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Authors: Good, Kyle M., Houser, AnnMarie, Arntzen, Lorraine, and Turnbull, Peter C. B.

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Naturally Acquired Anthrax Antibodies in a Cheetah (*Acinonyx jubatus*) in Botswana

Kyle M. Good,[1,5] AnnMarie Houser,[2] Lorraine Arntzen,[3] and Peter C. B. Turnbull[4] [1]Cheetah Conservation Botswana, Private Bag 0457, Gaborone, Botswana; [2]Cheetah Conservation Botswana, Jwana Game Reserve, Jwaneng, Botswana; [3]National Institute of Communicable Diseases, Sandringham, South Africa; [4]Arjemptur Technology Ltd., Science Park, Porton Down SP4 0JQ, United Kingdom; [5]Corresponding author (email: kmgood@accelerate-it.co.bw)

ABSTRACT: An outbreak of anthrax in the Jwana Game Reserve in Jwaneng, Botswana, was first observed when three cheetahs (*Acinonyx jubatus*) died of the disease in November 2004. In the aftermath of this event, banked serum samples collected from 23 wild-caught cheetahs were examined, by the inhibition enzyme-linked immunoassay (ELISA), for antibodies to the protective antigen (PA) of *Bacillus anthracis*. Of the 23 cheetahs, 16 regularly accessed the reserve. Antibodies to PA were detected in one cheetah collected in May 2004, indicating the disease was occurring well before it was first noticed. This appears to be the first demonstration of naturally acquired anthrax antibodies in cheetahs. The finding of one antibody-positive animal amongst at least 16 potentially exposed individuals is consistent with existing reports that it is uncommon for cheetahs to develop natural immunity to anthrax.

Key words: Anthrax, antibody, cheetah, immunity, protective antigen.

In November 2004, attention was drawn to an outbreak of anthrax in a 20,000-ha game reserve, Jwana Game Reserve located in Jwaneng, Botswana (central GPS; 24°43′21.53″E, 24°31′51.89″S), when three captive cheetahs (*Acinonyx jubatus*) died of the disease (Good et al., 2005). The three animals had been fed meat from a dead red hartebeest (*Alcelaphus buselaphus*) from the reserve; anthrax was subsequently confirmed in the red hartebeest. Retrospectively, it was determined that earlier cases had occurred in a red hartebeest and a wildebeest (*Connochaetes taurinus*) found in September 2004, and in another red hartebeest in October 2004. The anthrax outbreak in the reserve peaked between January and March 2005; by the end of March, confirmed or suspected anthrax deaths occurred in 44 zebra (*Equus burchellii*), 19 red hartebeest, eight springbok (*Antidorcas marsupialis*), four wildebeest, and three eland (*Tragelaphus oryx*). Cases ended in June, only to be followed by a resurgence over the next 6 mo involving an observed 20 wildebeest, 17 red hartebeest, five zebra, and two springbok. Sporadic cases occurred every month from January to May 2007; eight eland, six zebra, three wildebeest, one springbok, one giraffe (*Giraffa camelopardalis*), and one polecat (*Ictonyx striatus*) succumbed. In all, 142 affected animals were observed during this period. Previous anthrax records in the Botswana National Veterinary Laboratory annual reports include an outbreak affecting 42 goats in Jwaneng in 1985 and cases (numbers not specified) in cattle in this district in 1990; smaller numbers of cattle cases have occurred in neighboring districts of southern Botswana in the years 1985–87, 1990, and 1997–99. There were no prior records of the disease in wildlife in this area.

Due to the remoteness of the Jwana Game Reserve in relation to veterinary facilities, confirmation of anthrax as the cause of death, by polychrome methylene blue-stained smears or by using the PA detecting hand-held immunochromatographic (Burans et al., 1996; Tubbesing, 1997; Muller et al., 2004), could only be done in 17 (12%) of the cases; the remainder had to be recorded as suspected cases, based on the typical symptoms of sudden death with bleeding from orifices. On this basis, a further 129 deaths were not suspected to result from anthrax. Included in these 129 deaths were five ostriches (*Stuthio camelus*), 14 white-

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backed vultures (*Gyps africanus*), and a chacma baboon (*Papio ursinus*). Anthrax has been recorded previously in ostriches (Ebedes, 1976; Hugh-Jones and de Vos, 2002). There is at least one anecdotal report, in a vulture, of death from this disease (Turnbull et al., 2008), although in the cases here, 12 of the 14 vultures found dead were together at a water hole, and poisoning was suspected.

The three cheetah deaths stimulated questions related to the potential impact of anthrax on cheetahs. The reserve has a wild population of free-roaming cheetahs that access the reserve through warthog (*Phacochoerus africanus)* holes. Banked sera from 23 wild-caught cheetahs were examined by inhibition enzyme-linked immunoassay (ELISA), described elsewhere (Turnbull et al., 2004), for antibodies to the protective antigen (PA) of *Bacillus anthracis.* Twelve of the sera had been collected between March and September 2004, before the outbreak had become apparent; the remainder of the sera were collected between June and October 2005. Sixteen of the 23 cheetahs regularly frequented the reserve, and the serum from one of these, a male in a group of three collected in May 2004, had measurable antibodies to the anthrax antigen, indicating that anthrax was occurring in his home range at least several months before the outbreak in the reserve was recognized. To our knowledge, this is the first report of naturally acquired anthrax antibodies in cheetahs, although their ability to develop vaccine-induced antibodies to PA, in a manner similar to other species, has been demonstrated and shown to be paralleled by protective immunity (Turnbull et al., 2004). Lions in anthrax enzootic areas have been shown to develop these antibodies, and circumstantial evidence suggests that this confers a protective immunity (Turnbull et al., 1992).

Among wild carnivores, cheetahs appear to be unusually susceptible to anthrax, and this has been attributed to their lack of scavenging behavior and their consequential lack of opportunity to build up immunity through exposure to anthrax carcasses (Ewer, 1973; Lindeque et al., 1998). An interesting observation, during the events reported here, was that sightings, and spoor evidence of cheetahs frequenting the reserve, decreased during the peak of the outbreak; this may indicate decreased exposure to the bacteria (A.M. Houser, unpubl. data). These considerations are commensurate with the finding of just one serologically positive animal amongst at least 16 potentially exposed animals.

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