Lassa Fever is a Zoonosis Caused by Lassa Virus in Multimammate Rats, Côte d’Ivoire, 2013


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References


To the Editor: Lassa fever is a zoonosis caused by Lassa virus (LASV; family Arenaviridae, genus Lassavirus). The primary reservoir of LASV is the multimammate rat (Mastomys natalensis), which is found throughout sub-Saharan Africa. LASV outbreaks among humans occur only in West Africa in 2 noncontiguous areas: 1 in Guinea, Liberia, and Sierra Leone; and 1 in Nigeria. Rare cases and evidence of exposure of humans have been documented in neighboring countries (i.e., Benin, Burkina Faso, Côte d’Ivoire, Ghana, Mali, and Togo) (1). LASV RNA has been detected in only 4 patients: 1 in Germany who had traveled in Burkina Faso, Côte d’Ivoire, and Ghana (2); 1 in the United Kingdom who had returned from Mali (3); and 2 in
Ghana, for whom no viral sequence was available because detection was performed by reverse transcription PCR only (4). In the region in Mali where the patient from the United Kingdom was infected, identical LASV sequences were found in multimammate rats (5). The sequence of the strain identified from the patient in Germany, who was designated AV, is the closest known relative of the clade formed by sequences from Mali (5). However, LASV was not found in its natural host in any of the countries visited by patient AV (6,7).

For a study investigating zoonotic pathogens in rural habitats, we caught small mammals in 3 ecologic zones of Côte d’Ivoire: 1) dry bushland in northern Côte d’Ivoire, around Korhogo (2); semiarid bushland in central Côte d’Ivoire, around Bouake; and rainforest in southwestern Côte d’Ivoire, near the Taï National Park (3) (online Technical Appendix Figure, http://wwwnc.cdc.gov/EID/article/21/8/15-0312-Techapp.pdf). Traps were installed within and around 15 villages and enabled the capture of 27 eulipotyphlans and 254 rodents during August–October 2013. Animals were assigned at the genus level in the field on the basis of morphology. For 88% of them, assignment could later be refined to the species level by sequencing a fragment of the mitochondrial cytochrome b gene. A total of 14 animal species representing 8 genera were detected. All host sequences were deposited in Dryad (http://www.data dryad.org/; online Technical Appendix Table 1). Multimammate rats were the dominant commensals at all sampling locations, comprising 64.5% of the overall sample (online Technical Appendix Figure).

Tissue samples were collected from all animals according to standard protocols. Total nucleic acids were extracted from lung samples and tested for the presence of LASV RNA by using a real-time PCR system amplifying a 400-bp fragment of the large genomic segment (8) (online Technical Appendix). LASV RNA was detected in 4 of 18 specimens of M. natalensis captured in Gbalôhô, near Korhogo (online Technical Appendix Figure). This site is much farther north in Côte d’Ivoire than previously examined sites (6). The 4 PCR-positive animals were 3 males and 1 female that were all captured indoors, 3 in the same house. PCR products were sequenced according to the Sanger method (GenBank accession nos. LN823982–LN823985). According to phylogenetic analyses performed in maximum likelihood and Bayesian frameworks (online Technical Appendix), LASV sequences identified in multimammate rats from Côte d’Ivoire formed a robust clade with sequences from the human AV strain and the LASV infecting multimammate rats in southern Mali (bootstrap 97, posterior probability 1.00; Figure). This phylogenetic placement opens up the possibility that patient AV was infected during her travel through Côte d’Ivoire, possibly in or near the city of Korhogo. Tip date calibration of Bayesian analyses showed that the most recent common ancestor of all LASV sequences from Côte d’Ivoire and Mali circulated ≈90 years ago (Figure; online Technical Appendix Table 2).

Further studies will be needed to investigate the geographic distribution of LASV in Côte d’Ivoire and the frequency of human infections. The current lack of diagnosed cases in the area may be caused by underreporting. Sensitization campaigns are needed to increase awareness of the risk for LASV infection among the local population and to improve detection of cases by health workers.

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**Rickettsia felis** Infection among Humans, Bangladesh, 2012–2013


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To the Editor: *Rickettsia felis*, which belongs to the spotted fever group of rickettsiae, causes febrile illness in humans. The main vector of this bacterium is the cat flea (*Ctenocephalides felis*). Since publication of reports of *R. felis* as a putative pathogen of humans in the United States in 1994, *R. felis* infection in humans worldwide has been increasingly described, especially in the Americas, Europe, Africa, and eastern Asia [1,2]. *R. felis* infection is common among febrile patients (≈15%) in tropical Africa (3) and among apparently healthy persons in eastern coastal provinces of China (4). However, little is known about prevalence of *R. felis* infection of humans in southern Asia, although 3 serologically diagnosed cases in Sri Lanka have been described (5) and *R. felis* has been detected in rodent fleas in Afghanistan (6). Hence, we conducted a cross-sectional study in Bangladesh to explore the presence of rickettsial pathogens among patients with fever of unknown origin.

Study participants were 150 patients at Mymensingh Medical College (MMC) hospital in Mymensingh, north-central Bangladesh, from July 2012 through January 2014, and 30 healthy control participants from the staff at the same college. Selected patients met the following criteria: 1) fever (axillary temperature >37.5°C) for >15 days that did not respond to common antimicrobial drug therapy; 2) any additional clinical features including headache, rash, lymphadenopathy, myalgia, and eschars on skin; and 3) titre according to the Weil-Felix test (antibodies against any of 3 *Proteus* antigens) of >1:80. Patients with evident cause of fever (e.g., malaria diagnosed by blood smear or immunochromatography) were excluded from the study. This research was approved by the college institutional review board, and informed consent was obtained from patients (or guardians) and healthy controls before their entry into the study.

Venous blood samples were aseptically collected from the patients, and DNA was extracted by conventional method by using proteinase K and sodium dodecyl sulfate. Nested PCR selective for the 17-kDa antigen gene was used to screen for rickettsiae according to the method described previously (7); ≈100 ng of DNA in a 50-μL reaction mixture was used. For each PCR, a negative control (water) was included and utmost care was taken to avoid contamination. Among the 150 samples tested, results were positive with a 232-bp amplified product for 69 (46%) and negative for all controls.

PCR products from 20 samples were randomly selected for sequence analysis. All nucleotide sequences from