

Original Article

A study on transmission of Peste des petits ruminants virus between dromedary camels and small ruminants

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Abstract

Introduction: In recent years Peste des petits ruminants (PPR) disease caused several epidemics in a wide range of susceptible hosts. The ability of the peste des petits ruminants virus (PPRV) to cross the species barrier necessitates further research, particularly on disease circulation and cross-species transmission between typical and atypical hosts to guide and facilitate the eradication program anticipated by the Food and Agriculture Organization (FAO) and the World Organization for Animal Health (OIE) in 2030. The aim of this study is to explore the role of dromedary camels as transmitters for PPR.

Methodology: Four experiments were carried out on clinically healthy seronegative camels, sheep and goats. In experiment I, the animals were inoculated with a PPR- positive suspension of camel pneumonic lung homogenate. In the other three experiments either sheep and goats were inoculated and after three days were housed with camels or vice versa.

Results: Marked clinical signs suggestive of PPR were seen in sheep and goats while camels showed mild infection. Severe clinical signs of PPR were seen in sheep and goats when kept with inoculated camels. Postmortem examination revealed PPR lesions in all inoculated animals including camels.

Conclusions: This study showed that dromedary camels infected with PPRV can transmit the disease to sheep and goats, even when they developed mild clinical signs.

Key words: PPR, camel, sheep, goat, infection, transmission.

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Introduction

Peste des petits ruminants (PPR) is a severe fast spreading disease affecting mainly domestic small ruminants and characterized by sudden onset of depression, fever, discharge from the eyes and nose, soreness in the mouth, disturbed breathing and cough, foul smelling diarrhea and death [1]. This disease is one of the most economically important trans-boundary animal diseases as it causes death of small ruminants, hence impacting the livelihoods, food security and nutrition of millions poor people in developing countries. Outbreaks of PPR in sheep and goats in Africa and Asia are extensively reported.

According to OIE- World Animal Health Information System (WAHIS) data, PPR outbreaks have been reported in almost all countries except those in the southern region of Africa, including Zimbabwe, Botswana, Namibia, and South Africa. In Asia, outbreaks of PPR have been reported in Afghanistan, Bahrain, Bhutan, Iraq, Israel, Kuwait, Mongolia, Nepal, Oman, Palestine, Tajikistan, Turkey, United Arab Emirates, Yemen as depicted in OIE-WAHIS data, Pakistan [2-4], Bangladesh [5], India [6], Iran [7] and China [8,9].

PPR is mainly a disease that affects small ruminants, however reports describing the infection in

camels are continuously published from different production zones. Ismail *et al.* were the first to detect PPR antibodies in Sudanese camels in Egypt [10]. Roger *et al.* reviewed a new acute febrile epizootic disease characterized by highly contagious respiratory syndrome with elevated morbidity and low mortality rates in camel in Ethiopia during 1995-1996 [11]. Formal contacts revealed the presence of a similar camel disease incidence in Djibouti, Somalia, Eritrea, and Sudan. Detection of PPR virus (PPRV) antigen by antigen-detection enzyme-linked immunosorbent assay (ELISA) and antibodies by antibody-detection ELISA and PPRV RNA by reverse transcription polymerase chain reaction (RT-PCR) have been reported in these outbreaks. Roger *et al.* detected PPR antibodies in 7.8% of camels after occurrence of respiratory outbreak [12]. In Sudan PPRV antibodies were detected in camel sera [13-15]. In August 2004, a massive outbreak of disease with diarrhea and respiratory problems and up to 50% mortality in camels was reported in eastern Sudan. PPRV antigen and genes were detected in affected animals using immunocapture ELISA assay (Ic-ELISA) and RT-PCR, respectively, and partial genome sequences obtained [16,17]. It was noticed that PPR outbreak had been reported in sheep in the same area prior to the camel outbreak [16].

Recently, PPR outbreaks have been reported in camel in Iran [18] and Kenya [19]. These recent disease outbreaks necessitate investigating the exact role camels may play in the epidemiology in the affected zones. Within the atypical hosts of PPR such as pigs, buffaloes, and wild animals, the camels are distinctive in that they are geographically distributed within the PPR endemic areas of the world and herded together with the typical host, the small ruminants, in most parts of Africa and Asia and their numbers are increasing significantly. Accordingly, this work is intended to explore this debatable role of camels through experimental infection of camel, sheep, and goats by PPR virus originating from camel and sheep.

Methodology

Animals

All animals used in the four experiments were clinically healthy, belonging to local breeds, and sourced from areas with no history of either PPR vaccination or outbreak. All animals were screened seronegative for PPR using competitive ELISA (c-ELISA) [20]. The kit was obtained from the French Agricultural Research Centre for International Development (CIRAD), Montpellier, France. The OIE/FAO/European Union (EU) reference Laboratory

designed and validated the kit. Dromedary camels (*Camelus dromedarius*) of the local Arabi breed at the age of 2-3 years were brought from Central Sudan (Gezira state) and Western Sudan (Kordofan state). Sheep (*Ovis aries*) of the local Sahrawi breed in Sudan at the age of 8-12 months and Goats (*Capra aegagrus hircus*) of the Nubian goat breed in Sudan at the age of 8-12 months were brought from Khartoum state. The experiments were carried out in the closed semi-isolated animal house at the Central Veterinary Research laboratories (CVRL) in Khartoum, Sudan, with strict Biosecurity measures, including wearing protective clothing and rubber boots. Animals were kept at the animal house for 3 weeks prior to the experiments, during which they received anti-parasitic medicines and multivitamins, and were observed for general health condition. The rectal body temperature was recorded every day at a fixed time in the morning. The means of body temperature during the acclimatization period were 36.2 ± 2.2 °C. Between each experiment (I, II, III and IV) the animal house was cleaned and disinfected twice by formalin fumigation before the introduction of new animals.

Inocula preparation

Inoculation of animals was done by the subcutaneous route using a suspension of lung homogenate prepared using slaughterhouse-collected camel or sheep lung infected with PPR virus. A 20% tissue homogenate in phosphate buffered saline (PBS) containing antibiotic and antifungal solution was prepared. After centrifugation at 400 g for 10 minutes, the supernatants of the camel and sheep lung homogenates were collected. The homogenates were tested for PPR antigen using immunocapture ELISA (Ic-ELISA) and RT-PCR, as described below, and found positive. Furthermore, the inocula were checked for the presence of bovine herpes virus-1 (BHV-1), bovine viral diarrhea (BVD), respiratory syncytial virus (RSV) and parainfluenza virus 3 (PIV 3) using a Sandwich ELISA kit (tetra-pulmotest) according to the manufacturer's instructions (Bio-X Diagnostics, Jemelle, Belgium) and both lung homogenates from camel and sheep were found to be free of these respiratory pathogens.

Experimental design

The animal experiments were approved by the Ethical Committee on Animal Experiments of the Central Veterinary Research Laboratories (CVRL), Sudan. Animals, PPR virus used, and experimental protocols are summarized in Table 1. In experiment I,

we infected camels, sheep, and goats with a camel-derived PPRV to determine host susceptibility. To determine the animal host-to-animal host contact transmission, we conducted the following experiments. In experiment II-a, 3 sheep were infected with sheep-derived PPRV and were placed in contact with 3 camels, while in II-b, 2 goats were infected with sheep-derived PPRV and were placed in contact with 2 camels. In experiment III-a, 3 sheep were infected with camel-derived PPRV and were placed in contact with 3 camels, while in III-b 2 goats were infected and placed in contact with 2 camels. In the last experiment (IV), 5 camels were infected with camel derived-PPRV and were placed in contact with 5 sheep and 5 goats. Experimentally infected animals were introduced to in-contact animals 3 days post infection. In all experiments, animals were observed daily for the clinical signs of PPR. Body temperature was measured daily in the morning from day 0 of post infection (pi) till the day of the slaughter (21 pi). Blood samples were collected from all animals every 2 days for white blood cells (WBCs) count and weekly for PPR antibodies detection. Slaughtering of all surviving animals was done on day 21 pi.

Detection of PPR antigen using Ic-ELISA

Lung, spleen, and lymph node samples of dead and slaughtered animals from the four experiments (n = 144) were examined for PPR antigen using Ic-ELISA. The Kits were obtained from CIRAD, Montpellier, France and the test was performed according to the manufacturer’s instructions.

Detection of PPR N gene using RT-PCR

RNA was extracted from the tissue samples using Qiagen RNeasy Kits (Qiagen, Hilden, Germany), as instructed by the manufacturer. Lung samples of inoculated animals in each experiment were tested for PPR genome using RT-PCR. PCR was done using Qiagen One-step RT/PCR Kit (Qiagen, Hilden, Germany). The primers NP3 and NP4, which were selected for the determination of the C-terminus of the N gene of PPRV [21]. Five µL of RNA was amplified in a 45µL reaction mix containing: 10µL of 5X Qiagen buffer, 2µL of dNTP mix, 10µL of Q-solution, 2µL of One Step RT-PCR Mix, each primer at a final concentration of 0.6 µM, and water. Thermal cycler conditions were as follows: reverse transcription 30 minutes at 50 °C, initial PCR activation during 15 minutes at 95 °C, followed by 40 cycles of amplification corresponding to 30 seconds at 94°C, 30 seconds at 60 °C, 1 minute at 72°C and final extension of 10 minutes at 72 °C. RT-PCR resulted in an amplification product of 351 bp. Ten µL of the amplified products were analyzed by electrophoresis in 1.5 % agarose gel.

Detection of PPR antibodies using c-ELISA

Collected sera from the four experiments (n = 192) were examined for PPR antibodies using c-ELISA obtained from CIRAD, Montpellier, France; the test was performed according to the manufacturer’s instructions. This test was carried out before the adaptation of this c-ELISA to camels and dromedaries in order to improve the detection of antibodies directed against PPR present in their serum. This task has been

Table 1. Study design of peste des petits ruminants virus (PPRV) experimental infection and transmission trials with dromedary camels, sheep and goats using camel and sheep- derived PPRV for intra-nasal infection. Experimentally infected animals were added to in-contact animals 3 days post infection.

Experiment	Inoculum	Animal inoculated	Number (ID* of inoculated animal)	In contact animals (Number) ID*
I-a	PPR-positive camel lung-homogenate	Dromedary camels	5 (I-a-1-5)	-
I-b	PPR-positive camel lung-homogenate	Sheep	5 (I-b 1-5)	-
I-c	PPR-positive camel lung homogenate	Goats	5 (I-c1-5)	-
II-a	PPR-positive sheep lung homogenate	Sheep	3 (II-a 1-3)	Dromedary camels (3), II-a D 1-3
II-b	PPR-positive sheep lung homogenate	Goats	2 (II-b 1-2)	Dromedary camels (2), II-b D 1-2
III-a	PPR-positive camel lung homogenate	Sheep	3 (III-a 1-3)	Dromedary camels (3), III-a D 1-3
III-b	PPR-positive camel lung homogenate	Goats	2 (III-b-1-2)	Dromedary camels (2), III-b D 1-2
IV	PPR-positive camel lung homogenate	Dromedary camels	5 (IV 1-5)	Sheep (5), IV-S 1-5 + Goats (5), IV-G 1-5

* ID: Identity of animal in the experiment; D: dromedary (*Camelus dromedarius*) at the age of 2-3 years; breed: local Arabi camel breed of Sudan; S: sheep (*Ovis aries*) at the age of 8-12 months, breed: local sheep breed of Sudan; G: goat (*Capra aegagrus hircus*) at the age of 8-12 months, breed: Nubian goat breed of Sudan.

done since then and has resulted in improving the competitiveness of the serum of the camelids (having only one antibody fragment out of two) so that it has better affinity relative to the monoclonal antibody used in the test.

Results

Experiment I: Host susceptibility

Dromedary camels

The five dromedary camels inoculated with PPR-positive camel lung homogenate remained clinically healthy, except for slight fever and leukocytosis. They showed slight increase in temperature (38.6 – 38.8 °C) on days 7-11 pi. Total WBCs count decreased slightly (5750- 7500 cells/μL) on day 4 pi, and then markedly increased in four camels (16700 – 22200 cells/μL), reaching up to 37,000 cells/μL in one camel (ID-2) at day 14 pi.

Sheep

All sheep inoculated with PPR-positive camel lung-homogenate showed leukocytosis that reached up to 20,200 cell/μL in three animals (I-b 2, 3, 4), and fever (38.9-39.0 °C) followed by depression, nasal and lacrimal discharges, and later mouth ulcers and diarrhea. Sheep IS2 showed fever (39.0 °C) and died on day 8 pi.

Goats

In goats inoculated with PPR-positive camel lung-homogenate, typical clinical signs of PPR were observed. In all goats, total WBCs decreased (7500–8900 cells/μL) on day 4 pi, and then markedly increased up to 28,400 cell/μL. High fever (> 39 °C), coughing, and nasal discharge started at 3 pi followed by abdominal breathing and lacrimation. Besides these symptoms, two goats (I-c 2, 3) showed slight diarrhea.

No deaths were recorded till the day of slaughtering (21 pi).

Postmortem (PM) findings in experiment I

The inoculated camels, sheep and goats were euthanized on the day 21 pi. The postmortem observations in euthanized, and the dead sheep (I-b 2) were mainly congestion in lung, spleen, liver, intestine (enteritis), colon, slight congestion in kidneys, some tonsils and lymph nodes and blood vessels supplying the digestive organs. The spleen in some sheep and goats was found enlarged. Severe congestion was seen in some lymph nodes, especially the mesenteric ones.

Detection of PPR antigen in tissue samples using Ic-ELISA and confirmation by RT/PCR

Lung, spleen, and lymph node samples were collected during postmortem at 21 pi, homogenized, and processed for PPRV detection using Ic-ELISA. Positive results were obtained in samples collected from all the 15-inoculated sheep, goats, and camels (Table 2 experiment I). Three samples out of five from each animal species were tested using in RT/PCR for PPRV genome. All tested samples were found positive.

Detection of PPR antibodies using c-ELISA

Antibodies against PPRV were detected in all sheep and goat sera collected 3 weeks after the experiments, however none of camel sera were positive.

Experiment II

Dromedary camels housed with sheep infected with PPR-positive sheep lung homogenate (II-a-D)

All inoculated sheep developed symptoms similar to those seen in experiment I-b without mortality. All camels housed with infected sheep showed slight increase in temperature, between 38.5 °C and 38.8 °C on day 7. The temperature was 38.9 °C on day 19 pi in

Table 2. Summary for experiments I: II: III and IV of PPRV antigen detection through Ic-ELISA in lung: spleen and lymph node samples of camels: sheep and goats.

Experiment	Camel (positive/tested)			Sheep (positive/tested)			Goat (positive/tested)			Total (positive/ tested [%])
	Lung	Spleen	Lymph node	Lung	Spleen	Lymph node	Lung	Spleen	Lymph node	
I-a	5/5	5/5	5/5	-	-	-	-	-	-	15/15 [100]
I-b	-	-	-	5/5	5/5	5/5	-	-	-	15/15 [100]
I-c	-	-	-	-	-	-	5/5	5/5	5/5	15/15 [100]
II-a	3/3	3/3	3/3	2/3	3/3	2/3	-	-	-	16/18 [88.9]
II-b	2/2	2/2	2/2	-	-	-	2/2	2/2	2/2	12/12 [100]
III-a	3/3	2/3	1/3	2/3	2/3	3/3	-	-	-	13/18 [72.2]
III-b	2/2	1/2	1/2	-	-	-	2/2	2/2	1/2	9/12 [75]
IV	4/5	4/5	4/5	3/5	3/5	3/5	3/5	3/5	3/5	30/45 [66.7]
Total (%)	19/20 (95)	17/20 (85)	16/20 (80)	12/16 (75)	13/16 (81.3)	13/16 (81.3)	12/14 (85.7)	12/14 (85.7)	11/14 (78.6)	125/150 [83.3]

one camel (II-a 4). Leukocytosis (14,600, 22,400 and 19,600 cells/ μ L) was detected in 3 camels (experiment II-a 1, 2, 3).

Dromedary camels housed with goats infected with PPR-positive sheep lung homogenate (II-b-D)

Symptoms similar to those seen in experiment I-c were seen in the infected goats. Four out of five in-contact camels showed slight fever and leukocytosis.

Detection of PPR antigen in tissue samples using Ic-ELISA

Testing lung samples for PPR antigen using Ic-ELISA revealed that all infected sheep and goats were positive, except for one sheep (II-a 2). Ic-ELISA indicated positive results in lung samples collected from all camels exposed to infected sheep and goats (Table 2, experiment II).

Detection of PPR antibodies using c-ELISA

Antibodies against PPRV were detected in all sheep and goat sera collected at 3 weeks after the experiments, however none of camel sera were found to be positive.

Experiment III

Dromedary camels housed with sheep infected with PPR-positive camel lung homogenate (III-a D)

All inoculated sheep developed symptoms like those seen in experiment I-b without mortality. All camels housed with infected sheep showed slight rise in temperature ranged between 38.7–39.0 °C, as well as leukocytosis (13500–15700 cells/ μ L).

Postmortem (PM) finding

In-contact camels were euthanized on day 19 pi and on PM showed congestion in intestine and pneumonia lesion in lung.

Detection of PPR antigen in tissue samples using Ic-ELISA

Testing tissue samples for PPR antigen using Ic-ELISA showed positive results in three camels (III 1, 4, 5), one sheep (III-a1) and two goats (Table 2, experiment III).

Detection of PPR antibodies using c-ELISA

All sheep and goat sera collected after the experiments were found positive for PPR antibodies.

Experiment IV

Sheep housed with dromedary camels infected with PPR-positive camel lung homogenate (IV-S)

All infected camels showed mild rise in temperature (38.0–38.6 °C) starting at day 5–6 pi as well as leukocytosis. Sheep housed with infected camels

showed variable clinical signs including fever, depression, coughing, nasal and lacrimal discharges, and diarrhea. Two sheep (IV S 1, 4) died at days 4-7 after housing with infected camels.

Goats housed with dromedary camels infected with PPR-positive camel lung homogenate (IV-b-G)

All infected camels showed mild rise in temperature and leukocytosis (13900-5800 cells/ μ L). Goats housed with infected camels showed similar clinical signs observed in sheep. These symptoms disappeared on day 16-17 after housing with infected camels and there was no mortality.

PM findings

The camels, sheep and goats were euthanized on the day 21 pi. The postmortem findings for euthanized and dead animals were congestion in liver and intestine, pneumonia in lung and enlargement of mesenteric lymph nodes.

Detection of PPR antigen in tissue samples using Ic-ELISA

Four out of five infected camels (IV D 1, 2, 4, 5), 3 out of 5 in-contact sheep (IV S 1, 4, 5) and 3 out of the 5 in-contact goats (IV G 1, 2, 3) were found positive for PPR antigen using Ic-ELISA (Table 2).

Detection of PPR genome using RT-PCR

Some of PPR Ic-ELISA positive samples (n = 12 lung samples; 7 camels, 3 sheep and 2 goats) were selected for PPRV genome detection using QIAGEN One step RT/PCR Kit and PPRV-specific primers. All the samples tested positive for PPR genome (data not shown).

Detection of PPR antibodies using c-ELISA

Sera were collected from all camels, sheep and goats 21 days after the experiments and tested for PPR antibodies. Only sheep and goat sera were found positive.

Discussion

The advantage of doing transmission experiments compared to field studies is that they offer a controlled environment in which the effect of a single factor can be investigated, while variation due to other factors is minimized [22]. Evidence of clinical PPRV infection in camels and seroconversion are continuously being reported from endemic regions in Africa, the Middle East and Asia [23]. In this work, we explored the possible role of camels in the transmission of PPR. We carried out four experiments to infect sheep, goats and

camels with PPRV originating from camel and sheep. Experimental infections produce clinical signs which are specific for PPR disease [24-25] and are comparable to naturally infections [5,25-27]. Host responses are reflected by initial leukopenia followed by leukocytosis. Similar observation has been recorded by other authors [28-29]. The postmortem lesions seen in sheep and goats in the four experiments matched with the classical lesions caused by the virus previously reported in naturally as well as experimentally infected sheep and goats [30-33]. Antibodies against PPR were detected in all sheep and goats used in the four experiments with elevating titres in the three weeks, representing strong positive reaction. Interestingly, all camel sera in this study, collected till the end of the experiments, tested negative for PPR antibodies. This is contradictory to the antigen detection results which could be explained by the unusual nature of camel antibodies previously reported [34]. We noticed the same during the massive PPR outbreak in camels in 2004 [16]. In most of the previous reports, antibodies to PPR in camels were either detected in low levels [12,14,15,35-37] or it was not detected [38-41]. In this study, camels were however confirmed to be susceptible and able to replicate the virus as demonstrated, whether by inoculation or by in contact experiments. Since this study, the standard operating procedure of the c-ELISA kit that was originally validated for sheep and goats was modified to detect antibodies directed against the PPRV nucleoprotein in camelid serum or plasma, thus improving sensitivity of the kit to camel species. The test is commercialised by Innovative Diagnostics (IDVet, Grabels, France) and uses technology developed by CIRAD, Montpellier, France. Therefore, we highly recommend testing and validating the currently available ELISA kits for the detection of PPRV specific antibodies in atypical hosts, especially the camels.

Throughout the four experiments, artificially infected camels showed mild clinical signs and remained apparently healthy. There was slight rise in temperature and evidence of immune reactions based on the decrease in total WBCs count followed by marked increase, indicating that PPRV in experimental trials does not cause disease in dromedary camels but induces a low level of virus replication that is still detectable at 21 pi as demonstrated by the detection of PPR antigen in tissue samples using Ic-ELISA and confirmation by RT/PCR.

This result corroborates previous findings demonstrating that experimental infection of dromedary camels with PPRV does not produce

apparent clinical signs. The only experimental infection that resulted in mild respiratory disease with coughing, nasal discharge and fever was reported by Harrak *et al.* [28]. Wernery experimentally infected camels with PPRV and reported the failure to produce clinical signs [42]. Fakhri *et al.* observed no clinical infection of PPR in camels after experimental exposure and claimed that camels play no epidemiological role in the spread of the disease [43]. Recently, Schulz [44] showed that camelids including the dromedary camels developed no clinical signs, no viremia, shed no or low PPRV-RNA loads in swab samples and did not transmit any PPRV to the contact animals when experimentally infected with the virulent PPRV lineage IV (LIV)-strain Kurdistan/2011. It is concluded that PPRV causes subclinical infection in dromedary camels without apparent PPR clinical symptoms. The slight variations in the magnitude of the pathology induced by the virus could be related to the strain of the virus used as an inoculum, dosage, or the route of inoculation. However, more research is needed to determine the role played by these factors.

The most critical elements in the infection of dromedary camels with PPR are virus shedding and the transmissibility to the typical host such as sheep and goats. In the present study, we demonstrated severe clinical disease in sheep and goats housed with dromedary camels infected with PPRV prepared from camel lungs pointing to virus shedding from infected camels. Additionally, camels infected with PPRV from sheep can transmit the disease back to sheep.

These results contradict the recent findings by Schulz *et al.* who found that camels did not transmit any PPRV to the contact animals when experimentally infected with the virulent PPRV [44]. The failure of the dromedary camels experimentally infected with PPRV in the experiments by Schulz *et al.* could be related to the biological properties of the strain PPRV lineage IV (LIV)-strain Kurdistan/2011 primarily isolated in northern Iraq where there was no incidence of camel infection with PPRV or the breed of camels involved [44]. In a similar situation, Nawathe and Taylor previously performed an experimental investigation that revealed a subclinical infection of PPRV in pigs with no shedding and transmission of the virus to goats [45]. Later, Schulz *et al.* performed transmission trials with the same virus strain used in the above camel experiment (PPRV lineage IV (LIV)-strain Kurdistan/2011) and revealed that pigs and wild boar are possible sources of PPRV-infection to goats [46].

Sequence analysis of PPRV isolates obtained from camels and other atypical hosts with emphasis on the

glycoprotein HN gene, which is necessary for the virus attachment to and entry into the cells is expected to shed light on potential mutations that enable the virus to infect animal species other than small ruminants.

Contrary to the outcome of experimental infection of PPRV in dromedary camels which show subclinical disease, outbreaks of PPR in camels have been reported from four camel-raising countries (Ethiopia, Iran, Kenya and Sudan). Stress conditions in the field such as seasonal movement in search for green pasture coinciding with severe PPR in the comingling sheep flocks [16] could contribute to the appearance of the disease in camels. Interestingly, no further disease outbreaks were reported from Sudan, despite abattoir studies on slaughtered camels have reported detection of PPRV antigens in lung tissue with gross lesions, indicating a history of pneumonia [21,47,48]. Therefore, it is possible that after the primary severe outbreaks an endemic situation develops characterized by subclinical infection as animals build up immunity that can minimize the symptoms during further attacks of the virus [49].

The most significant finding presented in this work is that camels can develop subclinical infection by PPRV and can transmit it to sheep and goats. In agreement with the conclusion of Rahman *et al.* [21] that camels do not appear to be a dead-end host, the virus transmissibility between sheep, goats and camels needs to be considered when designing control measures for PPR.

Conclusions

This study provides unequivocal evidence of PPRV transmission from sub-clinically affected camels to in-contact sheep and goats highlighting the neglected role of the camels in transmitting this economically important disease.

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Authors' Contributions

IKS, GL, AD, YHA designed and supervised the study. IKS, MAH, SMA, SMM, BAM, KMT performed the experiments and lab work. YHA, AIK drafted the manuscript. IKS analyzed and interpreted the data. GL, AD, AIK revised and approved the manuscript.

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