An investigation into the source and spread of foot and mouth disease virus from a wildlife conservancy in Zimbabwe


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Summary
African buffalo were introduced into a wildlife conservancy in the southeast of Zimbabwe in an effort to increase the conservancy’s economic viability, which is primarily based on eco-tourism. The buffalo were infected with SAT serotypes (SAT-1, SAT-2 and SAT-3) of foot and mouth disease (FMD) virus, and in order to isolate the conservancy and prevent the transmission of FMD to adjacent populations of domestic livestock, the conservancy was surrounded by a double-fence system, 1.8 m in height. The intention was to prevent the movement of both wildlife and domestic animals across the perimeter. However, two years after the buffalo were introduced, FMD occurred in cattle farmed just outside of the conservancy. Using serological and molecular diagnostic tests, epidemiological investigations showed that it was most likely that antelope (impala or kudu), infected through contact with the buffalo herd within the conservancy, had jumped over the fence and transmitted the virus to the cattle.

Keywords

Introduction
In the early 1990s several wildlife conservancies, the largest of which is 340,000 ha, were formed through the amalgamation of groups of traditional cattle ranches in the lowveld (low altitude) areas of Zimbabwe (Fig. 1). These conservancies were established because it had been demonstrated that a greater return per hectare could be derived from purely wildlife-based enterprises, compared to cattle ranching, in this semi-arid part of the country (11). The conservancies were each surrounded by twelve-strand game-fencing standing 1.8 m high. Within the conservancies all internal fencing was removed to allow the wildlife complete freedom of movement.

The presence of African buffalo greatly enhances the financial return from wildlife-based enterprises because they are one of the ‘big five’ wildlife species, i.e. wildlife with high eco-tourist potential (elephant, lion, rhinoceros, buffalo and leopard). The owners of the conservancies were, therefore, anxious to obtain buffalo despite the fact that they may be carriers of foot and mouth disease (FMD)
The majority of conservancies were situated within Zimbabwe’s FMD control zone (the buffer zone and surveillance zone) and not within the (FMD free zone) beef export zone (Fig. 1). Following an agreement made by all sectors that would potentially be affected if FMD were introduced, the conservancies were granted permission to introduce wild buffalo, some of which were persistently infected with FMD viruses. A number of provisos were attached to this permission, as follows:

– all cloven-hoofed livestock were to be removed from the conservancies
– buffalo-proof fences (six strands of high tensile steel or barbed wire, 1.2 m in height) were to be constructed inside the perimeter fence and separated from the 1.8 m (12 strand) perimeter fence by a defoliated strip at least 7.5 m wide. Both the perimeter and inner ‘buffalo’ fences were to be electrified using solar power
– fence lines were to be inspected daily for breaks and the passage of any animal across them was to be recorded.

In 1993 buffalo were introduced into the conservancies from the Gona-re-zhou National Park in the southeast of the country. Additional buffalo were introduced in 1994 from the Hwange National Park in western Zimbabwe. Genomic sequencing of a portion of the ID gene, isolated from SAT-serotype FMD viruses prevalent in buffalo from the two national parks, has shown that the viruses (serotypes SAT-1, SAT-2 and SAT-3) isolated from these two locations belong to different evolutionary lineages (3, 4, 5). For the purposes of this paper SAT-2 viruses that are restricted to specific geographical locations are referred to as totopotypes.

The rationale behind the decision by the Department of Veterinary Services in Zimbabwe to allow the introduction of buffalo harbouring FMD viruses into the conservancies was based on a number of factors. Most importantly, past experience and experimental work on the transmission of FMD from carrier buffalo to cattle showed that direct and prolonged contact between the buffalo and cattle is necessary for this to occur (6, 7, 20). Furthermore, the circumstances that favour airborne transmission of FMD virus over long distances, which has infrequently been observed in outbreaks of the disease in Europe (8), do not usually exist in southern Africa (16, 17). The consensus within the Department of Veterinary Services was that if direct contact between buffalo and cattle could be prevented, FMD would not occur in livestock outside the conservancies. However, to quantify the risk to domestic livestock posed by the introduction of buffalo to the conservancies, the Department commissioned a risk assessment that was conducted by two independent consultants (funded by the British Department for International Development). The risk assessment showed
that the likelihood of outbreaks in domestic cattle outside of the conservancies, as a result of buffalo escaping from the conservancies or excreting wind-borne virus, was low. However, the study identified antelopes from the conservancies as an indirect risk factor in the transmission of FMD, because buffalo could infect antelopes that would subsequently jump over the fence lines and transmit the virus to domestic livestock (15).

In July 1997 an outbreak of FMD, caused by a SAT-2 virus, occurred in a herd of 700 cattle immediately adjacent to the perimeter fence of the largest of the conservancies (Fig. 1). This outbreak was successfully eliminated through the immunisation of the infected herd and any cattle potentially in contact with the herd, and the implementation of conventional zoosanitary measures.

This paper describes an investigation into the source of the virus responsible for the aforementioned outbreak of FMD. Recommendations to prevent similar outbreaks from occurring in the future have also been included.

Methods

Diagnosis of the cattle outbreak

Samples of epithelium were collected from tongue lesions of clinically affected cattle immediately after the disease was first suspected in the herd. The samples were packaged on dry ice and shipped via air to the World Reference Laboratory (WRL) for FMD at the Institute for Animal Health in Pirbright in the United Kingdom. In the laboratory the virus was cultured on primary calf thyroid cells and typed by enzyme-linked immunosorbent assay (ELISA) (13).

Identification of foot and mouth disease viruses in buffalo within the conservancy

Two buffalo herds, known as the Mukazi and Mkwazi herds, which commonly grazed in the area of the conservancy located the closest to where the outbreak in the cattle occurred (Fig. 1), were driven (using helicopters) into a temporary ‘boma’ (pen) constructed of plastic sheeting. Nine or ten animals from each herd were chemically immobilised and scrapings from the pharynx and anterior oesophagus were collected using a probang cup. The contents of the cup were dispensed into 5 ml of 0.08 M phosphate buffer, pH 7.2, and maintained at 4°C until completion of the sampling procedure. Each sample was then placed in a canister containing liquid nitrogen, packaged on dry ice, and shipped by air to the WRL. At the WRL the samples were cultured on thyroid cells and the viral isolates were identified using an ELISA.

The probang cup was washed with a strong solution of citric acid and rinsed in three separate buckets of clean water prior to being used in each animal. Blood for serum preparation was also collected from the immobilised buffalo.

Sampling of antelope

Impala (Aepyceros melampus) and kudu (Tragelaphus strepsiceros) were captured from three sites located within the general area where the Mkwazi buffalo herd grazed. Two of the sites (site A and B, Fig. 1) were within 2 km of the perimeter fence and the cattle paddock. Samples were collected from a total of 17 kudu and 23 impala at site A and 27 impala at site B. An additional 35 impala and two kudu were sampled at the site where the Mkwazi herd was captured (site C, Fig. 1). Blood for serum preparation was collected from all of the antelope and throat scrapings were taken from impala and kudu at site A. In addition, serum was collected from 149 culled animals; 141 impala, 6 kudu and 2 eland (Taurotragus oryx) that had been living in parts of the conservancy where other buffalo herds were present. Probang specimens were screened for FMD virus by the inoculation of primary pig kidney cells at the Agricultural Research Council-Onderstepoort Veterinary Institute (ARC-OVI), South Africa.

Serology

Antibody levels in sera to SAT-1, SAT-2 or SAT-3 viruses were determined with a competitive ELISA (14) at the Central Veterinary Laboratory in Harare.

Nucleotide sequencing and phylogenetic analysis

Genomic amplifications and nucleotide sequencing of the viruses were performed at the WRL and the ARC-OVI using the ID gene polymerase chain reaction and sequencing analysis approach described elsewhere (2).

Results

On site investigation

As previously mentioned, it was established that there were two buffalo herds, each consisting of approximately 50 animals, which frequently grazed within 2 km to 20 km of the perimeter fence adjacent to where the FMD outbreak in the cattle occurred. The Mkwazi herd, introduced from the Gona-re-zhou National Park in 1993, typically grazed the closest to the cattle outbreak focus (2 km to 10 km), while the Mukazi herd, obtained from Hwange National Park in
Viral recovery from clinically affected cattle and buffalo oesophageo-pharyngeal (probang) samples

The four SAT-2 viruses identified in samples from the cattle involved in the FMD outbreak were shown to be closely related to one another and to belong to the northwest Zimbabwe topotype, which includes virus strains isolated from the Hwange National Park (Fig. 2).

Two closely related SAT-2 viruses were recovered from the ten probang samples obtained from the Mukazi buffalo herd and one SAT-1 virus from the nine probang samples obtained from the Mkwazi herd. No viruses were recovered from clinically affected cattle in 1994, grazed slightly further away (15 km to 20 km). The fence guards in the area reported that no fence breaks had occurred and that there had been no movement of buffalo out of the conservancy or of cattle into it. However, approximately two weeks before the FMD outbreak in cattle occurred, a pride of lions had moved into the area and caused considerable disturbance among the buffalo herds inside the conservancy and the cattle on the outside (the lions were able to cross the electrified fences). Because of the cattle losses caused by the lions the owners moved the cattle, mostly adult cows, away from the perimeter fence. Clinical cases of FMD in the cattle were observed eight days later. The cattle had not been vaccinated against FMD.

* Indicates those viruses sequenced by the World Reference Laboratory for foot and mouth disease at the Institute for Animal Health in Pirbright in the United Kingdom. The scale is equivalent to a nucleotide distance of 2%

Fig. 2
Neighbour-joining tree depicting VP1 gene relationships of SAT-2 serotype foot and mouth disease viruses from buffalo and cattle in Zimbabwe

Bootstrap values based on 10,000 replications and > 60 are indicated next to the relevant nodes.
from the probang specimens collected from the antelope living within the conservancy. The SAT-2 viruses isolated from the Mukazi buffalo were grouped within the Hwange viral cluster (Fig. 2) and were determined to be closely related to the viruses isolated from the cattle.

Serology

The proportions of buffalo in the Mukazi herd that were serologically positive for SAT-1, SAT-2 and SAT-3 viruses were 30%, 70% and 40% respectively, while in the Mkwazi herd the percentages were 44, 44 and 56, respectively. Antibody titres to SAT-2 were higher in the Mukazi than in the Mkwazi herd.

Most of the ELISA tests performed on sera collected from antelope living within the conservancy were negative, except for site A where 17% of the impala and 23% of the kudu had significant levels of antibody to SAT-2 virus. Only one animal among the antelope tested from other parts of the conservancy (i.e. antelope located in areas other than Sites A, B and C where buffalo herds were present) was seropositive for SAT-2.

Discussion

This investigation demonstrated the importance of molecular epidemiological data in establishing the origin of FMD outbreaks. The results showed, surprisingly, that the virus responsible for the outbreak in cattle in the southeast of Zimbabwe originated in buffalo introduced into the conservancy, more than two years previously, from the Hwange National Park (located several hundreds of kilometres to the northwest). Hence, a ‘foreign’ viral topotype was introduced to southeast Zimbabwe by translocation of buffalo from the Hwange National Park. The appearance of a virus type that was not previously present in this area has implications for the effective control of the disease by vaccination (19). Since there is no evidence that the buffalo or the cattle crossed the perimeter fence of the conservancy, it was suspected that an intermediary was responsible for the transfer of the virus, based on what is currently known about the transmission of FMD, this could only have been air currents or another species of animal.

The double fence concept developed in Zimbabwe to provide a physical barrier between wildlife conservancies and domestic livestock was based on the premise that aerosol transmission of FMD viruses over distances of more than a few metres does not occur in southern Africa. On the rare occasions that long distance aerosol transmission has been shown to occur in Europe, large piggeries were usually the source of infection (A.I. Donaldson, personal communication), presumably because of the large viral load that is excreted by pigs, i.e. pigs excrete up to 3,000 times more virus than cattle (8). Southeastern Zimbabwe farms a small number of pigs and has no large piggeries. Furthermore, for aerosols to survive in air currents for any significant length of time the relative humidity must be at least 60% (8), a condition that is uncommon in this relatively dry area (except for in the early morning hours, particularly in the winter). The outbreak in question did occur during the winter, however, it has also been shown that although experimentally infected buffalo in the acute stage of infection with SAT virus excrete virus in expired aerosols, the amounts are too low to measure (9). The risk assessment that was conducted a few weeks prior to the outbreak examined the likelihood of airborne viral transmission but concluded that the risk was considerably lower compared to the other three scenarios that were considered (i.e. cattle entering the conservancy, infected wildlife exiting the conservancy, and infected wildlife products exiting the conservancy) (15). Based on the above observations, it is concluded that although airborne transmission of SAT-2 virus from buffalo to cattle could have precipitated the SAT-2 outbreak in July 1997, the probability is low.

The risk assessment conducted prior to the FMD outbreak in question revealed that antelope were occasionally able to jump across the double fence-line. Calculations based on this observation and antibody prevalence rates in impala revealed that this represented a significant risk in the transmission of FMD (15). Unfortunately, the results of the risk assessment were in the process of being analysed when the FMD outbreak occurred and there had been insufficient time for the Zimbabwean authorities to implement risk management strategies to address this threat. Following the outbreak of July 1997, a special investigation was conducted to assess the probability of antelope having been responsible. On the basis of serological data, it was shown that a proportion of both the impala and kudu in close proximity to the Mukazi buffalo herd, but not elsewhere in the conservancy, had been infected with SAT-2 virus. However, the serological data did not enable the authors to determine when infection of the antelope occurred.

An additional factor to be considered is the disturbance created by the presence of lions in the area of the conservancy adjacent to the infected cattle. It is very probable that this caused an increase in the movement of the antelope, especially the kudu, through or over the 1.8 m high perimeter fence. Infected antelope that had escaped from the conservancy would have been a source of infection to animals on the outside of the perimeter fence. As this outbreak occurred during the dry season, it was also more likely that the escaped antelope would have used the same water points as cattle. In the rainy season a greater availability of water would reduce this occurrence.
A number of studies on FMD in impala have shown that, unlike cattle and African buffalo, these animals do not become long-term carriers of FMD viruses (1, 9, 18; Van Vuuren et al., unpublished data). However, outbreaks of FMD in impala occur regularly in the Kruger National Park (KNP) in South Africa and there is historical evidence that impala may have been responsible for the transmission of FMD to cattle living in areas close to the KNP (12, 18). The sero-prevalence rate among impala at site A (17%) was similar to that found during an outbreak in this species in the KNP in 1992 (12). Less is known about the role of kudu in the epidemiology of FMD in southern Africa, but natural infection of kudu has been observed, and short-term carriers have been detected following experimental infection (10). Although serological evidence of infection among antelope in the vicinity of the FMD outbreak is not conclusive proof that the antelope were the source of the infection for the cattle, the circumstantial evidence is considerable.

It has been suggested that a wide cattle-free zone should be created outside wildlife areas containing FMD infected buffalo to preclude the possibility of FMD transmission to domestic livestock. This is often not a practical proposition in southern Africa where land availability has dramatically decreased and where there is a growing necessity to integrate and rationalise land use. There is intensified competition for land due to rapidly increasing numbers of people and the concomitant rise in poverty levels, and traditional inhabitants want access to the grazing areas and firewood that are available in wildlife reserves. At the same time the importance of eco-tourism for the economies of southern African countries is growing and there is a need to provide space for these enterprises to develop.

It is the opinion of the authors, based on this investigation, and that of Sutmoller et al. (15) that the risk of FMD being transmitted from animals inside of conservancies to adjacent domestic livestock can be reduced to acceptable levels. This can be achieved by increasing the height and strength of the perimeter fencing surrounding conservancies so as to render them impervious to antelope and buffalo. Additional measures, such as limiting the population densities of buffalo and antelope and vaccinating livestock grazing immediately outside of conservancies containing buffalo, may also reduce the risk of transmission. Following this incident the height of fencing surrounding conservancies in Zimbabwe was increased to a minimum of 2.3 m.

Enquête sur l’origine et la propagation de la fièvre aphteuse à partir d’une réserve naturelle au Zimbabwe


Résumé
Des buffles africains ont été introduits dans une réserve naturelle du sud-est du Zimbabwe en vue d’en améliorer la viabilité économique tributaire de l’écotourisme. Or, ces buffles étaient infectés par les sérotypes SAT (SAT 1, SAT 2 et SAT 3) du virus de la fièvre aphteuse. Une double clôture de 1,8 m de haut a été érigée autour de la réserve afin de l’isoler et prévenir la transmission de la maladie aux élevages proches. Ce dispositif devait éviter le franchissement du périmètre, que ce soit par les animaux sauvages ou les animaux domestiques. Cependant, deux années après l’introduction des buffles, un foyer de fièvre aphteuse était signalé dans un élevage bovin situé dans le voisinage immédiat de la réserve. Les enquêtes épidémiologiques réalisées à l’aide d’épreuves de diagnostic sérologiques et moléculaires ont montré que des antilopes (impalas ou koudous), infectées au contact du troupeau de buffles à l’intérieur de la réserve, avaient très probablement sauté par-dessus la clôture et transmis le virus au bétail.

Mots-clés
Investigación del origen y la propagación del virus de la fiebre aftosa a partir de una reserva de fauna salvaje de Zimbabue


Resumen
En una reserva del sureste de Zimbabue se introdujeron búfalos africanos entre los cuales se podían encontrar ejemplares infectados con serotipos SAT (SAT-1, SAT-2 y SAT-3) del virus de la fiebre aftosa. El objetivo era de acrecentar la viabilidad económica de la zona, que depende esencialmente del ecoturismo. A fin de aislar ese espacio e impedir la transmisión de la enfermedad a las poblaciones vecinas de ganado doméstico, se instaló un sistema de doble cercado de 1,8 metros de altura alrededor de la reserva. La idea consistía en evitar que los animales, tanto salvajes como domésticos, atravesaran el perímetro. Sin embargo, dos años después de la llegada de los búfalos se dieron casos de fiebre aftosa entre el ganado de explotaciones contiguas a la reserva. Mediante pruebas de diagnóstico serológica y moleculares, la investigación epidemiológica puso de manifiesto que lo más probable es que el contagio del ganado se debiera a antílopes (impalas o cudúes) que hubieran saltado por encima de las vallas tras quedar infectados por contacto con la manada de búfalos de la reserva.

Palabras clave

References


