a microplate reader µQuant (Biotek) at 492 nm filter. The reader was connected to a computer loaded with

KC software specific for the ELISA plate reader which was used to acquire the readings. The optical density (OD) values were converted to percentage inhibition by using the following formula:

PI=100-(absorbance of the test sample /absorbance of Cm wells) X100

Test Sera showing more than 40% PI of mean OD values of the Cm (monoclonal antibody control) wells were taken as positive for PPR antibodies.

The micro serum neutralisation test (mSNT) was performed in Vero cells for the detection of antibodies to PPRV with a fixed dilution of 1:8 for each test sera sample as per the method described in Rossiter *et al* (1985); Balamurugan *et al* (2012) and OIE (2012).

RESULTS AND DISCUSSION

cELISA responses (Mean ± SE) of serum samples collected for seromonitoring of PPRV antibodies in selected herds of cattle in the areas without previous history of PPRV disease incidence and PPRV vaccination in goats and sheep under field/ farmers holdings are presented in Table 3.

Table 3. PI values of cELISA for PPRV antibodies in cattle sera collected from areas with no previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	PI values (Mean±SE) for PPRV antibodies in cELISA		
	Thiruvallur District	Kancheepuram District	Tiruvannamalai District
Cattle (below 1 year)	Negative	45.76	Negative
Cattle (above 1 year)	46.83±0.43	49.31±0.52	45.98±0.57

The mean PI values was calculated only from those sera samples that showed positive PI values (>40) in cELISA

cELISA responses (Mean ±SE) of serum samples collected for seromonitoring of PPRV antibodies in selected herds of cattle in the areas with previous history of PPRV disease incidence and PPRV vaccination in goats and sheep under field/ farmers holdings are presented in Table 4.

Table 4. PI values of cELISA for PPRV antibodies in cattle sera collected from areas with previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	PI values (Mean±SE) for PPRV antibodies in cELISA		
	Tirunelveli District	Thoothukudi District	Pudukottai District
Cattle (below 1 year)	51.52	48.63	Negative
Cattle (above 1 year)	59.73±0.41	58.24±0.43	60.16±0.48

The mean PI values was calculated only from those sera samples that showed positive PI values (>40) in cELISA

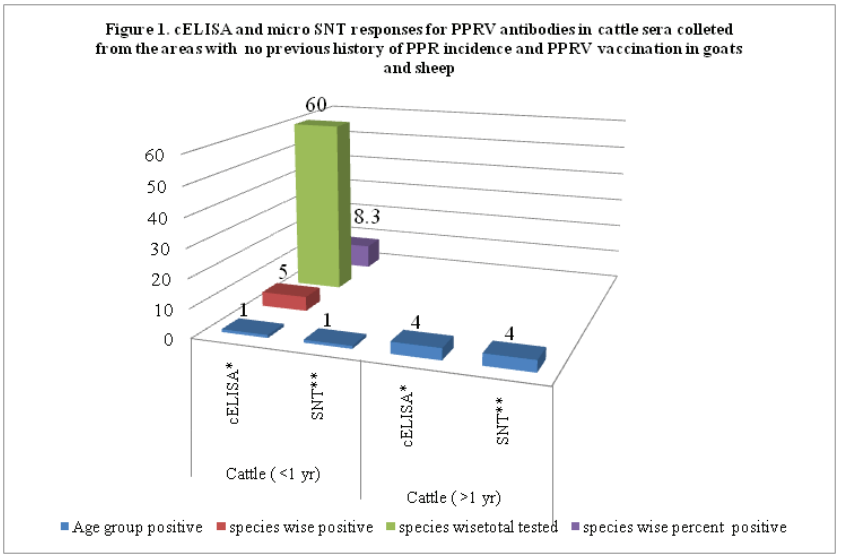
cELISA and micro SNT responses of serum samples collected for seromonitoring of PPRV antibodies in selected herds of cattle in areas without previous history of PPRV disease incidence and PPRV vaccination in goats and sheep under field/ farmers holdings are presented in Table 5 and shown in Figure 1 which showed 8.3% positivity .

Table 5. cELISA and micro SNT responses for PPRV antibodies in cattle sera collected from areas with no previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	Test	No. of animals positive/ No of animals tested			Age group wise Total No.	Species wise Total Positive by both	Species wise percent
		Thiruvallur District	Kancheepuram District	Tiruvannamalai District			
Cattle (< 1 yr)	cELISA*	0/10	1/10	0/10	1/30	5/60	8.3
	SNT**	0/10	1/10	0/10	1/30		
Cattle (> 1 yr)	cELISA*	2/10	1/10	1/10	4/30		
	SNT**	2/10	1/10	1/10	4/30		

* No. of sera positive / No. of animals tested in cELISA

** No. of sera showing ≥ 1 in 8 titre in micro SNT / No. of animals tested



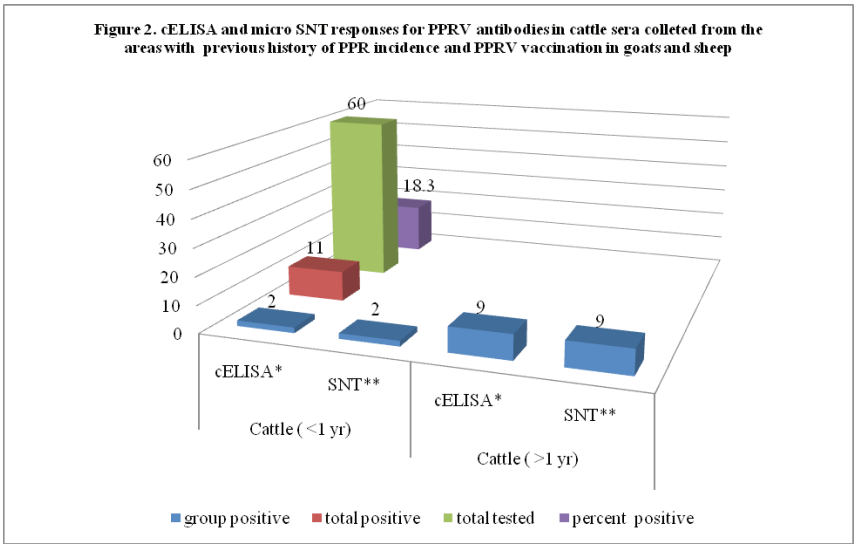
cELISA and micro SNT responses of serum samples collected for seromonitoring of PPRV antibodies in selected herds of in the areas with previous history of PPRV disease incidence and PPRV vaccination in goats and sheep under field/ farmers holdings are presented in Table 6 and presented in Figure 2 which showed 18.3% positivity.

Table 6. cELISA and micro SNT responses for PPRV antibodies in cattle sera collected from areas with previous history of PPR incidence and PPRV vaccination in goats and sheep

Species	Test	No. of animals positive/ No of animals tested			Age group wise Total No. Positive / Total No. tested	Species wise Total Positive by both cELISA and SNT / Total No. Tested	Species wise percent positive
		Tirunelveli District	Thoothukudi District	Pudukottai District			
Cattle (< 1 yr)	cELISA*	1/10	1/10	0/10	2/30	11/60	18.3
	SNT**	1/10	1/10	0/10	2/30		
Cattle (> 1 yr)	cELISA*	3/10	4/10	2/10	9/30		
	SNT**	3/10	4/10	2/10	9/30		

* No. of sera positive / No. of animals tested in cELISA

** No. of sera showing ≥ 1 in 8 titre in micro SNT / No. of animals tested



Cattle can be infected, but they do not seem to develop clinical signs and are not known to transmit PPRV to other animals, which appear to be dead-end hosts (www.cfsph.iastate.edu, 2015). Although cattle are unable to transmit the disease to another host, sero-conversion against the PPRV H protein has been observed (Khan *et al*., 2008).

Interestingly, epidemiological surveillance studies carried out in different enzootic regions have revealed PPRV seroprevalence in other ruminants including cattle, buffalo and camel. This seroprevalence can be as high as 67% in case of cattle. Cattle are considered as potential dead-end hosts for PPRV. It appears however that this virus, for reasons not yet elucidated, can occasionally overcome the innate resistance of these species, resulting in the development of clinical signs (Naveen kumar *et al*., 2014).

The prevalence of PPR virus antibodies among domestic animals (goat, sheep, cattle and camel) populations in NE Nigeria was studied by El-Yuguda *et al.* (2013), using virus neutralisation test (VNT) and c-ELISA. In VNT, seroprevalence in cattle was 16.7%. Similar pattern of prevalence was noted when the sera were tested for PPR antibodies using c-ELISA.

Balamurugan *et al.* (2014) studied the prevalence of PPRV antibodies in cattle, buffaloes, sheep and goats carried out during the period 2011 using the serum samples randomly collected from different villages of five states in India. A total of 605 cattle serum samples were collected from 52 districts in five states (Andhra Pradesh, Gujarat, Jammu and Kashmir, Maharashtra and Rajasthan) of India and screened for PPRV-specific antibodies by using PPR monoclonal antibody-based competitive ELISA kit. Analysis of 605 samples showed PPRV seroprevalence of 11.07 % in cattle.

Muthuchelvan *et al.* (2015) reported seroprevalence rate of PPRV at country level in goats and sheep as 43.56 % and 4.58% in cattle and buffaloes, respectively.

CONCLUSION

Cattle sera showed the prevalence of PPRV antibodies as high as 18.3% in Tamil Nadu indicating infection of PPRV without clinical signs. The presence of PPRV antibodies demonstrated that bovines are exposed to PPRV infection and it implied the importance of cattle as subclinical hosts for the virus, besides widespread presence of the disease in sheep and goats. Though the cattle are considered to be the dead end host, it is recommended to vaccinate the cattle when they are reared along with sheep and goats in the premises for National PPRV control to be successful.

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