

2008 showed 6 aa substitutions in the envelope gene: V129I, L131Q, I170T, E203D, M340T, and I380V. Our results support the notion that aa positions at 129 and 131 in the envelope gene are critical genetic markers for phylogenetic classification of DENV-2 (7–9).

Notably, residue 131 in the envelope gene is located within a pH-dependent hinge region at the interface between domains I and II of the envelope protein. Mutations at this region may affect the pH threshold of fusion and the process of conformational changes (10).

Our results suggest the circulation of genetically different DENV-2 in Brazil and that these viruses may have a role in severity of dengue diseases. These findings can help to further understand the complex dynamic pathogenic profile of dengue viruses and their circulation in dengue-endemic regions.

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Yersinia Species Isolated from Bats, Germany

To the Editor: Bats are distributed worldwide and are among the most diverse and species-rich mammals on earth. They exist in a large variety of distinct ecologic niches. Many bat species roost near humans, which is of particular interest for research on bat-to-human transmission of potential zoonotic pathogens. Moreover, migratory bats could act as long-distance vectors for several infectious agents. In recent decades, scientific interest in chiropteran species has markedly increased because bats are known hosts to zoonotic agents, such as henipaviruses, Ebola virus, and severe acute respiratory syndrome (SARS)-like corona viruses (1,2). However, investigations regarding bacterial pathogens with potential for mutual transmission between bats and humans are sparse. The effect of bacterial agents on individual bats is largely unknown and has been neglected in most studies published to date (3).

We conducted a broad study during 2006–2008 on diseases and causes of death in bats among 16 species found in Germany. Two hundred deceased bats, collected in different geographic regions in Germany (southern Bavaria, eastern Lower Saxony, eastern Brandenburg, and Berlin greater metropolitan area), were subjected to necropsy and investigated by using routine histopathologic and bacteriologic methods. During necropsy, instruments were dipped in 70% ethanol and moved into a Bunsen burner flame after every incision to prevent any cross-contamination. For bacteriologic examination, tissue samples were treated accordingly to prevent environmental contamination. A freshly cut tissue surface was plated onto Columbia agar (5% sheep blood; Oxoid, Wesel, Germany), Gassner agar (Oxoid), and MacCo-

nkey agar (Oxoid) and incubated at 37°C for 24–48 hours.

Twenty-five bacterial genera were cultured from bats, including 2 known human-pathogenic *Yersinia* spp., i.e., *Y. pseudotuberculosis* and *Y. enterocolitica*. The first *Yersinia* strain (Y938) was cultured from lung, heart, kidney (pure cultures), liver, spleen, and intestine (mixed cultures) of a greater mouse-eared bat (*Myotis myotis*). This isolate was identified as *Y. pseudotuberculosis* by Api 20E (bioMérieux, Nürtingen, Germany), Micronaut-E (Merlin Diagnostik GmbH, Bornheim-Hersel, Germany), and 16S rRNA gene analysis (Table). The sequence was deposited into GenBank under accession no. FN561631. Further serologic characterization by agglutination test (Denka Seiken, Tokyo, Japan) and multilocus sequence typing (4) identified *Y. pseudotuberculosis* serogroup 1, biovar 5, sequence type (ST) 43 in all tissue samples investigated. During necropsy, severe enlargement of the liver and a marked hemoperitoneum were seen. Microscopic examination showed multifocal severe necrotizing hepatitis and splenitis associated with numerous intralosomal gram-negative coccobacilli and a moderate to marked interstitial pneumonia. The remaining organs, including heart, kidney, and intestine, had no pathologic changes.

The second *Yersinia* strain (Y935), *Y. enterocolitica*, was isolated in pure culture from spleen and intes-

tine of a common pipistrelle (*Pipistrellus pipistrellus*) and identified by the methods described above (Table). The 16S rRNA sequence was deposited into GenBank under accession no. FN561632. No bacteria were cultured from any other organ. Based on results of an agglutination test (Denka Seiken), the isolate was characterized as *Y. enterocolitica* serotype O:6, biovar 1A. Necropsy and histopathologic examination showed no inflammatory changes, suggesting a subclinical state of infection.

Yersiniosis is a bacterial disease with a wide distribution and host range. *Y. pseudotuberculosis* and *Y. enterocolitica* are frequently isolated from a variety of wild and domestic animals (5), but little is known about the occurrence of yersiniosis in free-ranging chiropteran species. Only few reports of fatal *Y. pseudotuberculosis* infection in captive flying foxes have been published (6,7). In Europe, *Y. pseudotuberculosis* strains belonging to serogroup 1 are most common and cause most *Y. pseudotuberculosis* infections in humans and animals (5). Isolates of ST43 in the multilocus sequence typing database (4) came from humans, birds, hares, hedgehog, cat, dog, and pig in Europe; humans in Asia; marsupial in Australia; and deer in Australia and New Zealand. We report an isolate from a free-ranging bat in Germany. *Y. enterocolitica* biovar 1A has been found in a wide range

of human, animal, and environmental sources. Although often considered nonpathogenic, this biovar is described as an opportunistic pathogen (8), and serovar O:6 has been detected as the causative agent of ovine placentitis and abortion (9).

Transmission of both *Yersinia* species generally occurs after ingestion of contaminated food or water. All bat species in Germany are insectivorous, and insects can be infected with various microbial agents. Investigations concerning bacteria–insect interactions showed that insects may carry pathogenic bacteria, including *Yersinia* (10); thus, insects or contaminated water are possible sources of both species described.

In conclusion, *Y. pseudotuberculosis* and *Y. enterocolitica* were isolated from 2 bat species in Germany, representing evidence of *Yersinia* spp. in free-ranging vespertilionids. Histopathologic findings of the greater mouse-eared bat were consistent with those of systemic *Y. pseudotuberculosis* infection, rendering this species pathogenic for bats. The common pipistrelle was subclinically infected with *Y. enterocolitica*. The role of wild animals as reservoir hosts for bacterial pathogens such as *Yersinia* spp. is well known, underlining the need for biologists and persons handling wildlife to be aware of these zoonotic infectious agents.

Table. Strain typing results of *Yersinia* spp. isolates from free-ranging bats, Germany*

Method	<i>Y. pseudotuberculosis</i> †	<i>Y. enterocolitica</i> ‡
Api 20E profile§	1 0 1 4 1 1 2 1 7	1 1 5 4 7 2 3 1 7
Micronaut E,¶ % probability	100% <i>Y. pseudotuberculosis</i>	99.77% <i>Y. enterocolitica</i>
16S rRNA gene analysis, % similarity to sequences in GenBank	100% (1,058 bp) to <i>Y. pseudotuberculosis</i> YPIII#	100% (985 bp) to <i>Y. enterocolitica</i> strain 280**
Serologic characterization by agglutination test	Serogroup 1, biovar 5	Serotype O:6, biovar 1A
Multilocus sequence typing††	1 2 1 2 5 2 1	Not determined

**Y. pseudotuberculosis* obtained from lung, heart, kidney, spleen, liver, and intestine; *Y. enterocolitica* obtained from spleen and intestine. For *Y. pseudotuberculosis* spleen, liver, and intestine samples, isolates were obtained in mixed culture accompanied by colonies of 1–2 nonspecific bacteria. For all remaining tissues, *Y. pseudotuberculosis* and *Y. enterocolitica* were isolated in pure culture.

†GenBank accession no. FN561631.

‡GenBank accession no. FN561632.

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¶Merlin Diagnostik GmbH, Bornheim-Hersel, Germany.

#GenBank accession no. CP000950.

**GenBank accession no. FJ641888.

††Of gene loci *adk*, *argA*, *aroA*, *glnA*, *thrA*, *tmk*, *trpE*.

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Letters

Letters commenting on recent articles as well as letters reporting cases, outbreaks, or original research are welcome. Letters commenting on articles should contain no more than 300 words and 5 references; they are more likely to be published if submitted within 4 weeks of the original article's publication. Letters reporting cases, outbreaks, or original research should contain no more than 800 words and 10 references. They may have 1 Figure or Table and should not be divided into sections. All letters should contain material not previously published and include a word count.

Human Herpesvirus 8, Southern Siberia

To the Editor: Human herpesvirus 8 (HHV-8) is the etiologic agent of Kaposi sarcoma. Sequence analysis of the highly variable open reading frame (ORF)–K1 of HHV-8 has enabled the identification of 5 main molecular subtypes, A–E (*1*). A and C subtypes are prevalent in persons in Europe, Mediterranean countries, northwestern China, and the United States; subtype B, in persons in sub-Saharan Africa; subtype D, in persons in the Pacific Islands and Japan (*2–6*); and subtype E, in Native Americans in the United States.

Considering that K1 gene polymorphisms of HHV-8–infected persons reflect the divergence accumulated during the early migrations of modern humans out of Africa (*1*), it is tempting to put the polymorphisms observed in the different subtypes into an evolutionary perspective with their geographic distribution. It is thought that Native Americans infected by subtype E and Pacific Islanders, including those infected by subtype D in the Japanese archipelago, originated from a common ancestral genetic stock in continental Asia. Because Siberia constitutes the geographic link between mainland Asia, North America, and the Pacific (online Technical Appendix, www.cdc.gov/eid/content/16/3/585-Techapp.pdf), it is likely that the Siberian region has served as a source or a corridor of human dispersals to these regions. Thus, we conducted a molecular epidemiology HHV-8 survey of the Buryat population, a major indigenous group in southern Siberia, to gain new insights into the origins, possibly common, of HHV-8 subtypes D and E.

After consent of local authorities and participants, we collected 745 human blood samples in 1995 in 17 medicosocial structures (homes for elderly persons, veterans of the Russian army,