Short communication

Seroprevalence of Small Ruminant Lentiviruses (SRLVs) and *Peste des petits* Ruminants virus (PPRV) in Nonvaccinated Sheep and Goats in Sarawak, Malaysia

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ABSTRACT

Small ruminant lentiviruses (SRLVs) and Peste des Petits Ruminants Virus (PPRV) are economically important viral diseases that threaten small ruminant production globally. While previous studies have reported their serological status in Peninsular Malaysia, there is limited epidemiological information on their occurrence in East Malaysia. This study aimed to determine the seroprevalence and risk factors of SRLVs and PPRV among nonvaccinated sheep and goats from selected smallholder farms in Sarawak. A total of 245 serum samples collected from sheep (n=117) and goat (n=128) were screened using commercial ID Screen® PPR competition (PPRC ver. 0821) and MVV/CAEV indirect (VISNAS ver. 0922) ELISA test kits. The results of ELISA assays indicate 7.4% (95% CI: 4.7-11.3) apparent and 6.9% (95% CI: 4.0-11.3) true seroprevalence for SRLVs and 5.7% (95% CI: 3.4-9.4) apparent and 5.1% (95% CI: 2.6-9.1) true seroprevalence for PPRV in the sample. Further exploratory univariable logistic regression analysis revealed that PPRV seropositivity was significantly associated with the farm management system (OR= 4.176, 95% CI: 1.14-15.36, p = 0.031), indicating a higher risk of exposure in semi-intensive management system. To the best of our knowledge, this study provides the first preliminary serological evidence of SRLVs and PPRV among nonvaccinated smallholder sheep and goat flocks in Sarawak, suggesting the need for surveillance and full scale epidemiological study.

Keywords: Small ruminant; Peste des Petits Ruminants; Small ruminant lentiviruses, Seroprevalence; Sarawak

INTRODUCTION

Caprine arthritis encephalitis virus (CAEV) and Maedi-Visna virus (MVV) of the genus *Lentivirus* and family *Retroviridae* are collectively called small ruminant lentiviruses (SRLVs) (Larruskain & Jugo, 2013). Transmission in sheep and goats occurs through ingestion of infected colostrum or via aerosolized respiratory secretions (Davies *et al.,* 2023), causing persistent multisystemic disease involving chronic arthritis and encephalitis syndrome in CAE or progressive respiratory disease and wasting due to meningoencephalitis in Maedi-Visna (MV) (Barták *et al.,* 2018; Kalogianni *et al.,* 2021). Both CAE and MV are globally recognized as economically significant diseases (Ali *et al.,* 2016; Araújo *et al.,* 2020; Gosselin *et al.,* 2018; Jimale *et al.,* 2024; Mekibib *et al.,* 2018; Michiels *et al.,* 2020) causing decreased milk yield, impaired reproductive performance, increased disease susceptibility, and premature culling of small ruminants (Davies *et al.,* 2023; Jimale *et al.,* 2024). Given the global distribution and economic importance of SRLVs coupled with its prevalence among domestic and wild ruminants in West Malaysia (Balakrishnan *et al.,* 2025; Jesse *et al.,* 2018; Jimale *et al.,* 2024; Paul *et al.,* 2021), it is timely to investigate its occurrence among farm animals in Sarawak.

Peste des Petits Ruminants (PPR) is a severe and highly contagious disease caused by the PPR virus (PPRV), a *Morbillivirus* which is closely related to the Rinderpest virus (Singh & Bandyopadhyay, 2015). PPR occurs globally as a notifiable transboundary animal disease (TAD) of small ruminants (Clemmons *et al.,* 2021) and it is currently targeted for global eradication by the FAO and WOAH in 2030 (Cano-Terriza *et al.,* 2020; Fentie *et al.,* 2018; Gelana *et al.,* 2020). It is an acute febrile disease manifesting signs of anorexia, depression, coughing, tachypnoea, oculonasal discharges, mouth ulcers, and diarrhoea, followed by death or full recovery (Balamurugan *et al.,* 2014). However, despite the economic significance and the urgent need to eradicate PPR globally, there are only two published reports on its prevalence status among small ruminants (Jimale *et al.,* 2024) and deer (Balakrishnan *et al.,* 2025) in Malaysia, suggesting potential exposure of both wild and domestic ruminants to the antigens of PPRV in the country requiring further investigation, especially in the Borneo region.

Although there are several published reports suggesting possible exposure to SRLVs and PPRV antibodies in small ruminant (Jesse *et al.,* 2018; Jimale *et al.,* 2024; Paul *et al.,* 2021) and deer herds (Balakrishnan *et al.,* 2025) in West Malaysia, to the best our knowledge, there is no published report on the serological status of SRLVs and PPRV among sheep and goat flocks in the Borneo region and Sarawak in particular. To address this, a preliminary cross-sectional survey was undertaken to assess the seroprevalence and potential determinants of SRLVs and PPRV in selected smallholder sheep and goat farms in Sarawak, Malaysia. It was hypothesized that lower prevalence rates of SRLVs or PPRV are linked to specific explanatory variables associated with smallholder sheep and goat flocks within the region.

MATERIALS AND METHODS

Study design and sampling

This study was approved by the Institutional Animal Care and Use Committee (UPM/IACUC/AUP-R037/2023) Universiti Putra Malaysia and the individual farmers also gave their consent to participation. This study focused on nonvaccinated sheep and goat flocks kept by smallholder farmers under semi-intensive or oil palm-integrated farming systems in Bintulu (3.1739° N, 113.0428° E) and Miri (4.4180° N, 114.0155° E) divisions of Sarawak. The farmers did not use vaccines, and the local veterinary authority is not implementing any vaccination programme against SRLVs or PPRV in the study area. The sample size used for this study was calculated using the formula $n = (Z^2 \times P \times (1 - P))/d^2$, where Z = standard normal distribution for 95% confidence interval (Z=1.96 for 95% CI), P is the expected true proportion = 20.6% (Paul *et al.,* 2021), d is the desired precision (5% absolute precision, representing half the desired confidence interval width) (Thrusfield, 2005). Although 260 samples were initially collected to increase

precision, nine were excluded due to incomplete data, poor serum separation, or spillage, leaving 245 valid samples (sheep = 117; goats = 128) for analysis. The farms were selected based on farmer consent, but sampling units were selected randomly to minimize selection bias. Pregnant and very young animals were excluded from the study because the farmers declined due to personal reasons. Blood samples were collected from sheep or goat by jugular venipuncture after restraining each animal in a standing position on a non-slip surface with the head and neck extended and stabilised to expose the jugular vein. Using gentle digital pressure, the jugular vein was partially occluded at the base of the neck. A vacutainer tube containing clot activator and fitted with sterile 18G 1.5″ hypodermic needle was gently inserted into the jugular vein at an angle of 30 degrees cranially to collect approximately 5 mL of whole blood. Each sample was properly labelled, kept undisturbed in a cold box, and transported to the laboratory where the clotted blood samples were centrifuged at 3000 rpm for 5 mins to collect sera in 1.5 mL microcentrifuge tubes. The sera were temporarily held at -20 °C in the serum bank at the Clinical Research Laboratory, Faculty of Veterinary Medicine, Universiti Putra Malaysia, prior to the serological assays.

Detection of serum antibody by ELISA

Commercial ID Screen® PPR competition (PPRC ver 0821 with a label specificity = 99.4% and sensitivity = 94.5%), and indirect MVV/CAEV (VISNAS ver 0922 with label specificity = 98.9% and sensitivity = 91.7%) ELISA screening tests were used for the detection of serum antibodies against PPRV and SRLVs in the test sera according to manufacturer instructions. The absorbance was quantitatively measured using Infinite® F50 microplate reader (Tecan Trading AG, Switzerland) at a wavelength of 450 nm to record the optical densities (OD) of the ELISA test medium. Samples with a S/N % OD ≤ 50% were considered positive for PPRV competition, while samples with an S/P % OD ≥ 60 % were considered positive for SRLVs.

Statistical analysis

The EpiTools statistical software (Sergeant, 2018) was used to calculate the apparent and true (TP = (AP+Sp-1)/Se+Sp-1) seroprevalence and their respective confidence intervals (CI) using Rogan and Gladen method (Rogan & Gladen, 1978) based on the sensitivity and specificity of PPRC ver 0821(Sp = 99.4% and Se = 94.5%) and VISNAS ver 0922 (Sp = 98.9% and Se = 91.7%) commercial ELISA test kits (IDvet, Montpellier, France). The univariable binary logistic regression was computed with Statistical Package for Social Sciences (SPSS) using SRLVs or PPRV serological status as dependent variable and the species, gender, age, physiological stage, prophylactic measures, and management system of small ruminants as independent variables. The odds ratios, corresponding 95% confidence intervals (CIs), and p-values for different categories of explanatory

variables were tabulated. The proportion of missing data for each explanatory variable was minimal and unlikely to have a significant impact on overall estimates. All the records with missing information were excluded from the analyses to minimise bias.

RESULTS AND DISCUSSION

The results of ELISA assay for SRLVs revealed that 18/245 sheep (7; 6.0%) and goats (11; 8.6%) were positive suggesting an overall 7.4% (95% CI: 4.7-11.3) apparent and 6.9% (95% CI: 4.0-11.3) true seroprevalence **(Table 1)**. In terms of PPRV, antibodies were detected in 14/245 sheep (8; 6.8%) and goats (6; 4.7%) indicating an overall 5.7% (95% CI: 3.4-9.4) apparent and 5.1% (95% CI: 2.6-9.1) true seroprevalence (**Table 2**). Further exploratory analysis using univariable binary logistic regression of PPRV serological status and the explanatory epidemiological variables showed a significant higher risk of infection in the semi-intensive management system (OR= 4.176, 95% CI: 1.14-15.36, p = 0.031) (**Table 3**). In terms of SRLVs, there was no significant associations between the seropositivity and the epidemiological variables of sheep and goats (**Table 4**), suggesting an equal chance of exposure and susceptibility among the examined sheep and goats due to similar management and environmental conditions (Paul *et al.,* 2021). The observed seroprevalence of SRLVs and PPRV in this study indicates a very low level of exposure. Although seropositivity does not confirm present or past clinical infection (Ghanem *et al.,* 2009), the observation of serum antibodies among farm animals in this study is epidemiologically important since there is no routine vaccination against SRLVs or PPRV in the state of Sarawak. Technically, this finding suggests the absence of herd immunity, past natural exposure, or silent subclinical circulation of the viruses within the small ruminant population. During outbreaks PPR leads to 100% morbidity and 90% mortality with survivors becoming life-long carries, serving as reservoirs of infection in the herd (Santhamani *et al.,* 2016). Similarly, chronic SRLVs infection produce subclinical life-long carriers which serve as permanent reservoirs of infection in the herd (Plaza *et al.,* 2009).

The present 6.9% seroprevalence of SRLVs among sheep and goats agrees with previous studies that reported 6.0% in Somalia (Ghanem *et al.,* 2009) and 6.4% and 6.7% in goat and sheep in Brazil (Alves *et al.,* 2017). However, the result of this study is lower than other countries such as Iraq with 65% (Hamza & Özkan, 2017), Thailand with 75.9% (Panneum & Rukkwamsuk, 2017), Belgium with 95% (Adjadj *et al.,* 2019), and Italy with 100% (Crespo *et al.,* 2016). Furthermore, our result is also lower than 8.8% (Jesse *et al.,* 2018), 21.4% (Paul *et al.,* 2021), and 45.4% (Jimale *et al.,* 2024) seroprevalence rates among sheep and goats from West Malaysia. Although our studies in West Malaysia employed the same ID Screen® ELISA testing system used in the current study, the significantly lower

seroprevalence of SRLVs observed in the current study may be explained by differences in geographical location, management practices, genetic composition and structure of the small ruminant population, and origins of the livestock (Jesse *et al.,* 2018; Jimale *et al.,* 2024; Paul *et al.,* 2021). Additionally, the higher seropositivity in the West Malaysian studies may be linked with a more intensive management practice, faulty biosecurity, uncontrolled cross border movement of animals, and poor hygienic measures (Ghanem *et al.,* 2009) coupled with the practice of rearing sheep and goats together increasing the risk of interspecies transmission (Gjerset *et al.,* 2009). Other researchers have also observed and reported that intensive management practices such as vaccination, animal identification, and milking operations increase within herd risk of SRLVs transmission (Barquero *et al.,* 2011; Tu *et al.,* 2017).

The observation of 5.1% (95% CI: 2.6-9.1) seroprevalence of PPRV in sheep and goats is almost similar to the result of a previous study in West Malaysia (Jimale *et al.,* 2024). The current result is however significantly lower than endemic proportions reported in developing countries such as 16.2% in Ethiopia (Mebrahtu *et al.,* 2018), 29.0% in Cameron (Awa *et al.,* 2002), 35.8% in Pakistan (Zahur *et al.,* 2011), 60.0% in Jordan (Al-Majali *et al.,* 2008), and 76.5% in Nigeria (El-Yuguda *et al.,* 2013). According to the WOAH, PPR is a significant infectious TAD of major concern to small ruminant producers (Cano-Terriza *et al.,* 2020) in developing countries of Africa, Asia, and the Middle East (Baron *et al.,* 2017; Kumar, 2014; Santhamani *et al.,* 2016), where its high prevalence is accompanied by severe losses in productivity and mortality (Cano-Terriza *et al.,* 2020). PPR is a highly contagious and rapidly spreading disease that frequently occurs as outbreaks in sheep and goats farms (Lembo *et al.,* 2013). The spread of disease and propagation of outbreaks in developing countries is facilitated by cross border animal movement and trading (Balamurugan *et al.,* 2019). Seasonal factors, especially prolonged draught, forcing herders to move animals over long distances to find feed also leads to outbreaks (Abubakar *et al.,* 2009). The increased risk of exposure to PPRV among semi-intensively managed sheep and goats in our study disagrees with a previous study which reported that high frequency of animal movement and communal grazing were associated with increased risk of exposure to PPRV under extensive system (Wendimu *et al.,* 2024). However, a multivariable logistic regression analysis was not performed to control for any confounding variables due to the presence of only one significant variable and small sample size, limiting the translational value of this result. Therefore, in the future, further studies using larger representative samples will enhance our understanding of disease risk in the population.

In conclusion, this study is to the best of our knowledge the first study to describe the serological status of SRLVs and PPRV sheep and goat flocks in East Malaysia. The low levels of SRLVs and PPRV exposure detected among

nonvaccinated small ruminants is suggestive of possible spread. Due to the limitation in interpretation and applications of serology, further comprehensive epidemiological investigations are required for isolation and molecular characterization of the viruses and elucidate their risk factors toward designing data-driven control and eradication program in line with the WOAH and FAO mandates.

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Table 1. Sample Distribution and the results of ELISA for SRLVs amongst small ruminants.

Variable	Tested	Positive	Apparent prevalence		True prevalence	
			Estimate (%)	95% CI	Estimate (%)	95% CI
Species						
Goat	128	11	8.6	4.9-14.7	8.3	4.2-15.1
Sheep	117	7	6.0	2.9-11.8	5.4	2.0-11.9
Gender						
Male	81	6	7.4	3.4-15.2	7.0	2.6-15.6
Female	164	12	7.3	4.2-12.4	6.9	3.5-12.4
Age						
Young (up to 1yr)	48	4	8.3	3.3-19.6	8.0	2.4-20.4
Adult (Above 1 yr)	197	14	7.1	4.3-11.6	6.6	3.5-11.6
Production phase						
Young stock	52	4	7.7	3.0-18.2	7.3	2.1-18.8
Adult male	37	4	10.8	4.3-24.7	10.7	3.5-26.1
Adult female	100	6	6.0	2.8-12.5	5.4	1.9-12.6
Lactation	56	4	7.1	2.8-17.0	6.7	1.9-17.5
Prophylactic measures						
Yes	125	11	8.8	5.0-15.1	8.5	4.3-15.4
No	120	7	5.8	2.9-11.6	5.2	1.9-11.5
Management						
[1]Semi intensive	119	9	7.6	4.0-13.8	7.1	3.2-14.0
[2]Integrated	126	9	7.1	3.8-13.0	6.7	3.0-13.2
Total	245	18	7.4	4.7-11.3	6.9	4.0-11.3

CI: confidence interval
[1]Semi-intensive: raised animals in group pens within a slated-roofed and zero grazing.
[2]Integrated: raised animals in oil palm plantation and allowed to graze occasionally.

Table 2. Sample Distribution and the results of ELISA for PPRV amongst small ruminants.

Variable	Tested	Positive	Apparent prevalence		True prevalence	
			Estimate (%)	95% CI	Estimate (%)	95% CI
Species						
Goat	128	6	4.7	2.2-9.9	4.0	1.2-9.7
Sheep	117	8	6.8	3.5-12.9	6.3	2.7-13.0
Gender						
Male	81	4	4.9	1.9-12.0	4.2	0.9-12.1
Female	164	10	6.1	3.4-10.9	5.5	2.5-10.8
Age						
Young (up to 1yr)	48	1	2.1	0.4-10.9	1.1	0.1-10.8
Adult (Above 1 yr)	197	13	6.6	3.9-11.0	6.1	3.1-10.9
Production phase						
Young stock	52	4	7.7	0.3-18.2	7.6	2.6-18.7
Adult male	37	1	2.7	0.5-13.8	2.2	0.5-14.1
Adult female	100	6	6.0	2.8-12.5	5.8	2.3-12.7
Lactation	56	3	5.4	1.8-14.6	5.1	1.3-14.9
Prophylactic measures						
Yes	125	5	4.0	1.7-9.0	3.6	1.2-9.0
No	120	9	7.5	4.0-13.6	7.4	3.6-13.9
Management						
[1]Semi intensive	119	11	9.2	5.2-15.8	9.2	4.9-16.2
[2]Integrated	126	3	2.4	0.8-6.8	1.9	0.2-6.6
Total	**245**	**14**	**5.7**	**3.4-9.4**	**5.1**	**2.6-9.1**

CI: confidence interval

[1]Semi-intensive: raised animals in group pens within a slated-roofed and zero grazing.

[2]Integrated: raised animals in oil palm plantation and allowed to graze occasionally.

Table 3. Univariable logistic regression analysis for the associations of PPRV serological status and the epidemiological variables of small ruminants in Sarawak.

Variables	Examined	Positive	Negative	OR (95% CI)	*p*
Species					
Sheep	128	6 (4.7)	122 (95.3)	1.492 (0.50-4.44)	0.471
Goat	117	8 (6.8)	109 (93.2)	**1.0 (Reference)**	
Gender					
Female	81	4 (4.9)	77 (95.1)	1.250 (0.38-4.11)	0.714
Male	164	10 (6.1)	154 (93.9)	**1.0 (Reference)**	
Age					
Young (up to 1yr)	48	1 (2.1)	47 (97.9)	3.321 (0.42-26.03)	0.253
Adult (Above 1 yr)	197	13 (6.6)	184 (93.4)	**1.0 (Reference)**	
Production phase					
Young stock	52	4 (7.7)	48 (92.3)	1.472 (0.31-6.92)	0.913
Adult male	37	1 (2.7)	36 (97.3)	0.491 (0.05-4.91)	
Adult female	100	6 (6.0)	94 (94.0)	1.128 (0.27-4.69)	
Lactation	56	3 (5.4)	53 (94.6)	**1.0 (Reference)**	
Prophylactic measures					
Yes	125	5 (4.0)	120 (96)	1.946 (0.63-5.98)	0.245
No	120	9 (7.5)	111 (92.5)	**1.0 (Reference)**	
Management					
[1]Semi intensive	119	11 (9.2)	108 (90.8)	4.176 (1.14-15.36)	0.031*
[2]Integrated	126	3 (2.4)	123 (97.6)	**1.0 (Reference)**	

CI: confidence interval, *P*-values with an asterisk (*) are statistically significant.
[1]Semi-intensive: raised animals in group pens within a slated-roofed on zero grazing.
[2]Integrated: raised animals in oil palm plantation and allowed to graze occasionally.

Table 4. Univariable logistic regression analysis for the associations of SRLVs serological status and the epidemiological variables of small ruminants in Sarawak.

Variables	Examined	Positive	Negative	OR (95% CI)	*p*
Species					
Goat	128	11 (8.6)	117 (91.4)	1.477 (0.55-3.95)	0.436
Sheep	117	7 (6.0)	110 (94.0)	**1.0 (Reference)**	
Gender					
Female	81	6 (7.4)	75 (92.6)	1.013 (0.37-2.81)	0.98
Male	164	12 (7.3)	152 (92.7)	**1.0 (Reference)**	
Age					
Young (up to 1yr)	48	4 (8.3)	44 (91.7)	1.188 (0.37-3.79)	0.77
Adult (Above 1 yr)	197	14 (7.1)	183 (92.9)	**1.0 (Reference)**	
Production phase					
Young stock	52	4 (7.7)	48 (92.3)	1.083 (0.26-4.57)	0.913
Adult male	37	4 (10.8)	33 (89.2)	1.576 (0.37-6.74)	
Adult female	100	6 (6.0)	94 (94.0)	0.830 (0.22-3.07)	
Lactation	56	4 (7.1)	52 (92.9)	**1.0 (Reference)**	
Prophylactic measures					
No	125	11 (8.8)	114 (91.2)	1.558 (0.58-4.16)	0.377
Yes	120	7 (5.8)	113 (94.2)	**1.0 (Reference)**	
Management					
[1]Semi intensive	119	9 (7.6)	110 (92.4)	1.064 (0.41-2.78)	0.900
[2]Integrated	126	9 (7.1)	117 (92.9)	**1.0 (Reference)**	

CI: confidence interval, *P*-values with an asterisk (*) are statistically significant.
[1]Semi-intensive: raised animals in group pens within a slated-roofed on zero grazing.
[2]Integrated: raised animals in oil palm plantation and allowed to graze occasionally.