ORIGINAL RESEARCH ARTICLE

Seroprevalence of Rift Valley fever virus in sheep and goats in Zambézia, Mozambique

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Background: The Rift Valley fever virus (RVFV) is a vector-borne virus that causes disease in ruminants, but it can also infect humans. In humans, the infection can be asymptomatic but can also lead to illness, ranging from a mild disease with fever, headache and muscle pain to a severe disease with encephalitis and haemorrhagic fever. In rare cases, death can occur. In infected animals, influenza-like symptoms can occur, and abortion and mortality in young animals are indicative of RVFV infection. Since the initial outbreak in Kenya in the 1930s, the virus has become endemic to most of sub-Saharan Africa. In 2000, the virus appeared in Yemen and Saudi Arabia; this was the first outbreak of RVF outside of Africa. Rift Valley fever epidemics are often connected to heavy rainfall, leading to an increased vector population and spread of the virus to animals and/or humans. However, the virus needs to be maintained during the inter-epidemic periods. In this study, we investigated the circulation of RVFV in small ruminants (goats and sheep) in Zambézia, Mozambique, an area with a close vector/wildlife/livestock/human interface.

Materials and methods: Between September and October 2013, 181 sheep and 187 goat blood samples were collected from eight localities in the central region of Zambézia, Mozambique. The samples were analysed for the presence of antibodies against RVFV using a commercial competitive ELISA.

Results and discussion: The overall seroprevalence was higher in sheep (44.2%) than goats (25.1%); however, there was a high variation in seroprevalence between different localities. The data indicate an increased seroprevalence for sheep compared to 2010, when a similar study was conducted in this region and in overlapping villages. No noticeable health problems in the herds were reported.

Conclusions: This study shows an inter-epidemic circulation of RVFV in small ruminants in Zambézia, Mozambique. Neither outbreaks of RVF nor typical clinical signs of RVFV have been reported in the investigated herds, indicating subclinical infection.

Keywords: *Rift Valley fever virus*; *Mozambique*; *Zambézia*; *seroprevalence*; *goats*; *sheep*

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Rift Valley fever virus (RVFV) is a mosquito-borne virus that can infect domestic ruminants, such as cattle, sheep and goats. The outcome varies from influenza-like symptoms to abortion and mortality in young animals. Susceptibility to RVFV infection has been suggested to depend on factors such as age and species. The virus can also infect humans, and the outcome varies from asymptomatic to fever, encephalitis and haemorrhagic fever. In addition to health implications for animals and humans, the infection also has severe economic impact through animal loss and/or trade restrictions (1–4).

RVFV was first reported in Kenya in the 1930s (5) and is now considered endemic in most parts of sub-Saharan Africa. In 2000, there was an outbreak of RVF in Yemen and Saudi Arabia, which was the first time the disease was observed in a country outside of Africa (6). How the

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virus was introduced is not known, but it is believed to have been through transportation of infected livestock (7). The first report of RVF in Mozambique was in the 1960s (8). Since then, reports of the virus in the country have been scarce. A sero-epidemiological survey of pregnant women in eight provinces of Mozambique from 1981 to 1983 showed a seroprevalence of 2% (9). In 2010–2011 in Maputo, the capital of Mozambique, the RVFV seroprevalence was 36.9% in cattle (10). Analysis of goat and sheep serum samples collected in 2007 and 2010 in the Zambézia Province in Mozambique showed a seroprevalence of IgG antibodies of 35.8% in sheep and 21.2% in goats in 2007 compared to 9.2% in sheep and 11.6% in goats in 2010 (11).

The maintenance of RVFV between epidemics is not completely understood. However, the general view is that the virus is maintained through vertical transmission in its arthropod vector. Wildlife is also considered as a possible reservoir for RVFV (12). A serological study in Kenya found RVFV antibodies in black rhino, buffalo, elephant, warthog and wildebeest, further supporting this notion (13); however, the data in wildlife are scarce and more studies are needed to fully understand the possible role of wildlife in RVFV transmission. Epidemics are connected to an increase in vector population, often following heavy rain.

The abundance and diversity of wildlife and mosquito species, as well as a close wildlife/human/livestock/vector interface, makes Zambézia Province ideal for RVFV circulation. In this study, we analyse the seroprevalence of RVFV in sheep and goat samples collected in 2013 in Zambézia Province to further investigate the inter-epidemic circulation of RVFV in the region.

Materials and methods

Study area and sampling
The study was conducted in Zambézia Province (Fig. 1) from September to October 2013. In the province, eight localities in three districts (Mopeia, Nicoadala and Quelimane) were selected. The study area was selected because, although most bovines in Zambézia Province are vaccinated, no vaccination of small ruminants had been performed and the local veterinary service had identified this province as a potential high-risk area for RVFV. The farms were selected based on willingness of farmers and overlap with the regions investigated by Fafetine et al. (11). Blood samples were collected from goats ($n = 187$) and sheep ($n = 181$). Both female and male animals were sampled, and the animals were categorised into three age groups (0–6 months, 6–12 months and >12 months), see Table 1 for further details. Animals were bled from the jugular vein using a vacutainer. Serum was separated from the whole blood and was stored at +4°C until tested. The
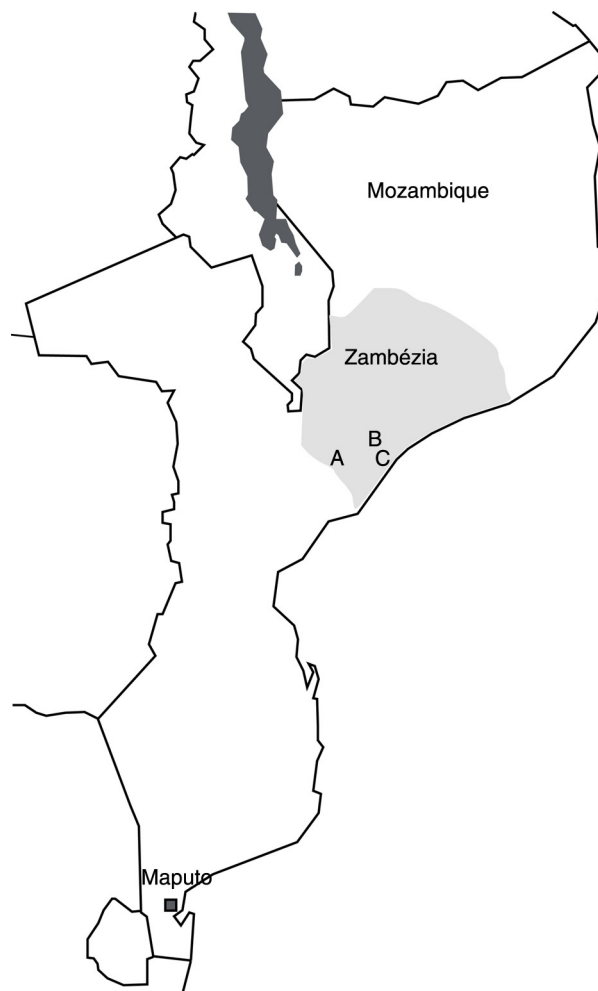

Fig. 1. A map of Mozambique, highlighting Zambézia Province and displaying the three areas (Mopeia (a), Nicoadala (b) and Quelimane (c)) where the sampling has occurred.

animal keeper was interviewed about the animal health and mortality on the farm.

Serology
The 368 serum samples were analysed using the ID Screen Rift Valley Fever Competition Multi-species ELISA (ID-Vet) according to the manufacturer's instruction. Optical reading was performed at 450 nm, and the cut-off values suggested by the manufacturer were followed. The seroprevalence was calculated, with a 95% confidence interval, using the modified Wald method (14). The seroprevalence was compared between species, gender and age using a chi-square test.

Results
In total, 368 ruminants (187 goat and 181 sheep) were sampled in the three districts (Table 1). Most of the samples were from adult animals (>12 months) but animals <12 months were also sampled. Antibodies specific for RVFV were detected in animals from all localities

Table 1. Details of the seroprevalence of RVFV in goat and sheep populations sampled in 2013 in Zambézia, Mozambique

			No. sampled	No. positive	Seroprevalence (%)	95% CI
Goats	All		187	47	25.1	19.4–31.8
	Sex	Female	135	33	24.4	17.9–32.4
		Male	52	14	26.9	16.7–40.4
	Age	0–6 months	29	6	20.7	9.5–38.8
		6–12 months	55	4	7.3	2.4–17.8
		>12 months	103	37	35.9	27.3–45.6
Sheep	All		181	80	44.2	37.2–51.5
	Sex	Female	123	60	48.8	40.1–57.7
		Male	58	20	34.5	23.5–47.4
	Age	0–6 months	31	5	16.1	6.7–33.1
		6–12 months	46	11	23.9	13.8–38.1
		>12 months	104	64	61.5	51.1–70.3

For each species, the overall data have been grouped according to the sex and age of the sampled animals.

included in this study (Table 2). One hundred twenty-seven of the 368 analysed serum samples (37.5%; 95% CI 29.8–39.5) were positive for RVFV antibodies. The overall seroprevalence in sheep was significantly higher than in goats ($p < 0.001$), with 44.2 and 25.1% seropositive animals, respectively (Table 1). No statistically significant difference in seroprevalence was observed between gender for sheep or goats. However, adult animals (> 12 months) had a higher seroprevalence than younger animals ($p < 0.001$). The seroprevalence for both goat and sheep varied between districts and between individual localities (excluding places with < 30 animals sampled), with a seroprevalence range of 7.5–40.6% for goats and 19.7–90.9% for sheep.

In connection to the sampling activities, the animal keepers were asked about animal health and mortality on their farm. All the farms had experienced death of animals during the sampling year; however, the deaths did not appear to be associated with typical RVF symptoms. No malformed foetuses were reported on any of the farms. Three farms reported abortions, but these were most likely linked to starvation.

Discussion

In this study, we show that RVFV is circulating during inter-epidemic periods in the Zambézia Province in both goats and sheep. The overall seroprevalence in 2013 was 44.2 and 25.1% for sheep and goat, respectively. The difference in seroprevalence between sheep and goat was significant ($p < 0.001$). Why the seroprevalence is different in goat and sheep, sometimes even on the same farm, is not known; however, it could be that one species is more susceptible than the other, which would be supported by the fact that in this study we consistently found a higher seroprevalence in sheep than goats. The possible difference in susceptibility could be due to a number of unidentified factors, such as host genetic background and differences in animal management, but more studies are needed to elucidate this.

The highest seroprevalence for both goat and sheep was observed in adult animals (> 12 months). The higher seroprevalence in adult animals was expected as they have been exposed to the risk of infection for a longer period. For goats, the youngest animals (0–6 months) had a higher seroprevalence (20.7%) than goats between 6 and

Table 2. The seroprevalence of RVFV in goat and sheep sampled in different localities in the three districts – Mopeia, Nicoadala and Quelimane

District (Localities)	No. goat	Seroprevalence (%)	95% CI	No. sheep	Seroprevalence (%)	95% CI
Mopeia (Deda)	5	60.0	22.9–88.4	–	–	–
Mopeia (Chimuara)	47	21.3	11.8–35.0	42	40.5	27.0–55.5
Mopeia (South)	–	–	–	33	90.9	75.7–97.6
Nicoadala (Mucelo)	1	0.0	0.0–83.3	66	19.7	11.8–31.0
Nicoadala (Amed)	40	7.5	1.9–20.6	8	37.5	13.5–69.6
Nicoadala (Mingano)	33	26.5	12.6–41.3	–	–	–
Quelimane (Dona ana)	32	40.6	25.5–57.8	–	–	–
Quelimane (Padeiro)	30	30.0	16.5–48.0	32	53.1	36.5–69.1

12 months of age (7.3%). The reason for this could be that maternal immunity can last up to 6 months of age.

A previous study by Fafetine et al. (11) investigated serum samples collected in 2007 and 2010 from sheep and goats in the same regions as those included in this study. Together, the data show that the overall seroprevalence in Zambézia Province varies between years. In 2007, the seroprevalence in goat and sheep, respectively, was 21.2 and 35.8%; in 2010, it was 11.6 and 9.2%; and in 2013, it was 25.1 and 44.2%. There are several potential reasons for the differences in seroprevalence. One reason could be precipitation because high rainfall is an important risk factor of RVF. In 2013, the average monthly precipitation from January to April was 333 mm, compared to 114 mm in 2010 and 210 mm in 2007 (Instituto Nacional de Metreologica, Maputo Mozambique, 2013). Increased rain could lead to a rise in seroprevalence through an increased vector population. Noteworthy, Fafetine et al. (11) used an ELISA detecting only IgG antibodies, whereas, in this study, we used an ELISA detecting both IgG and IgM antibodies. This difference could lead to slightly higher prevalence rates in this study. Also, although the same region in Zambézia was studied in both studies, it was not always possible to sample exactly the same farms. The seropositivity in livestock during inter-epidemic periods has been investigated in other counties. Sumaye's et al. (15) showed that in the Kilombero River Valley, Tanzania, the overall prevalence in livestock (cattle, sheep and goat) born after the RVFV outbreak in 2006/07 (15) was 5.55%, much lower than observed in this study.

The seroprevalence varied between the sampled farms, even within the same district. The reason for this is not known, but factors such as closeness to fresh water or 'dambos' during the dry period, the presence of containers or tires on the farm that could serve as mosquito breeding grounds, possible effects of vaccination of cattle in the area for transmission of RVFV to small ruminants, the size of the herds, closeness to wild fauna and possible wildlife reservoirs could play a role in the transmission of RVFV.

In summary, the high level of RVFV seroprevalence in small ruminants in Zambézia suggests viral circulation, even without reports of clinical signs in susceptible hosts. Therefore, it is important to monitor the RVFV circulation in Mozambique for the early detection of epidemic occurrence of disease and prevention of transmission to humans.

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