Review Article

Special Issue: Molecular Virology and Control of *Peste des Petits Ruminants* Virus

Eradicating *Peste des Petits Ruminants* - The Challenges Ahead

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Abstract | *Peste des petits ruminants* virus causes an economically important disease of small ruminants that causes extensive livestock losses across areas where it is endemic. Further the recent description of the disease in naïve populations of animals have demonstrated an expansion of the distribution of the disease. Having been in the shadow of rinderpest virus for decades little is understood regarding factors that influence the outcome of infection with both viral and host genetics being thought to influence the outcome of disease. Following the successful global eradication of rinderpest virus, *peste des petits ruminants* virus has also been targeted for global eradication by the OIE. Whilst the tools are available to attempt eradication there are a number of factors that will influence the strategy taken to progress with the aim of eradication. Here we describe current areas where knowledge improvement is required to shape our understanding of the disease and its distribution alongside areas of investigation that require consideration in any viral eradication campaign.

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Peste des petits ruminants virus (PPRV) has been endemic across much of Africa and Asia since it was first described as a viral entity distinct from rinderpest virus (RPV) several decades ago (Gibbs et al., 1979). PPRV was in the shadow of rinderpest virus for centuries and until the 1950's little consideration was given to this plague of small ruminants and its effect on small ruminant agriculture. Indeed PPRV was likely confused with rinderpest for decades if not centuries prior to the characterisation of the virus as a distinct viral entity. In the last ten years PPRV has become of increased interest to the veterinary and scientific community, not least due to the successful global eradication of RPV. As such, probably as a result of the increased focus on PPRV as a pathogen of economic significance, the virus has been reported more frequently in areas where it has long been considered to be endemic as well as in areas where the virus has never been described before (Banyard et al., 2014; OIE, 2015; OIE, 2016). Its expansion into regions bordering the EU have been of particular interest (OIE, 2016; Parida, 2016). Furthermore, due to both an increased availability of molecular tools in countries where the virus is endemic and an increased interest in PPRV following RPV eradication outbreaks of the virus have been genetically typed enabling an epidemiological assessment of lineage distribution across endemic areas. This in turn has led to

speculation surrounding the emergence of a dominant lineage of the virus in new areas (Kwiatek et al., 2011). Certainly, the recognition of the importance of PPR as an obstacle to the development of sustainable agriculture across Africa and Asia, areas where for the majority of countries PPR is considered to be endemic, has led to the laboratory confirmation of disease with subsequent reporting of cases to scientific press.

From a historical perspective, early reports of the disease were predominantly associated with mortalities in small ruminant populations across West Africa. During the 1980's the detection of the virus in the Sultanate of Oman on the Arabian Peninsula led to the recognition of the virus in India (Taylor, 1984; Shaila et al., 1989) and the subsequent reporting of the disease across much of Asia. During the last few years the virus has extended its geographical range across vast swathes of Asia (Wang et al., 2009; Parida et al., 2015), in particular following an outbreak that resulted in the detection of the virus across much of China during late 2013 and 2014 (Banyard et al., 2014).

The advent of molecular technologies over the last 20 years has led to studies into the genetic composition of different isolates of PPRV which has fostered a greater understanding of the molecular epidemiology of the disease in endemic regions. From a genetic perspective, four distinct lineages of PPRV have been described based on genetic data derived from either the nucleoprotein gene or the fusion protein gene of the virus (Couacy-Hymann et al., 2002; Forsyth and Barrett, 1995). With increased genetic surveillance it has become clear that for closely related strains of PPRV the N gene is more divergent and therefore represents a more suitable locus for the genetic resolution of isolates (Kwiatek et al., 2007; Kerur et al., 2008; Kumar et al., 2014) although several countries still perform F gene based PCRs (Banyard et al., 2010). Furthermore, the rapid development of sequencing technologies has revolutionised sequence generation from samples with the result that full genome data can be derived from samples with relative ease enabling phylogeneticists to assess isolates based on any part of the genome. Most recently, the first Bayesian full genome assessment of PPRV has, based on full genome sequence data available, concluded that from an evolutionary perspective, emerged at the beginning of the twentieth century, a few decades before the first recorded description of PPRV in 1942 (Muniraju et al., 2014). A Bayesian phylogenetic analysis of all PPRV lineages mapped Lineage III PPRV as the first to have diverged from an ancestral virus. The estimated probability for the root location of an ancestral PPRV and individual lineages were determined as being Nigeria for PPRV as a whole, Senegal for lineage I, Nigeria/Ghana for lineage II, Sudan for lineage III, and India for lineage IV. In recent years PPRV had extended its boundaries southwards in Africa as far as southern Tanzania (2008) and Zambia (2015) and the Democratic Republic of Congo and Angola (2012) (Banyard et al., 2010; FAO, 2013). PPR outbreaks have also been reported across North Africa including within Tunisia (2006), Morocco (2008) and Algeria (2011). Alongside this, within Europe, Turkey reported approximately twenty laboratory confirmed PPR outbreaks in sheep and goats during 2011-2012 (Sevik and Sait, 2015). In East Asia, the virus spread to Tibet (2007) and has recently been reported across much of China (2013-2014) (Banyard et al., 2014).

The reporting of different genetic lineages in different regions has been sporadic as often facilities have not had the technological capability to genetically type virus. The accessibility of molecular tools and more recently the ability to generate full genome data with relative ease has enhanced the full genome dataset available for PPRV.

The hypothesis that dominant lineages may enter areas and overwhelm existing lineages of the virus is of great interest. Certainly the extensive detection of lineage IV virus in Africa in recent years has led some to hypothesise that lineage IV is a dominant lineage that is replacing other lineages across different areas. Historically, the detection of lineage IV PPRV in Central Africa was the first detection of this lineage on the African continent. Since then, and following the increased interest in the disease, this lineage has continued to be detected in other regions across Africa. Whilst reports of PPRV infection are frequently reported in the scientific literature and to the OIE, genetic characterisation is required to understand the epidemiology of the virus. The expansion of lineage IV virus across Africa has been a slow process when considering reports of genetic characterisation of this lineage in different areas. A single, report of lineage III in India exists but little evidence exists to validate this occurrence from the perspective of the existence of this lineage within India (Dhar et al., 2002). The occasional reports of lineage III existence on the tip

of the Arabian Peninsula have to the speculation that lineage IV strains of PPRV quickly become dominant within regions at the expense of other lineages. Whether local extinction of other lineages occurs is unclear as reports where genetic data exist for local outbreaks are few. The reported expansion of lineage IV virus across Central and East Africa and the explosion of lineage IV virus between 2008 and 2012 across North Africa have further fuelled ideas surrounding the enhanced pathogenicity of lineage IV strains of the virus (Kwiatek et al., 2011). A recent study in Nigeria has demonstrated an extensive distribution of lineage IV virus alongside a fairly limited distribution of the lineage II virus that has been documented historically (Woma et al., 2015). Whether this increased representation of lineage IV virus in different regions indicates a dominance of viruses from this lineage is unclear. Certainly, as with other morbilliviruses (Banyard et al., 2005; Baron et al., 2005) the determinants of virulence for PPRV remain almost completely undefined and as such potential lineage driven dominance remains speculative, especially as many regions are unable to genetically type circulating viruses. Further, where controlled experimentation with different lineages of PPRV have been carried out there is little to suggest significant differences in pathogenicity, rather host variability in immune status and coinfection with other parasites drive the overall outcomes of infection. Alongside this recent reports of lineages I, II and IV in Uganda (Luka et al., 2012) and Tanzania (Mahapatra et al., 2015), lineage II and IV in Nigeria (Woma et al., 2015) and lineage III in the Sudan suggest at least that other lineages of virus remain present in the region despite the emergence of lineage IV (Banyard et al., 2010). As the application of molecular tools to investigate the presence of different lineages of PPR expands across endemic areas, the existence and potential expansion of different lineages may become more apparent. However, without further evidence of either the host factors associated with the outcome of infection or identification of viral genetic elements that influence pathogenicity, a clear understanding of the epidemiology of individual lineages, their pathogenicity and their existence will not be possible.

From an epidemiological perspective it is clear that small ruminants play the most important role in spread of the disease. A role for different wildlife species has not yet been well defined although numerous studies have implicated PPRV in disease in different species (Mahapatra et al., 2015). Of these camels have, on occasion, been profoundly affected by PPRV infection with high mortalities being seen (Roger et al., 2001; Khalafalla et al., 2010; Saeed et al., 2010; Kwiatek et al., 2011). The potential for subclinical infection of camelids and transmission to in contact ruminants has not yet been fully explored although experimental infection has demonstrated the potential for mild disease in these animals (El Hakim, 2006). Alongside camels, although long thought of as insignificant factors in the spread of PPRV (Anderson and McKay, 1994), several studies have implicated a potential role for large ruminants in the epidemiology of PPRV with reports detailing serological evidence of infection (Govindarajan et al., 1997; Sen et al., 2010; Lembo et al., 2013). Furthermore, some have speculated on the utility of serological surveillance of large ruminants to determine the circulation of wildtype virus in the face of small animal vaccination (Abubakar et al., 2015). However, further studies are required to assess any potential role of large ruminants in the epidemiology of PPRV.

With the successful eradication of rinderpest virus the World Animal Health Organisation (OIE) has targeted PPRV for global eradication by 2030. Current vaccines are sufficient to enable a successful eradication programme (Albina et al., 2013). However, there is still room for improvement of the existing vaccines not least in the areas of thermostability and enabling differentiation between naturally infected and vaccinated animals- the so called DIVA principle. The former is of great importance when delivering vaccines to animal populations in tropical areas. Currently, the maintenance of a cold chain is costly and frequently unrealistic in some endemic areas often leading to a reduction in the quality of product administered in vaccination campaigns. Several laboratories are trying to overcome these issues (Sen et al., 2010; Silva et al., 2011) although currently a thermostable vaccine preparation is not commercially available for use against PPRV in the field. The issue of DIVA is more complicated. Whilst not essential, lessons learnt from the rinderpest eradication programme demonstrated clearly that towards the end of an eradication program, where vaccination ceases and a period of serosurveillance is initiated, the ability to differentiate serologically between vaccinated and naturally infected animals would aid serological assessment and the potential detection of antibodies against virus circulating in a silent, subclinical form. Some authors have

recently highlighted the utility of serological surveillance of in contact large ruminant populations for a similar reason. As large ruminants do not generate disease symptoms, and would not be vaccinated in any eradication programme, signs of serological positivity in cattle and buffalo would indicate naturally circulating virus (Abubakar et al., 2015). However, this may be applicable in some farm environments whilst it will not be sufficient in all areas. To this end DIVA formulations are required for the latter stages of the eradication campaign and several approaches are currently being taken. Most recently approaches including epitope deletion (Buczkowski et al., 2012; Muniraju et al., 2015), subunit vaccines (Herbert et al., 2014) and the insertion of positive marker genes (Hu et al., 2012; Muniraju et al., 2015). Several of these have shown promise and have been reported towards the end of an eradication programme as deemed necessary. The pathway to eradication will not be easy but with the date set for 2030 it will take cooperation on an international scale to repeat the successes seen with eradication of rinderpest virus and remove the burden of this plague of small ruminants globally.

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