Article One

Epidemiological Investigation of an Anthrax Outbreak at the Wildlife-Human Interface in Mosi-oa-Tunya National Park, Zambia, October-November 2025

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Abstract

Introduction: Anthrax is an acute zoonotic disease caused by Bacillus anthracis, primarily affecting herbivores, but also carnivores and humans through contaminated meat or environments. On 30 October 2025, a dead hippopotamus from Mosi-oa-Tunya National Park was fed to 36 captive lions, causing morbidities and mortalities, prompting an outbreak investigation. The objectives were to confirm the outbreak, assess the timeliness of detection, notification, and response, map wildlife cases, and evaluate knowledge and practices among at-risk populations.

Methods: A descriptive case series investigation was conducted from 16-20 November 2025, and characterized the outbreak by time, place, and host (animal/person). Tissue, soil, swabs, and water samples were collected and cultured to isolate B. anthracis. Timeliness was assessed using the 7-1-7 metrics by reviewing patient health records, veterinary reports, and key informant interviews. Wildlife case distribution was mapped using Quantum Geographic Information System (QGIS 3.44.7). A structured questionnaire was used to collect data on knowledge and practices. Data were cleaned in Microsoft Excel and analysed in Stata 17. Bloom's taxonomy scale was used to assess knowledge and practices, with ≥60% score regarded as adequate.

Results: A total of three confirmed wildlife cases and 13 suspected wildlife cases, with ten epidemiologically linked (probable) human cases were identified. The outbreak detection occurred exactly 7 days after emergence. Notification was done 6 days after detection, while public health responses were completed 10 days post-notification. Sixteen (16) wildlife cases were identified, and a spot map was generated. The case fatality rate (CFR) in captive lions was 42.9% (3/7). Among the 25 respondents (80% male; 60% <40 years; 40% livestock farmers, 60% wildlife facility workers), knowledge about anthrax was high, with 76% (19/25) having heard of the disease, 72% (18/25), and 64% (16/25) knew it affects wildlife and livestock, respectively. Overall, 60% (15/25) were aware of human infection and vaccination as means of livestock protection. Only 28% (7/25), however, practiced safe carcass disposal, and 48% (12/25) reported handling or consuming potentially contaminated meat.

Conclusion: The outbreak was characterized by delayed notification and response. High-risk community

practices were also observed. We recommend the implementation of coordinated, multisectoral risk communication and community engagement on safe carcass disposal.

Keywords: Anthrax, knowledge, practices, outbreak, Zambia

Background

Anthrax is an acute, fatal, zoonotic bacterial disease caused by the spore-forming, gram-positive bacterium called Bacillus anthracis (1). A defining feature of the pathogen is its ability to form spores that can persist in the environment for decades, creating long-term hotspots for infection (2). All warm-blooded animals are susceptible to B. anthracis; however, herbivores easily get infected due to grazing low on the ground in contaminated soils (3). Carnivores and humans on the other hand can become infected through exposure to contaminated environments or consumption of infected meat. In humans, the clinical presentation varies by route of exposure, manifesting primarily as cutaneous anthrax (through broken skin), gastrointestinal anthrax (through ingestion), or pulmonary anthrax (through spore inhalation) (4).

Climatic factors such as seasonal rainfall and temperature fluctuations play a pivotal role in the occurrence of anthrax outbreaks, apart from physiological stress and population density of hosts (5). Heavy rains can concentrate and deposit spores onto grazing areas and water sources, while drought forces animals to graze closer to contaminated soil (6). The long-term persistence of B. anthracis in the environment is largely determined by soil characteristics, particularly alkaline, calcium-rich soils, which favor spore survival (5).

Globally, anthrax is a priority pathogen in 65 countries, with 20,000-100,000 human cases occurring annually, posing a high risk particularly to communities in low and middle-income countries that are heavily dependent on livestock and wildlife for subsistence (2,3). This is because of low levels of awareness coupled with limited routine veterinary services.

In Sub-Saharan Africa, outbreaks are most common in the Congo Basin (Cameroon, Central African Republic, Democratic Republic of the Congo, Republic of the Congo, Equatorial Guinea and Gabon) and neighboring countries (7).

In Zambia, anthrax is recognized as an endemic threat, with persistent foci of infection documented in the Luangwa valley ecosystem and also in Southern and Western provinces. Outbreaks in these regions are typically precipitated by cycles of flooding and drought, which expose anthrax spores to grazing animals. The situation is exacerbated by critical gaps in surveillance systems, low livestock vaccination coverage, and poor carcass handling practices (8).

On 30 October 2025, a dead hippopotamus (Hippopotamus amphibius) was found in Mosi-oa-Tunya National Park. Its meat was fed to 36 lions (Panthera leo) at a private game facility on 31 October 2025, about 10 Km away from Mosi-oa-Tunya National Park. By November 6, 2025, seven lions began to show symptoms, which included facial swelling, loss of appetite, lethargy, and difficulty in breathing. This situation led to an outbreak investigation conducted from 16 November 2025 to 20 November 2025, which aimed to confirm the anthrax outbreak and to assess the promptness of detection, notification, and response. The study also aimed to map the distribution of wildlife cases and assess the knowledge and practices related to anthrax among the at-risk population around the park. These were exposed to the hippopotamus through handling of the carcass or consumption of meat.

Methods

Study Design Setting

The Mosi-oa-Tunya National Park, in Zambia's Southern Province, is located 12 km from Livingstone city. It spans about 66 km² along the Zambezi River (Zambia-Zimbabwe border). It lacks a game management area and hosts elephants, lions, antelopes, zebras, giraffes, warthogs, buffaloes, crocodiles, hippopotamuses, monkeys, birds, and fish. It is open all year-round, featuring the Victoria Falls, a United Nations Educational, Scientific and Cultural Organization (UNESCO) World Heritage Site, drawing more than 300,000 visitors annually. It is part of the Kavango-Zambezi (KAZA) Transfrontier Conservation Area (9). Figure 1 below shows the Mosi-oa-Tunya National Park in Livingstone District, where the study was conducted.

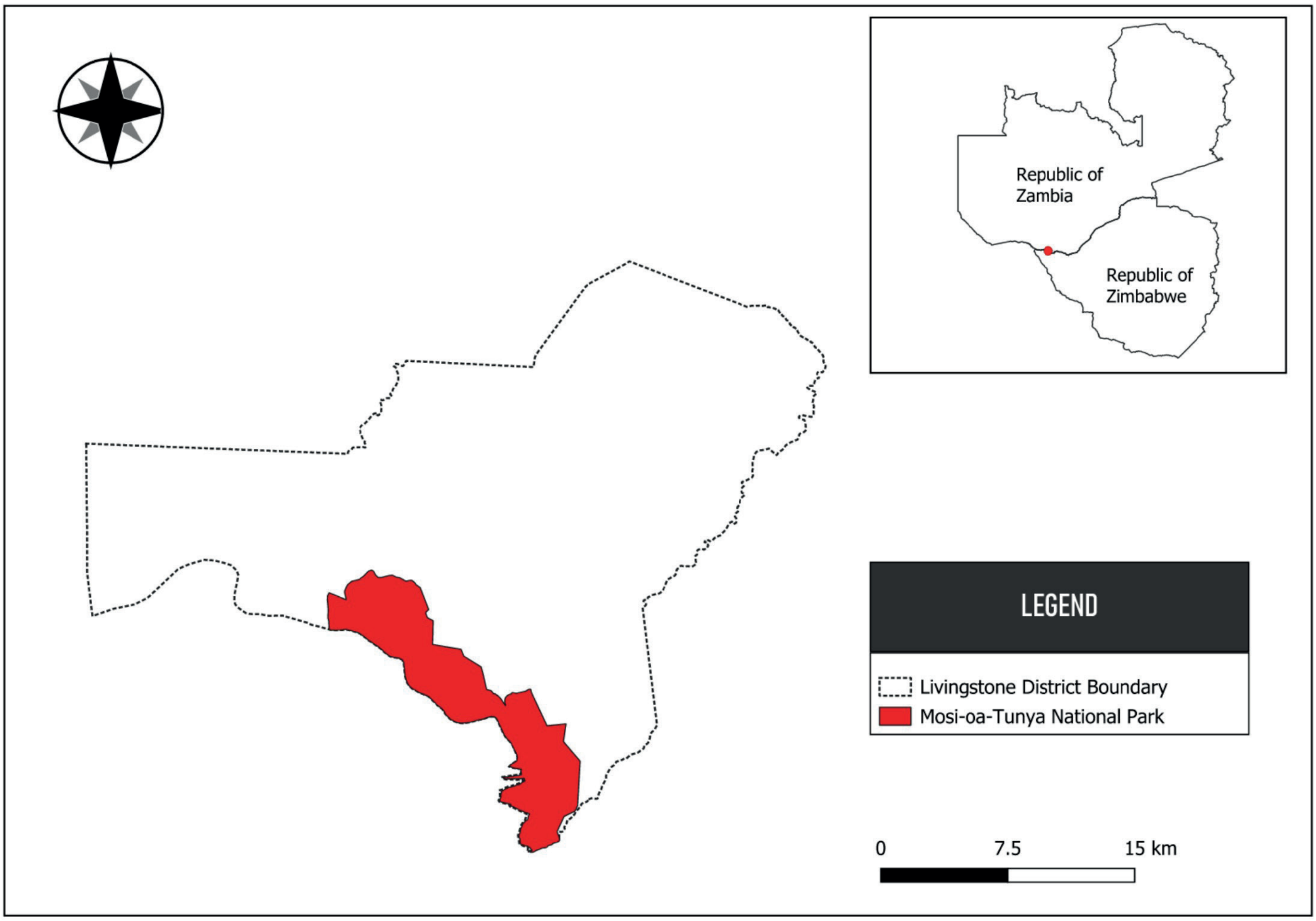

Figure 1: Map of the Study Site for an Investigation of an Anthrax Outbreak at the Wildlife-Human Interface in Mosi-oa-Tunya National Park, Zambia, October-November 2025.

Case Definitions

The case definitions we used were guided by the World Health Organization (WHO) and the World Organization for Animal Health guidelines (WOAH) (10,11).

Human Anthrax Cases

Suspected human anthrax case: Any person with acute illness showing one of the clinical forms-cutaneous (skin lesion evolving to a black eschar with edema), gastrointestinal (nausea, vomiting, anorexia, followed by fever), or pulmonary (viral-like respiratory symptoms rapidly progressing to breathing difficulty and fever)-between 1 October 2025 and 16 November 2025 residing in communities around Mosi-oa-Tunya National Park.

Probable human anthrax case: A suspected case with an epidemiological link to a confirmed case or exposure but without laboratory confirmation.

Confirmed human anthrax case: It is a clinically and epidemiologically compatible case that is laboratory-verified by isolation of B. anthracis from tissue or clinical specimens, detection of B. anthracis DNA by PCR, or other laboratory evidence such as anthrax toxins or serological confirmation.

Animal Anthrax Cases

Suspected animal anthrax case: Any animal found dead suddenly or within 24 hours of onset of illness exhibiting blood-stained, edematous fluid or exudate from external orifices in or around Mosi-oa Tunya National Park between 1 October 2025 to 16 November 2025.

Probable animal anthrax case: A suspected case with an epidemiological linkage to a confirmed case.

Confirmed animal anthrax case: An animal case that is a clinically and epidemiologically compatible case in which B. anthracis is isolated from clinical specimens (blood, tissues), or detected through culture and isolation, detection of B. anthracis DNA by PCR, or other laboratory evidence such as anthrax toxins or serological confirmation.

Study Design and Population

We conducted a descriptive case series study, purposively enrolling 25 individuals who were exposed to a dead hippopotamus through handling or consumption of its meat. 16 wildlife of Mosi-oa-Tunya National Park were also identified through case search and included in the study.

Data Collection

Swabs and tissues were collected from wildlife and one human. Environmental samples collected included soil and water. The samples were transported at 4°C in sterile leak-proof containers to the Central Veterinary Research Institute (CVRI) laboratories in Choma and Lusaka for analysis. To assess timeliness, review of reports from the District Veterinary Office and Department of Wildlife and National Parks was conducted alongside key informant interviews from health, veterinary and wildlife authorities. A review of patient health records was also done. In addition, the Global Positioning System (GPS) coordinates were collected for all wildlife mortalities. A structured questionnaire was generated using Kobo Toolbox to capture key information on knowledge and practices pertaining to anthrax among community members around the Mosi-oa-Tunya National Park.

Laboratory analysis

Samples were processed at CVRI laboratories in Choma and Lusaka using guidelines stipulated in the World Organization for Animal Health Manual of Diagnostic Tests and Vaccines for Terrestrial Animals Chapter 3.1.1 (12).

Culture and isolation were conducted in biosafety level 2 (BSL-2), utilizing Biosafety level 3 (BSL-3) practices. 5% sheep blood agar plates were inoculated with smears of tissues and swabs and incubated aerobically at 37°C for 24 hours. The cultured colonies were then observed using a magnifying glass for identification of characteristic colonies measuring 0.3-0.5 cm, appearing grey-white, non-hemolytic with ground-glass texture and "Medusa head" edge curling. This was done in biological safety cabinets using N95 respirators, double gloves, and gowns. The samples and cultures were then autoclaved at 121°C for 60 minutes before disposal to prevent environmental contamination.

Data Analysis

The 7-1-7 metrics were used to assess timeliness, where detection is expected to be ≤7 days post-emergence, notification ≤1-day post-detection and early response activities completed in ≤7 days post-notification (13). The GPS coordinates for all wildlife cases were exported to Microsoft Excel and cleaned. The Quantum Geographic Information Systems (QGIS 3.44.7) was used to develop a spot map utilizing the GPS coordinates for the wildlife cases. For the assessment of knowledge and practices, the data were imported into STATA version 17 for descriptive analysis. Bloom's taxonomy was used to classify respondents' knowledge and practice levels. Each "Yes" response scored 1 point (correct), while "No" or "Don't know" scored 0 for each knowledge or practice question. The sum of the "Yes" scores was calculated and used as the numerator, while the denominator was the total number of respondents who answered each question, respectively. This was then multiplied by 100 to find the percentage score. A score of ≥60% was considered an adequate level (14).

Ethical consideration

The Zambia National Public Health Institute (ZNPHI) has a waiver for outbreak investigations stipulated in the ZNPHI Act No. 19 of 2020 to generate quick evidence for informed public health decision-making. However, approval was obtained from Livingstone District Health Office and health facility staff to review patient medical records. All participants gave verbal consent, and their data were handled with strict confidentiality and anonymity throughout the investigation. Non-maleficence was ensured to both humans and animals during the study.

Results

Out of the 25 respondents, 80% (20/25) were male and 20% (5/25) were female, while 60% (15/25) were below 40 years old. Among these participants, livestock farmers were 40% (10/25), while wildlife facility workers were 60% (15/25). Table 1 below shows the demographic characteristics of the individuals who participated in this study.

Table 1: *Demographic Characteristics of Respondents in an Anthrax Outbreak Investigation at the Wildlife-Human Interface in Mosi-oa-Tunya National Park, Zambia, October-November 2025 (n=25)*

Demographic Characteristics	Number (%)
Age	
<40	15 (60%)
>40	10 (40%)
Gender	
Female	5 (20%)
Male	20 (80%)
Occupation	
Livestock farmers	10 (40%)
Wildlife farm workers	15 (60%)

The findings of this study showed that 76% (19/25) of the respondents had eaten hippopotamus meat while 24% (6/25) didn't. Furthermore, 60% (15/25) confirmed handling hippopotamus carcass and 40% (10/25) didn't. The study revealed that 40% (10/25) reported illness, while 60% (15/25) remained asymptomatic. All the symptomatic respondents were treated with ciprofloxacin 500mg (q12) orally for 7 days before the investigation was conducted. Table 2 below summarizes the symptoms reported by the respondents during the investigation.

Table 2: *Symptoms reported among the Respondents in an Anthrax Outbreak Investigation at the Wildlife-Human Interface in Mosi-oa-Tunya National Park, Zambia, October-November 2025 (n=25)*

Symptom	Number (%)
Fever and chills	6 (24%)
Stomach pain	5 (20%)
Nausea and vomiting	1 (4%)
Diarrhea	1 (4%)
Sore or black eschar	1 (4%)
Sore throat	1 (4%)
Hoarseness of voice	1 (4%)
Pain when swallowing	1 (4%)
No symptoms	15 (60%)

A total of 23 samples were collected, which included seven tissues, six soil samples, two water samples, eight animal swabs and one human swab. Three tissue samples, one from a lion and two from hippopotamuses, tested positive for anthrax. However, the only human swab from a wound (presenting with a black eschar) tested negative despite being epidemiologically linked to the index wildlife case through the opening of the carcass and consumption of meat. Nine out of the ten respondents presented with compatible symptoms of gastrointestinal anthrax and one respondent presented with cutaneous anthrax. Furthermore, all of them were epidemiologically linked to the dead hippopotamus, which tested positive for B. anthracis through contact with the carcass or consumption of meat. Therefore, the investigation identified 10 probable human anthrax cases.

The anthrax outbreak was detected on 6 November 2025, exactly 7 days after the emergence of the outbreak on 30 October 2025, meeting the 7-1-7 target's detection threshold of ≤7 days. However, an outbreak report was submitted to the National Livestock Epidemiology and Information Center (NALEIC) on 12 November 2025, 6 days after the detection of the outbreak, indicating delayed notification. Implementation of early response activities was completed on 22 November 2025, 10 days after notification of the outbreak. The response activities included active search for human and wildlife cases, sample collection, alerting nearby health facilities of the outbreak (Maunga and Mosi-oa-Tunya health facilities), reminder of case definition to health facility workers, risk communication and community engagement (RCCE), and assembling the One Health team for implementation of control and preventive measures. Figure 2 below summarizes the 7-1-7 metrics assessment through review of health records and reports and interviews with health workers, veterinary staff and wildlife authorities.

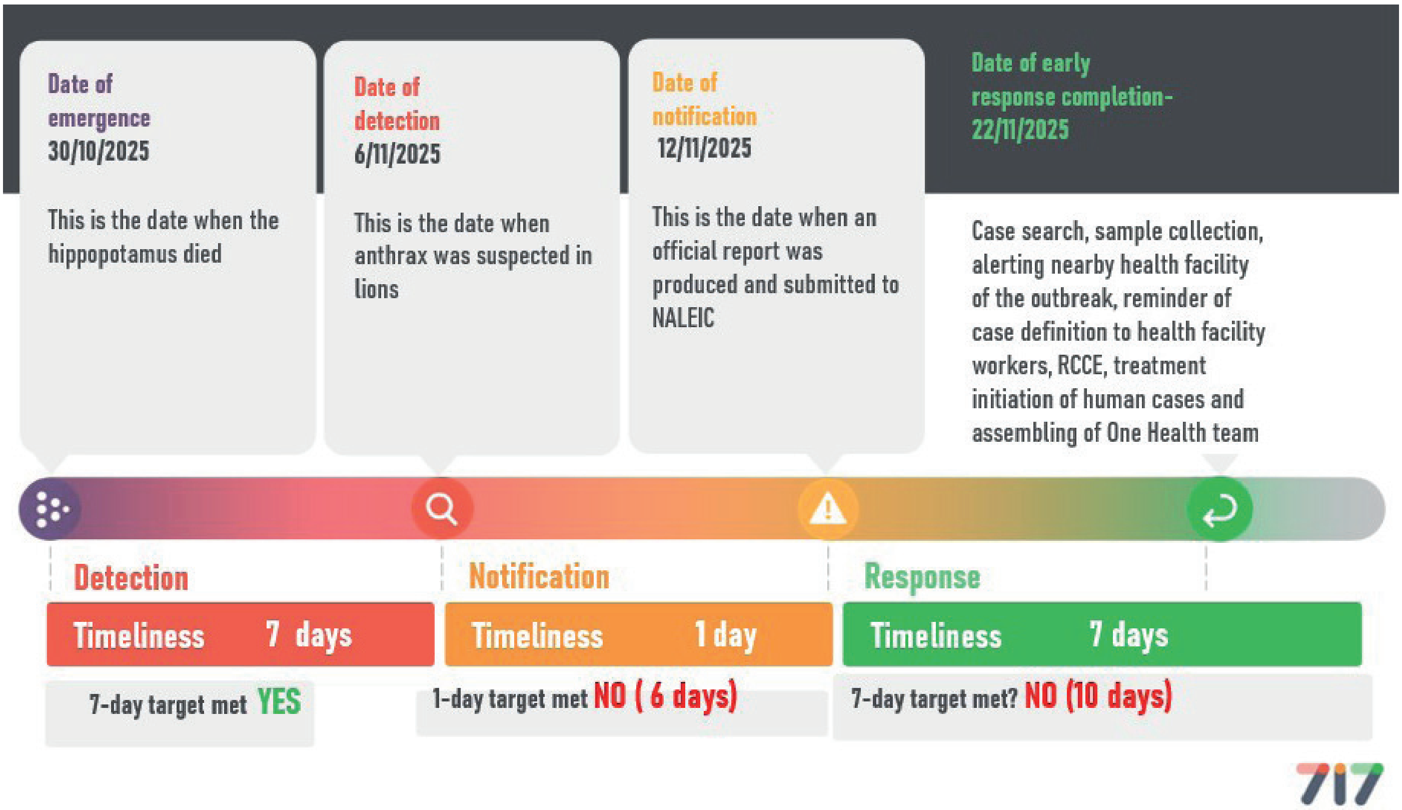

Figure 2: Summary of the assessment of the timeliness of detection, notification and response using the 7-1-7 metrics during an Anthrax Outbreak, Livingstone District, 2025.

During the investigation, we identified strong clinical suspicion or awareness of case definition of anthrax by the wildlife veterinarian as an enabler to early detection of the outbreak. On the other hand, One Health information sharing failure was identified as a bottleneck for delayed notification. Multi-agency coordination failure and risk communications delay were bottlenecks identified for delayed response. Table 3 below is a summary of the bottlenecks and enablers of detection, notification and response regarding the anthrax outbreak in Mosi-oa-Tunya National Park.

Table 3: Bottlenecks and Enablers of anthrax Detection, Notification and Response, Livingstone District, 2025.

Metric	Delay	Bottleneck/Enabler	Comment
Detection	Met the target	Strong clinical suspicion or awareness of case definition by wildlife veterinarian	The wildlife veterinarian had good suspicion of anthrax based on 7 captive lions showing facial edema, inappetence, recumbency and dyspnea
Notification	Did not meet the target	One Health information sharing failure	Immediate reporting to responsible authorities and One Health stakeholders was not done on time despite knowing that anthrax is a notifiable zoonotic disease
Response	Did not meet the target	Multi-agency coordination failure	There was delayed assembly of One Health response team involving MOH*, DVS†, and DNPW§ due to absence of pre-existing district-level coordination
		Risk communications delay	Late implementation of RCCE activities to high-risk communities

**MOH-Ministry of Health, †DVS-Department of Veterinary Services, §DNPW-Department of National Parks and Wildlife, RCCE- Risk Communication and Community Engagement*

Among the 36 captive lions at the private game facility, seven had presented with facial and oral swelling, loss of appetite (inappetence), recumbency (lying down and reluctance to move) and labored breathing (dyspnea). Three mortalities out of the seven suspected cases died, representing a Case Fatality Rate (CFR) of 42.9% (3/7).

Wildlife mortalities totaling 16 were recorded in Mosi-oa-Tunya National Park and the private game facility exhibiting blood-stained, edematous fluid or exudate from external orifices. These included four hippopotamuses (H. amphibius), three lions (P. leo), five buffaloes (Syncerus caffer), two zebras (Equus quagga), and two warthogs (Phacochoerus africanus). Three tissue samples had tested positive for anthrax, indicating three confirmed cases and 13 suspected cases in wildlife. Figure 3 below shows the distribution of the wildlife mortalities in Mosi-oa-Tunya National Park and the private game facility identified during the investigation.

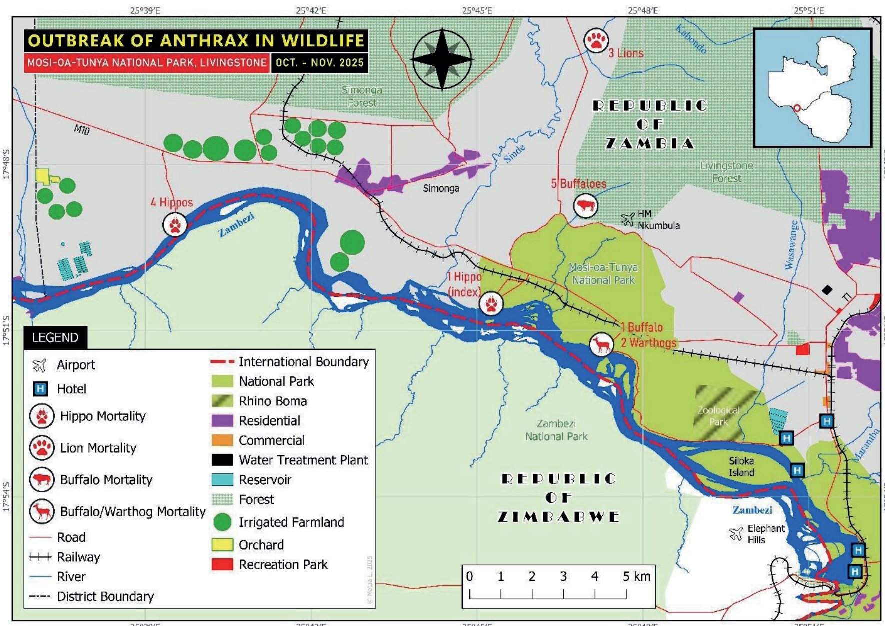

Figure 3: Distribution of wildlife mortalities in Mosi-oa-Tunya National Park and private game facility, Livingstone District, 2025.

The epidemic curve for this outbreak showed a temporal clustering of cases occurred 6 to 7 days after exposure. Figure 4 below is epidemic curve showing the number of human and wildlife cases per day and the date of onset of clinical signs in Livingstone District.

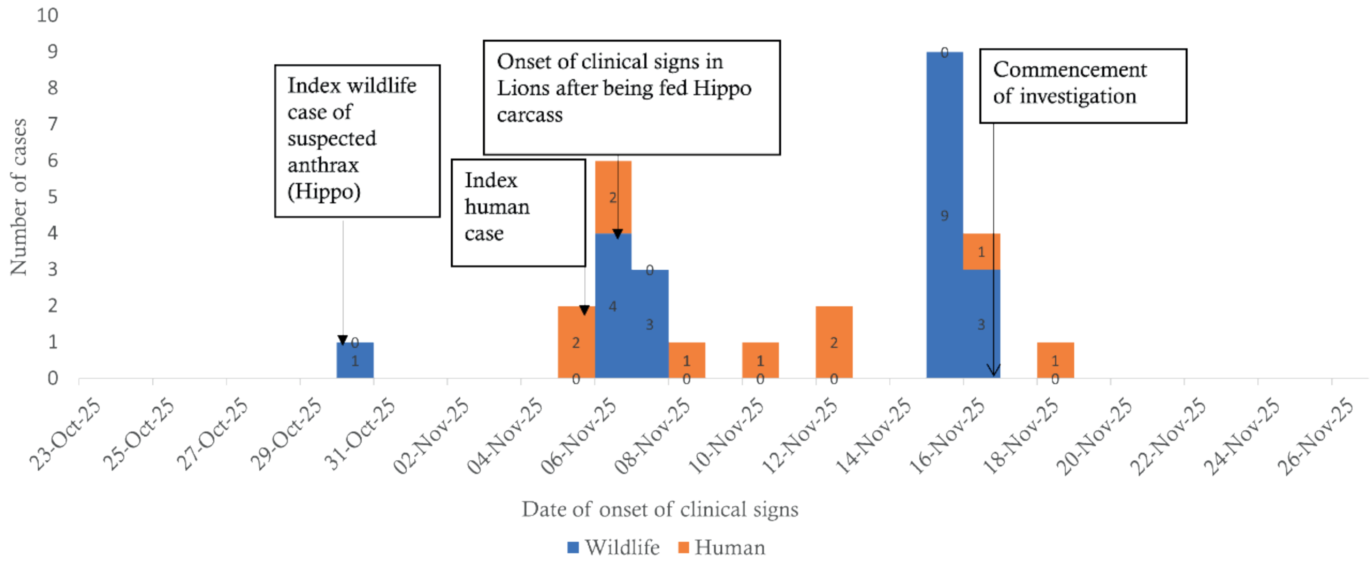

Figure 4: Epidemic curve of the anthrax outbreak in Mosi-oa-Tunya National Park and adjacent communities in Livingstone District, October-November 2025.

In terms of knowledge assessment regarding anthrax among the respondents, 76% (19/25) had heard of anthrax, 72% (18/25) were aware that it affects wildlife, 64% (16/25) knew it affects livestock. Additionally, 60% (15/25) recognized that anthrax can infect humans, and 60% (15/25) had knowledge that vaccination of livestock can prevent anthrax infection. Table 4 below shows the knowledge questions asked to the respondents and the frequencies in terms of response, as well as the score assessment.

Table 4: Knowledge about anthrax among respondents, Livingstone District, 2025.

Question	Yes n (%)	No n (%)	Don't know n (%)	Score Assessment
Have you heard about anthrax?	19 (76)	6 (24)	0 (0)	Knowledgeable
Does anthrax affect game?	18 (72)	5 (20)	2 (8)	Knowledgeable
Does anthrax affect livestock?	16 (64)	7 (28)	2 (8)	Knowledgeable
Does anthrax affect humans?	15 (60)	8 (32)	2 (8)	Knowledgeable
Does vaccination prevent anthrax?	15 (60)	4 (16)	6 (24)	Knowledgeable

In terms of practices concerning anthrax, safe carcass handling was low at 28% (7/25) of respondents, veterinary consultation was high at 84% (21/25), and 48% (12/25) of the respondents consumed potentially contaminated meat. Table 5 below shows the practice questions asked to the respondents and the frequencies in terms of response as well as the score assessment.

Table 5: Practices regarding anthrax among respondents, Livingstone District, 2025.

Practice	Yes n (%)	No n (%)	Score Assessment
Do you handle animal carcasses that die suddenly with precaution?	7 (28%)	18 (72%)	Low practice of safe carcass handling
Do you consult veterinary staff when your animals are sick?	21 (84%)	4 (16%)	High consultation rate with veterinary staff
Do you eat meat sourced from animals that died of unknown cause?	12 (48%)	13 (52%)	High-risk behavior in consuming unknown meat

Discussion

This investigation confirmed an anthrax outbreak at the wildlife-human interface of Mosi-oa-Tunya National Park. The failure to isolate B. anthracis from the human swab could be as a result of prior antibiotic treatment which was administered five days before sample collection underscoring the critical importance of collecting specimens for laboratory investigation before initiating antimicrobial therapy (2). Despite the lack of laboratory confirmation in the human cases, a strong epidemiological link was established through direct handling and consumption of the contaminated hippopotamus carcass which was a confirmed positive case, followed by anthrax compatible symptoms. This finding agrees with most of the anthrax outbreaks in Zambia affecting humans and livestock, where these cases primarily originate from wildlife (15). For instance, between 1 June to 28 July, 2023, 26 people in Sinazongwe District were infected with anthrax after consuming contaminated hippopotamus meat (16).

The practice of carcass scavenging as a disposal method, while economically motivated, is discouraged because of its potential to transmit pathogens to other animals and the ecosystems they are part of (17). This investigation revealed the transmission of B. anthracis from a dead hippopotamus to lions at a private game facility after being fed the meat. Anthrax being an infectious zoonotic disease requires careful adherence to protocols laid down in Chapter 4.13 of the Terrestrial Animal Health Code on disposal of dead animals (18). It recommends incineration as the ideal method of disposal of an anthrax carcass. Where this method is not possible, deep burial is the alternative. Unlike burial, burning has the advantage of destroying anthrax spores and reducing the number of spores available in the environment and, therefore, reducing the chance of spores resurfacing later.

This outbreak investigation serves as a compelling case study for the One Health approach in achieving the 7-1-7 targets. It demonstrates the complex links between environmental health, animal health, and human health in the successful control of Zoonoses. The investigation revealed critical bottlenecks in the notification and response system, including One Health information sharing failure, multi-agency coordination failure and risk communications delay, which delayed notification and intervention. These gaps are similar to findings in a retrospective observational study conducted in Brazil, Ethiopia, Liberia, Nigeria, and Uganda reviewing timeliness of detection, notification and response of public health events (13). Addressing these gaps requires the establishment of coordinated One Health committees that integrates wildlife authorities, veterinary services, human health sectors, environmental sectors and other relevant sectors to enable rapid detection, notification and joint response of zoonoses.

The wildlife cases of anthrax in Mosi-oa-Tunya National Park were mostly distributed along the Zambezi River and the outbreak occurred at the onset of the rain season, supporting the known ecology of this disease (19). During this period, flooding and soil disturbance expose dormant spores, while short and sparse vegetation forces herbivores to graze closer to the ground because grasses are short and sparse, and there is increased movement and mixing of animals in search of pasture and drinking water (20). This climatic and environmental situation increases the likelihood of spore ingestion and creates a high-risk interface where wildlife, livestock, and humans converge around shared water and pasture resources (21). Additionally, the Zambezi River serves as a natural corridor for wildlife movement and a shared resource for communities, particularly within the KAZA, one of the world's largest conservation zones. This outbreak therefore poses a high-risk for transboundary transmission of anthrax to Angola, Botswana, Namibia and Zimbabwe.

Finally, the assessment of community knowledge and practices revealed high knowledge about anthrax and low safe practices despite the small sample size. This means that high levels of awareness regarding anthrax risks did not translate into safe behaviors, aligning with studies conducted around national parks and pastoral communities, where economic pressure, food insecurity, and cultural norms drive continued consumption of contaminated meat, despite awareness of the zoonotic risk (15,22,23). The high level of veterinary consultation in this study, however, suggests a trusted entry point for tailored risk communication and community engagement (RCCE), livestock vaccination, and community-based surveillance, for anthrax prevention and control, while embracing the One Health approach (3,24).

Conclusion

This investigation confirmed the anthrax outbreak. It was characterized by delayed notification and response. The wildlife cases were distributed along the Zambezi River, with high-risk community practices despite high knowledge among sampled high-risk respondents.

Recommendations

Based on this investigation, MOH, DVS, DNPW and other relevant stakeholders should consider establishing a functional One Health coordination platform for formalized reporting and a multi-agency response plan for Zoonotic diseases. They should also consider enforcement of carcass disposal in accordance with Chapter 4.13 of the Terrestrial Animal Health Code. Moreover, targeted RCCE campaigns should be undertaken by a One Health team to promote safe handling and disposal of suspected anthrax carcasses. The MOH, DVS and DNPW should advocate for transboundary collaboration within the KAZA on the implementation of anthrax control and prevention measures.

Limitations

The findings of this study must be interpreted within the context of several limitations, including the lack of laboratory confirmation for the epidemiologically linked human cases. The small sample size of 25 participants limits the generalizability of the knowledge and practices assessment to the wider community and different settings. Additionally, the potential for recall bias exists in the self-reported data, and the delay in the investigation may have compromised the quality of environmental samples. Furthermore, prior antibiotic treatment before specimen collection may have reduced laboratory confirmation in human cases.

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Contributors

Mathews Sichalwe and Humphrey Banda developed the protocol and led the outbreak investigation. Benson Bowa and Brian Musalo were responsible for sample collection, while Benson Bowa conducted the laboratory analysis. Mathews Sichalwe, Mwendalubi Hazyondo and Conrad Chibale Collected data through questionnaire administration to community members. Data analysis and initial manuscript drafting were undertaken by Mathews Sichalwe, Humphrey Banda, and Paul Mulopa. Nyambe Sinyange, Dabwitso Banda, Gregory Bwalya, and Liywaali Maata conducted data quality checks. All authors reviewed and approved the final version of the manuscript.

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