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ORIGINAL ARTICLE

Field immobilization for treatment of an unknown illness in a wild chimpanzee (*Pan troglodytes schweinfurthii*) at Gombe National Park, Tanzania: findings, challenges, and lessons learned

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Abstract Infectious diseases are widely presumed to be one of the greatest threats to ape conservation in the wild. Human diseases are of particular concern, and the costs and benefits of human presence in protected areas with apes are regularly debated. While numerous syndromes with fatal outcomes have recently been described, precise identification of pathogens remains difficult. These diagnostic difficulties are compounded by the fact that direct veterinary intervention on wild apes is quite rare. Here we present the unique case of a wild chimpanzee at Gombe National Park that was observed with a severe illness and was subsequently examined and treated in the field. Multiple specimens were collected and tested with the aim of identifying the pathogen responsible for the illness. Our findings represent the first extensive screening of a living wild chimpanzee, yet despite our efforts, the cause and source of illness remain unknown. Nevertheless, our findings represent valuable baseline data for the ape conservation community and for comparison with other recent findings. In addition, we present the case here to demonstrate the planning required and multiple types of expertise necessary to maximize the amount of data obtained from such a rare intervention, and to provide lessons learned for future studies.

Keywords Apes · Health · Disease · Intervention · Diagnostics

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Introduction

Infectious diseases are widely recognized as a critical threat to conservation for many wildlife populations (Daszak et al. 2001). In great apes, numerous epidemic disease outbreaks have been associated with mortality events (Leendertz et al. 2006; Ryan and Walsh 2011). High-profile examples include Ebola virus in Taï chimpanzees (Formenty et al. 1999) and western lowland gorillas (Walsh et al. 2003; Bermejo et al. 2006), anthrax in Taï chimpanzees (Leendertz et al. 2004), an "AIDS-like" disease in Mahale chimpanzees (Nishida et al. 2003), as well as pathogenic SIVcpz infections in Gombe chimpanzees (Keele et al. 2009). Due to their physiological and genetic similarities to humans, nonhuman primates may be highly susceptible to diseases common to humans (Wolfe et al. 1998). Indeed, there is a widespread perception that human infectious diseases pose one of the greatest risks to the survival of apes in the wild (Homsy 1999; Leendertz et al. 2006; Altizer et al. 2007; Chi et al. 2007; Pederson et al. 2007; Köndgen et al. 2008). Although confirming cross-species transfers of infectious agents from humans to wild nonhuman primates can be difficult, numerous suspected and/or confirmed transmission events have been documented in the wild (Nizeyi et al. 2001; Köndgen et al. 2008; Rwego et al. 2008) and in captive settings (Schaumberg et al. 2012; Kooriyama et al. 2013).

Direct human–great ape interaction in protected habitats is usually limited to behavioral and/or conservation research, tourism, and variable levels of exposure to local people (e.g., crop-raiding by apes, local people visiting relatives in the protected area). Indirect exposure can also occur through human-associated factors such as domestic animals (pets or livestock), latrines, and garbage pits. While the risks of such close interaction should be assessed and mitigated (Garber 2008; Köndgen et al. 2008; Pusey et al. 2008), research and conservation efforts present invaluable opportunities to gather information about free-ranging animals. In particular, identifying diseases in wild primate populations and understanding their transmission dynamics provide the foundation upon which conservation management strategies can be designed and implemented (Travis et al. 2008). Individual cases where examination and/or treatment by a veterinarian is deemed necessary can also shed light on how wild animals contract diseases and cope with illnesses. However, veterinary treatment of wild nonhuman primates, and chimpanzees in particular, is complicated due to anesthesia-associated risks and habituation issues (Lukasik 2002; Travis et al. 2008; Ryan and Walsh 2011), and as such, each protected area allows differing levels of intervention.

The study of chimpanzees by Goodall and colleagues at Gombe National Park, Tanzania constitutes the longest continuous study of any great ape population. Chimpanzees of the two habituated communities, Mitumba in the north (studied since 1985, habituated by the mid-1990s) and Kasekela in the center of the park (studied since 1960, habituated by the mid-1960s), are followed and monitored daily by research assistants who collect long-term behavioral data. Members of the Kalande community in the south of the park remain unhabituated but have been monitored regularly since 1999 (Rudicell et al. 2010). The total chimpanzee population at Gombe appears to have declined from as many as 120–150 in the 1960s (Pusey et al. 2007) to 99–105 at the beginning of 2010 (Rudicell et al. 2010), with death associated with observable signs of disease as the leading cause of mortality (Williams et al. 2008). Major epidemics at Gombe included suspected polio in 1966, respiratory syndromes in 1968, 1987, 1996, 2000, and 2002, and sarcoptic mange in 1997 (Goodall 1983, 1986; Nutter 1996; Mlengeya 2000; Walton et al. 2004; Williams et al. 2008). Most recently, SIVcpz infection has been found to result in increased mortality (Keele et al. 2009) and has been identified to be a major contributing factor in the decline of the Kalande community (Rudicell et al. 2010).

A comprehensive noninvasive health-monitoring program was initiated in these communities in 2004 with the aim of understanding baseline levels of health in the population and identifying the presence and impacts of various pathogens (Lonsdorf et al. 2006; Travis et al. 2008; Gillespie et al. 2010; Terio et al. 2011). In the second year of the project, a chimpanzee known as Faustino (FO) was observed by field researchers with extreme emaciation and suspected paralysis of the hindlimbs. Due to the long history of human presence in the park, research and management staff were particularly concerned about the possibility that Faustino was infected with a human pathogen. Therefore, a field intervention by a trained veterinarian was conducted in an attempt to determine the cause of illness. Direct veterinary intervention in the form of immobilization occurred only 3 times in Gombe prior to the case presented here (Table 1). These included one chimpanzee that was caught in a snare and required hand amputation (Loretta), one moribund chimpanzee that received palliative care and subsequently survived (Goblin), and one chimpanzee that developed a facial abscess that was sampled and treated (Gilka).

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Table 1 Field immobilizations conducted at Gombe National Park (1960–2012)

Chimpanzee ID	Year	Reason	Source
Gilka	1970	Take biopsy of bulbous facial abscess	Goodall 1986
Goblin	1989	Examine and treat testicular wound	Goodall 1992
Loretta	1996	Amputation of hand as a result of snare injury	GSRC, unpublished records
Faustino	2005	Extreme illness with hindlimb weakness	This report

GSRC Gombe Stream Research Centre

While the historic interventions focused on diagnosis and treatment to prevent animal suffering, the intervention reported here (Faustino) was conducted for both welfare and investigative purposes. In this case, extensive serological testing provided unique information regarding reactivity to antibody tests, and hence agents to which this chimpanzee may have been exposed via people, other animals or the environment. While it is difficult to draw general conclusions based on the analysis of a single individual, the intervention provided a unique opportunity to conduct testing on a living, wild chimpanzee and thus represents an important case study. Moreover, a recent serological survey conducted in group-living nonvaccinated captive chimpanzees by Kooriyama et al. (2013) allows a reference point for initial comparisons. As such, we present this case here in order to (1) describe the first reported field intervention and the subsequent diagnostic test results on a wild chimpanzee, and (2) demonstrate the planning required and multiple types of expertise necessary to maximize the amount of data obtained from such a rare intervention and provide lessons learned for future studies. Given the unique situation of working in a field setting with limited equipment and electricity, this report diverges in format and detail from a typical veterinary case report. However, our intent here is to describe our experiences to other primatologists working in the field who are increasingly faced with similar situations.

Case report

Case history

A 16-year-old adult male chimpanzee (*Pan troglodytes schweinfurthii*)—Faustino (FO)—of the Kasekela community at Gombe National Park presented with deteriorating hindlimb locomotion in late 2005. Faustino had been observed on 17 November 2005 by researchers who noted his unusual behavior. Field researchers subsequently recorded observational data each time FO was seen (Table 2). No other animal in the study group was observed to have similar clinical signs.

For almost 2 weeks following the initial observation, Faustino's condition deteriorated (see Fig. 1). He became progressively unresponsive to external stimuli and uninterested in provisioned food, only ingesting some water. He had watery diarrhea throughout this period, and flies continuously congregated around him. With respect to locomotion, Faustino lay recumbent without sitting upright, and his only method of locomotion was to roll laterally. After continued inappetance, permission to provide Faustino bananas was requested from and granted by the Chief Park Warden. The food items were cleaned with the human skin disinfectant solution trichlorocarbanilide (Dettol^{TM}) and rinsed before distribution to the chimpanzee. Medical intervention for parasite control was considered but deferred pending veterinary assessment. Tanzania National Parks staff, who have regulatory and management authority over the wildlife in the park, made the decision to allow a trained veterinarian (R.S.) to administer supportive care 13 days after the first indication of illness. While park staff and researchers have generally maintained a hands-off philosophy towards animal health, in this case the decision to intervene was made due to concern over its prolonged nature (welfare) and potential infectiousness (population risk). Thus, the case presented herein complied with the legal authority and permitting requirements of the host country.

Intervention and recovery

On 1 December 2005, Faustino was by himself, close to the research staff quarters, immobile and largely unresponsive. Due to the subject's condition, R.S. determined that it was possible and preferable to attempt hand-injection, and Faustino was subsequently immobilized with 10 % ketamine concentration (Vetelar Fort Dodge Laboratories Inc., 250 mg i.m.) by hand-injection in the right triceps muscle. During the intervention, it was mild but sunny (approximately 75–80 °F), so the procedure took place in the shade (underneath trees) to prevent hyperthermia. Due to potential zoonotic disease risks, all staff participating in the intervention wore personal protection equipment including Tyvek suits, face masks, boot coverings, gloves, and eye protection (see Fig. 2).

Oxygen saturation determined by pulse oximeter (Nellcor^{TM}) varied from 86 to 93 %, and respirations under

Table 2 Behavioral observations and details reported by Gombe Stream Research Centre field staff until November 30th when reported by veterinarian R.S.

Date	Observations	Comments
17 Nov 2005	Slight wobble in left hindlimb	
	Tendency to slump against ground or a support to rest while following females—noted 14 times during the day	
	Watery diarrhea and flatulence	
24 Nov 2005	Thin and weak, visible loss of body condition	See Fig. 1 for photographic observation. A video of FO's rolling movement was recorded and sent to a neurologist for analysis
	Difficulty moving and chewing, unusual head movements made when tearing off pieces of fruit	
	Legs buckled when attempting to stand so focal rolled on his side in pursuit of food	
	Did not move when a large dead branch fell near him	
	Eyes bright and alert	
25 Nov 2005	Tried to climb into trees but was largely unsuccessful. Used arms only to climb into a tree to nest 2 meters off ground	Bananas offered
26 Nov 2005	Only left nest when offered bananas	
	Ate 15 bananas, 4 wild fruits, and some leaves	
	Stationary entire day other than rolling occasionally for short distances	
27 Nov 2005	As above, but with further reduced appetite	
28 Nov 2005	Consumed only one banana offered	
	Shivering and curled up to avoid rain and cold	
30 Nov 2005	Dehydrated, lethargic, and severely cachexic	
	Subject remained immobile and recumbent on side, unresponsive to stimuli	
	Very thin mucoid diarrhea	

The intervention took place on December 1st

Fig. 1 Photograph of Faustino taken on 24 November 2005, 7 days after he presented with abnormal behavior. Photo by M.W.

Fig. 2 Photograph taken during the intervention, showing shaded location and personal protection equipment worn by staff. Photo by M.W.

anesthesia were 15 breaths per minute. His heart rate was approximately 75 beats per minute (bpm), and rectal temperature from a digital thermometer (Elektronisches Fieber^{TM}) averaged 37.1 °C during the procedure. Normal reference ranges for pulse oximetry, rectal temperature, resting heart and respiratory rate of free-ranging chimpanzees do not exist to our knowledge. In a published report of an intervention on a wild chimpanzee that had

been speared, Hyeroba et al. (2011) reported a heart rate of approximately 94–98 bpm and a rectal temperature of 36.7–37.8 °C. In one captive study of 7 individuals, chimpanzees under anesthesia had the following ranges: rectal temperature 36.8 ± 0.6 °C, heart rate 110.8 ± 18.8 bpm, respiratory rate 24.4 ± 6.5 breaths per minute, and mean oxygen saturation between 87.8 ± 1.7 % at the initial time of contact to 93.0 ± 1.7 % after 20 min (Kearns et al. 2000). However, these values are highly dependent on the type of anesthetic utilized, ambient temperatures, and whether the animal was administered supplemental oxygen, so direct comparison is not appropriate.

A chest auscultation and examination of the lymph nodes did not detect any abnormalities. Faustino's gums were pale and dry, and capillary refill time was slow (greater than 5 s). Although the teeth were in good condition, his body condition was poor, with an estimated body score of 2 on a 5-point scale and signs of emaciation. Dried diarrhea was present around the anus. No obvious external signs of physical trauma were evident.

During the clinical examination, numerous biological samples were collected and preserved based upon the local availability of preservatives. Readers are referred to Leendertz et al. (2006) for a full description of ideal sample collection and preservation techniques that can be used in the field. In this case, whole blood was collected into ethylenediamine tetraacetic acid (EDTA) tubes and serum vacutainers and stored in a −13 °C solar freezer. Blood smears were prepared, and whole blood was spotted on filter paper, which was air-dried and stored with silica gel in a plastic zip-lock bag. Additional blood samples were preserved in glycerin and RNAlater (Ambion™, Austin, TX, USA). Fecal samples in glycerin and RNAlater were also collected.

Following physical examination, Faustino (estimated weight 30 kg) was treated empirically for systemic bacterial infection with parenteral enrofloxacin (Baytril™ 50; Bayer Australia Ltd, 150 mg i.m.) and amoxicillin (Clavulox™; Pfizer, 200 mg i.m.). Although he was not febrile, the subject received anti-inflammatories (flunixin meglumine, Finadyne™; Schering Plough Animal Health, 75 mg i.m.) and intravenous fluids (50 % dextrose 5 ml; 5 % dextrose IV 750 ml) as supportive measures. To reduce potential parasite burden effects on overall systemic health, a parenteral antihelminthic (ivermectin, Bimectin™; Bimeda, 12 mg s.c.) was administered.

Faustino awoke from anesthesia after 1 h and appeared calm during recovery. He rested quietly on the ground, looking directly at the observers without showing any signs of anxiety (which was considered normal for this well-habituated animal). The following day (2 December 2005) Faustino moved for the first time, relocating 20 m away, and sat upright for more than 16 min before returning to his laterally recumbent position. He showed interest in food and consumed wild fruits and provisioned bananas, but attempts to provide water failed and no bowel movements were observed. Within 3 days of the intervention (3 December 2005), he was alert and responsive and began to locomote, first attempting to walk bipedally and then dropping to all fours. Although bowel movements were still not seen, his appetite and frequency of locomotion increased. On day four post intervention, he was seen walking on all four limbs for approximately three meters and showed aggression towards baboons in defense of his food for the first time since the illness began. Faustino continued gaining strength and began socializing with other chimpanzees of his community 11 days after the procedure, albeit still showing some signs of weakness. By 23 December 2005, he had regained normal motor function in all four of his limbs and has maintained normal function until the present (May 2013).

Test results

Blood diagnostics

Blood (preserved in EDTA and a smear) was immediately taken to Kigoma Hospital, Kigoma Region, Tanzania and analyzed with the following results: total white blood cell count (6.6×10^9/l), neutrophils (59 %; 3,894 cells/µl), monocytes (2 %; 132 cells/µl), lymphocytes (36 %; 2,376 cells/µl), and eosinophils (3 %; 198 cells/µl), which are considered within species normal ranges (Hainsey et al. 1993; Howell et al. 2003). Malarial parasites were not noted upon examination of blood smears. After shipment to the USA, serum and plasma were submitted to the Simian Diagnostic Laboratory, Ltd. (San Antonio, TX 78229, USA) for serologic testing using standard techniques (Table 3). Blood typing, red cell indices, and dry chemistries were unsuccessful, due to either limited volume or sample preservation.

SIVcpz diagnostics

Blood collected during the intervention was also tested for SIVcpz-specific antibodies and viral nucleic acids. Serum (0.5 ml) was centrifuged at 20,000×*g* for 1 h, and the resulting pellet was resuspended for DNA extraction using the QIAamp DNA Blood mini kit (Qiagen). The remaining 0.5 ml of serum was used to isolate viral RNA using the QIAamp Viral RNA mini kit (Qiagen). Complementary DNA (cDNA) synthesis and diagnostic polymerase chain reaction (PCR) were performed as described (Santiago et al. 2003), targeting fragments in the SIVcpz *pol* (330 bp) and *gp41/nef* (740 bp) regions. Clarified serum

Table 3 Antibody tests run on Faustino blood samples (Simian Diagnostic Laboratory, Ltd., San Antonio, TX 78229, USA)

Pathogen	Reactive/nonreactive	Test type
Adenovirus, group-specific antibody	Reactive	Dot immunobinding assay (DIA)
Chimpanzee cytomegalovirus (CMV)	Reactive	DIA
Encephalomyocarditis (EMC)	Nonreactive	DIA
Epstein–Barr virus (EBV)	Reactive	DIA
Filovirus-Ebola-like (Reston)	Nonreactive	DIA
Hepatitis A	Nonreactive	Microparticle enzyme immunoassay
Hepatitis B	Nonreactive	Microparticle enzyme immunoassay
Hepatitis C	Nonreactive	Microparticle enzyme immunoassay
Herpes simplex-1 (HSV-1)	Reactive	DIA
Herpes simplex-2 (HSV-2)	Nonreactive	DIA
Herpesvirus-African monkey (SA 8)	Nonreactive	DIA
Human varicella-zoster (chicken pox)	Reactive	DIA
Influenza A	Reactive	DIA
Influenza B	Nonreactive	DIA
Lymphocytic choriomeningitis (LCM)	Nonreactive	DIA
Measles	Nonreactive	DIA
Mumps	Nonreactive	DIA
Parainfluenza type 1	Reactive	DIA
Parainfluenza type 2	Reactive	DIA
Parainfluenza type 3	Reactive	DIA
Respiratory syncytial virus (RSV)	Nonreactive	DIA
Rotavirus (SA 11)	Nonreactive	DIA
Simian immunodeficiency virus (SIV)	Nonreactive	Enzyme-linked immunosorbent assay (ELISA)

Table 4 Fecal sedimentation results for potentially pathogenic parasites (Lincoln Park Zoo Veterinary Laboratory, Chicago, IL 60614, USA)

Parasite	Date collected					
	7, Oct, 05	3, Nov, 05	27, Nov, 05	7, Dec, 05	24, Dec, 05	29, Jan, 06
Strongyloides sp.	Present	0	Present	0	0	0
Strongyle-type	Present	Present	Present	Present	Present	Present

was also probed for virus-specific antibodies using enhanced chemiluminescence Western blot analysis (Santiago et al. 2003). Furthermore, feces collected at the time of the intervention and preserved in RNAlater were tested using established methods (Santiago et al. 2003). Neither blood nor fecal analyses provided evidence that FO was SIVcpz positive at the time of intervention.

Fecal analyses

Fecal samples collected in 10 % neutral buffered formalin (Meridian Bioscience, Cleveland, OH, USA) before and after the incident as part of the ongoing health-monitoring project were analyzed for ova and parasites by sedimentation and floatation following standard methods (Gillespie 2006); Strongyle-type and *Strongyloides* spp. were present before, during, and after the illness (Table 4). Fecal samples for these dates were negative for the pathogenic protozoan parasites *Cryptosporidium* and *Giardia* when examined using immunofluorescent microscopy and pathogen-specific test kits (Salzer et al. 2007). Negative stain electron microscopy was also performed on a fecal sample obtained on the day of examination and was negative for viral particles.

DNA extracted from feces preserved in 10 % glycerin and blood preserved in RNAlater and dried on filter paper was tested for the presence of potentially pathogenic bacteria using a 16S ribosomal DNA (rDNA) PCR targeting highly conserved regions of the bacterial genome. The sequences obtained were heterogeneous as in healthy chimpanzees and did not show similarities to bacteria of known pathology. A specific PCR for *Bacillus anthracis* (anthrax) DNA was negative. Amplification products were obtained by PCR for adenovirus (Wellehan et al. 2004)

from two fecal samples, and sequences identical to each other and most closely related (94 % identical) to the DNA polymerase sequence of a chimpanzee adenovirus, simian adenovirus 25 (Davison et al. 2003) were found. Herpesvirus PCR (Ehlers et al. 2003) performed on whole blood identified DNA from *Pan troglodytes* lymphocryptovirus 1 (PtroLCV1), a member of the *Gammaherpesvirinae* subfamily with close similarity to Epstein–Barr virus (Ehlers et al. 2003). In addition, samples were tested for various enteroviruses following Harvala et al. (2011), but no positive signal was revealed by PCR.

Video assessment

Faustino's abnormal locomotion prior to the intervention was videotaped and sent to a veterinary neurologist for consultation. Based on visual assessment of movement and the rolling motion, it was suggested that the subject could move his limbs independently and maintained sensation in response to stimulation. These findings suggested that proprioception and voluntary motor function were intact and that the subject had muscular weakness rather than a primary neurological problem of the spinal cord. Traumatic injury was thought unlikely, as it would have resulted in limping or dragging of the affected area. Additionally, the rolling motion observed in Faustino would have put pressure on the injured area causing pain and would not have been a preferred mode of locomotion (W. Berry, personal communication).

Discussion

Laboratory results did not suggest any sign of systemic infection, as elevated white blood cell counts or reduced lymphocyte numbers were not present. Fecal samples obtained at the onset of clinical signs were negative for pathogenic protozoa, but positive for other potentially pathogenic nematode parasites such as Strongyle-type and *Strongyloides* spp. However, these parasites are commonly found at relatively high concentrations in the Gombe chimpanzees without causing such dramatic clinical signs (e.g., Gillespie et al. 2010; Terio et al. 2011). Fecal samples were also negative for pathogenic bacteria, anthrax, and various enteroviruses. Finally, consultation with a veterinary neurologist resulted in an assessment of generalized muscular atrophy, which may have been secondary to severe dehydration and extended anorexia. No visible external signs indicated trauma as a source, but because no radiographs could be taken, damage to the hips or spine could not be completely discounted. In an optimal diagnostic situation, radiographs and a muscle biopsy would have assisted in understanding the cause of the clinical signs. The eventual resolution of clinical signs argues against progressive conditions such as certain infectious diseases (e.g., poliovirus) and spinal trauma from which there would have been residual deficits in hindlimb function.

Since SIVcpz is endemic in Gombe, we considered the possibility that Faustino suffered from an acute retroviral syndrome. Acute HIV-1 infection in humans can be severe with newly infected individuals frequently seeking medical care with a variety of different symptoms, including fatigue, decreased appetite, general malaise, and even neurological problems (Cohen et al. 2011). Serum and fecal samples obtained at the time of intervention were negative for both SIVcpz-specific antibodies and nucleic acids by Western blot and nested PCR, respectively. Since high plasma viral load is a characteristic feature of acute HIV/SIV infection (Cohen et al. 2011), our failure to amplify SIVcpz sequences from a blood sample taken during FO's illness argues strongly against an acute viral syndrome. However, there are caveats, since subsequent noninvasive testing, which resumed 10 months after FO's recovery (12 August 2006), revealed that he had acquired SIVcpz (Keele et al. 2009); For example, suboptimal storage of the serum sample could have caused degradation of the viral RNA. There may also not have been sufficient cellular debris to detect proviral sequences. Finally, it is well known that there is a considerable time lag between HIV/SIV infection and the production of antibodies, which could explain the negative Western blot results (Cohen et al. 2011). Thus, we cannot exclude an acute retroviral syndrome with absolute certainty.

Despite a detailed veterinary assessment (albeit limited by field conditions) and a more complete testing of samples than had ever been undertaken for a Gombe chimpanzee, a firm diagnosis of the cause of Faustino's ailment remains elusive. Although seroreactive to several viruses (Table 3), none of these would have been expected to cause the noted clinical signs (Table 2). Nevertheless, this intervention did provide a rare opportunity to gather valuable information on potential pathogen exposure of Gombe chimpanzees. The test results must be interpreted with appropriate caution as some antibody titers to human viruses may have been due to cross-reaction with closely related chimpanzee virus. For example, seroreactivity to two alphaherpesviruses (herpes simplex virus and varicella-zoster virus) and one gammaherpesvirus (Epstein–Barr virus) could simply represent cross-reaction with chimpanzee herpesviruses (Ehlers et al. 2003; Luebcke et al. 2006) rather than exposure to these human pathogens. In fact, the DNA polymerase sequence of a close relative of Epstein–Barr virus, PtroLCV1 (Ehlers et al. 2003), was found in Faustino's blood, further supporting the possibility of cross-reactivity with a chimpanzee virus. The pathogenic potential of PtroLCV1 is not known, and therefore its potential role in the subject's disease

outcome cannot be assessed. Serum was also reactive against adenovirus, however the adenoviral DNA isolated from this chimpanzee was most closely related to simian adenovirus 25 (SAdV25) (Wevers et al. 2011). SAdV25, originally isolated from a latently infected chimpanzee (Basnight et al. 1971), and three other closely related chimpanzee adenoviruses (SAdV22–SAdV24) cluster with human adenoviruses of species E, which are mainly associated with infections of the respiratory tract and the eye (Schmitz et al. 1983). However, in humans the association of adenovirus with certain diseases is not exclusive, and these associations may be different in nonhuman primates. Thus, the importance of this finding, as either a primary or secondary pathogen, is uncertain.

Despite initial concerns, no conclusive evidence of anthropogenic disease was found in Faustino. As previously noted, although seroreactive for a number of potential human viruses, the assays are known to cross-react with closely related chimpanzee viruses. Given the available assays, the most that can be said regarding seroreactive test results is that Faustino had antibodies to a virus from a particular group. Antibodies against a number of influenza and parainfluenza viruses were noted, which may be the result of prior human exposure given the long history of respiratory outbreaks in the Gombe chimpanzees (Goodall 1983, 1986; Mlengeya 2000; Nutter 1996; Williams et al. 2008). Recent evidence for anthropozoonotic respiratory disease transmission between humans and wild chimpanzees in the Taï Forest (Köndgen et al. 2008) and between humans and wild mountain gorillas (Palacios et al. 2011) supports this hypothesis.

Recent research in African sanctuaries (Schaumberg et al. 2012) and in a captive facility in Japan (Kooriyama et al. 2013) has also documented human-associated pathogens in chimpanzees. In chimpanzee sanctuaries in Zambia and Uganda, Schaumberg et al. (2012) found a high prevalence of drug-resistant human-associated *Staphylococcus aureus*, and suggest that this may be due to treatment of individual apes with antibiotics or the close caregiver–chimpanzee contact required for rehabilitation of these animals. Kooriyama et al. (2013) recently reported the results from a serology survey of 14 captive socially housed chimpanzees in Japanese primate research centers and detected antibodies against 29 of 62 human pathogens. Like Faustino, these chimpanzees had test results that suggest previous exposure to parainfluenza viruses, Epstein–Barr virus, and varicella-zoster virus (but see discussion above regarding possible cross-reactivity with chimpanzee viruses). However, they also found antibodies against human respiratory syncytial virus (RSV), measles, hepatitis A, and rotavirus, which were not present in Faustino. Different laboratories were used for our investigation and that of Kooriyama et al. (2013), so this initial comparison should be interpreted with caution with regards to whether these similarities and differences truly reflect differing levels of human–chimpanzee contact. Our aim here is simply to demonstrate how little is still known about the pathogens circulating in captive and wild chimpanzees and emphasize how even a single data point (in this case, Faustino) provides an interesting comparative case study and basis for future studies.

We learned many lessons from this event that are likely to be helpful and relevant to other primatologists. At Gombe, as at many other wildlife research sites, veterinary intervention for acute disease outbreak investigation is fraught with both philosophical and logistical difficulties. First, in observational studies interested in life history patterns and fitness outcomes, veterinary intervention can distort what would otherwise be natural outcomes (Goodall 1986; Altmann 1987). Wild animals suffer disease and injury as a matter of course; the ability of individuals to withstand such insults, and the consequences of morbidity and mortality, are key parts of the data being collected in most long-term studies. Decisions to intervene may also be idiosyncratic, with intervention on the behalf of particular favored individuals, rather than being applied consistently across all individuals in the population. In this case, intervention was agreed upon due to the concern that Faustino may have contracted a disease of human origin that could threaten an entire small population of chimpanzees. Realizing that we did not have a standard rule for when/if to intervene was an important lesson. In cases where intervention is considered acceptable or necessary, there are additional concerns about the impacts of immobilization on individual animal safety and disruption to habituation (Travis et al. 2008). We did not suffer any adverse consequences in this case, given that Faustino was already largely immobile and very well habituated. In contrast, logistical issues provided very large additional challenges. Several procedures that would have been ideal, such as collection of cerebrospinal fluid, radiographs, and/or a muscle biopsy (as discussed above), were not possible. The capability to properly process, preserve, and store blood and tissue samples is critical, as is access to adequate health infrastructure and diagnostic laboratories (Leendertz et al. 2006). In this case, samples from Faustino's intervention were sent to six different laboratories and/or experts for testing, requiring a tremendous amount of human effort and logistical organization.

As a result of the lessons we learned, we suggest that the ape conservation and research community at large should support the development of contingency plans to assist with both the decision-making process as well as logistical preparation to maximize chances of success if/when outbreaks occur and interventions are necessary. Currently, the Mountain Gorilla Veterinary Project (MGVP) is the only group which commonly intervenes and immobilizes

wild great apes, and demographic analyses suggest that these interventions may be responsible for increased population growth rates in groups that receive interventions versus those that do not (Robbins et al. 2011). The MGVP has spent twenty-five years discussing and refining answers to these questions (Decision Tree Writing Group, 2006), but nothing similar exists for chimpanzees, which likely present different challenges for immobilization given, among other things, their more arboreal nature. For now, we make a minimum number of recommendations for ape conservation/research sites:

1. Establish guidelines regarding if/when and under what conditions intervention would be acceptable. This should be agreed upon by the appropriate governing officials and be embedded in operational documents to reduce the timeline for gaining approval from national wildlife managers.
2. Assess local expertise available for assistance in the face of an outbreak. This includes knowledge of local veterinary expertise or formulation of partnerships with those closest and/or most prepared to help; for this case, experts from the Ngamba Island Chimpanzee Sanctuary assisted. It is also critical to understand local laboratory resources and capabilities.
3. Create a contingency plan for the decision-making process, the logistics of the work/procedures, preservation and storage of samples, shipment and diagnostic evaluation of samples, financial responsibility, actions to be taken upon receipt of results, and communication of important information to all pertinent stakeholders.
4. Procure the necessary equipment (or know where it can be found) and training of personnel to implement the investigation/intervention plan.
5. Develop and implement training plans and procedures (including strict biosafety protocols) for conducting necropsies of deceased animals when possible to further investigate causes of mortality (e.g., Terio et al. 2011).

Without such guidelines and procedures, opportunities for diagnostic testing may be missed due to lack of appropriate collection, preservation, and storage capabilities. Ideally, such plans and experiences will be published and shared via other mechanisms throughout the conservation community so that lessons can be learned and strategies and procedures can be refined to better protect the health of wild primates.

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