Transactions of the Royal Society of Tropical Medicine and Hygiene (2007) **101**, 64—78

available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevierhealth.com/journals/trst

Morbidity and mortality of wild animals in relation to outbreaks of Ebola haemorrhagic fever in Gabon, 1994—2003

Sally A. Lahm[a,*], **Maryvonne Kombila**[b], **Robert Swanepoel**[c], **Richard F.W. Barnes**[d,e]

[a] *Institute for Research in Tropical Ecology, Makokou, Gabon*
[b] *Department of Tropical Medicine and Parasitology, University of Health Sciences, Libreville, Gabon*
[c] *National Institute for Communicable Diseases, Sandringham, South Africa*
[d] *Division of Biological Sciences 0116, Ecology, Behavior and Evolution Section, University of California, San Diego, CA, USA*
[e] *Environmental Sciences Research Centre, Anglia Ruskin University, East Road, Cambridge CB1 1PT, UK*

Received 15 November 2005; received in revised form 1 July 2006; accepted 4 July 2006
Available online 28 September 2006

KEYWORDS
Ebola haemorrhagic fever;
Filoviridae;
Wild animals;
Veterinary;
Morbidity;
Mortality;
Gabon

Summary Antibody to Ebola virus was found in 14 (1.2%) of 1147 human sera collected in Gabon in 1981—1997. Six seropositive subjects were bled in the northeast in 1991, more than 3 years prior to recognition of the first known outbreak of Ebola haemorrhagic fever (EHF), whilst eight came from the southwest where the disease has not been recognised. It has been reported elsewhere that 98 carcasses of wild animals were found in systematic studies in northeastern Gabon and adjoining northwestern Republic of the Congo (RoC) during five EHF epidemics in August 2001 to June 2003, with Ebola virus infection being confirmed in 14 carcasses. During the present opportunistic observations, reports were investigated of a further 397 carcasses, mainly gorillas, chimpanzees, mandrills and bush pigs, found by rural residents in 35 incidents in Gabon and RoC during 1994—2003. Sixteen incidents had temporal and/or spatial coincidence with confirmed EHF outbreaks, and the remaining 19 appeared to represent extension of disease from such sites. There appeared to be sustained Ebola virus activity in the northeast in 1994—1999, with sequential spread from 1996 onwards, first westwards, then southerly, and then northeastwards, reaching the Gabon—RoC border in 2001. This implies that there was transmission of infection between wild mammals, but the species involved are highly susceptible and unlikely to be natural hosts of the virus.

* Corresponding author. Present address: Division of Biological Sciences 0116, Ecology, Behavior and Evolution Section, University of California, San Diego, 9500 Gilman Drive, La Jolla, CA 92023, USA. Tel.: +1 619 287 1375; fax: +1 619 287 7611.
E-mail address: sallyalahm@aol.com (S.A. Lahm).

0035-9203/$ — see front matter
doi:10.1016/j.trstmh.2006.07.002

1. Introduction

The filoviruses include Marburg virus and three serotypes of Ebola virus (Ebola-Zaire, Ebola-Sudan and Ebola-Ivory Coast) that cause outbreaks of haemorrhagic fever among humans in Africa, plus Ebola-Reston from the Philippines that affects monkeys but appears to be non-pathogenic for humans (Peters et al., 1996). Three species of bats were recently identified as potential reservoirs of Ebola-Zaire (Leroy et al., 2005), but the natural hosts of the other viruses remain unknown.

Marburg virus was discovered in 1967 when laboratory workers in Germany and Yugoslavia acquired fatal illness from contact with the tissues of imported monkeys, many of which were sick or dead on arrival from Uganda (Siegert et al., 1967). Attempts to find evidence of infection in non-human primates in Africa were inconclusive, partly because existing serological techniques lacked sensitivity and specificity. Since the animals appeared to be highly susceptible to infection, it was concluded that they were unlikely to be capable of serving as reservoir hosts responsible for perpetuation of the virus in nature (Malherbe and Strickland-Cholmley, 1971; Simpson et al., 1968; Slenczka et al., 1971). Small outbreaks of Marburg haemorrhagic fever were subsequently observed in South Africa in 1975, with infection apparently originating in Zimbabwe, and in Kenya in 1980 and 1987, whilst large epidemics were recorded in the northeastern Democratic Republic of the Congo (DRC) in 1998—2000 and in Angola in 2004—2005 (Gear et al., 1975; Johnson et al., 1996; Smith et al., 1982; WHO, 1999, 2005). Although various species, including bats, were investigated as potential reservoir hosts of the virus during the outbreaks in Africa, no disease or deaths were noted in domestic or wild animals subsequent to the original episodes in Europe (Conrad et al., 1978).

Ebola virus was discovered in 1976 when simultaneous outbreaks of haemorrhagic fever occurred in northern DRC and southern Sudan, which were found to be caused by two different serotypes of a new virus, designated Ebola-Zaire and Ebola-Sudan (Cox et al., 1983; McCormick et al., 1983; Richman et al., 1983). A single case of Ebola-Zaire infection was recognised in DRC in 1977, and an outbreak of Ebola-Sudan infection recurred in southern Sudan in 1979 (Baron et al., 1983; Heymann et al., 1980). No deaths of livestock or wild animals were observed during these four initial outbreaks of Ebola haemorrhagic fever (EHF), but the position changed when the disease re-appeared in Africa in November 1994 after an interval of 15 years, when a new serotype of the virus designated Ebola-Ivory Coast was isolated from a biologist who became infected during an autopsy on a chimpanzee in which the infection was confirmed by immunohistochemistry (Le Guenno et al., 1995). The virus is believed to have caused epidemics in the chimpanzee community that had been under observation for 14 years (Formenty et al., 1999).

During the same month (November 1994), there was an outbreak of EHF in gold mining camps in the Minkébé region of northeastern Gabon, where residents noted the simultaneous occurrence of deaths among gorillas, monkeys and bush pigs (red river hogs) in the surrounding forest. This proved to be the first of three epidemics to occur in Gabon from 1994—1997, each associated with deaths of wild animals and each caused by a genetically distinct lineage of Ebola-Zaire virus (Georges et al., 1999; Georges-Courbot et al., 1997). After a 4-year interval during which no virus activity was detected, a further six epidemics occurred in Gabon and neighbouring northwestern Republic of the Congo (RoC) in 2001—2005. The first four of these six epidemics involved multiple incidents of humans acquiring infection from contact with wild animal carcasses and were associated with multiple genetic lineages of Ebola-Zaire virus (Leroy et al., 2004a; Rouquet et al., 2005). Meanwhile, there had been a large outbreak of EHF associated with Ebola-Zaire virus in the DRC in 1995, as well as epidemics caused by Ebola-Sudan virus in Uganda in 2000—2001 and in Sudan in 2004 (WHO, 2001, 2004).

The senior investigator (S.A.L.), who was conducting unrelated ecological studies in Gabon, was prompted by these outbreaks to make observations on morbidity and mortality in wild animals in Gabon and RoC from 1994 to 2003. The opportunity was also taken to perform tests for antibody to Ebola virus on human sera that had been collected earlier for an unrelated survey. The findings of that study are presented here and suggest that Ebola virus activity has been more widespread and sustained in Gabon than previously suspected, and provide an indication of the manner in which the infection may have spread.

2. Materials and methods

2.1. Study area

Gabon straddles the equator within the Guineo-Congolian phytogeographic region (White, 1983). The classic dense evergreen forest characteristic decreases with distance from the Atlantic coast towards the northeastern mixed evergreen semi-deciduous forest (Caballé, 1978). Approximately 15% of the habitat is savannah. Annual rainfall varies from a maximum of 3200 mm in the northwest to 1400—1600 mm in southern savannah regions. The three climatic regimes described are: (1) northeastern equatorial climate of two wet and two dry seasons; (2) central transitional tropical climate with a 3-month dry season and a 9-month variable wet season; and (3) southern tropical climate of 4—5 dry months and a 7—8-month wet season (Richard and Leonard, 1993).

2.2. Human serosurvey

From 1981—1997, 3531 serum samples were obtained by the Department of Tropical Medicine and Parasitology, University of Health Sciences, Libreville, Gabon, with informed consent from people in six rural communities in northeastern, southeastern and western Gabon for onchocerciasis research. The sera were stored at −70 °C. In 1998, 1147 (32.5%) of the samples were tested at the National Institute for Communicable Diseases (NICD), South Africa, for anti-Ebola IgG and IgM antibody using an ELISA with Ebola-Zaire virus antigen, as described previously (Ksiazek et al., 1999). In the absence of a panel of sera derived from known Ebola-infected individuals for proper validation and standardisation of the ELISA, the cut-off optical density (OD) value used for interpretation of results was based on