A226V Strains of Chikungunya Virus, Réunion Island, 2010

Eric D'ortenzio, Marc Grandadam, Elsa Balleydier, Marie-Christine Jaffar-Bandjee, Alain Michault, Elise Brottet, Marie Baville, Laurent Filleul

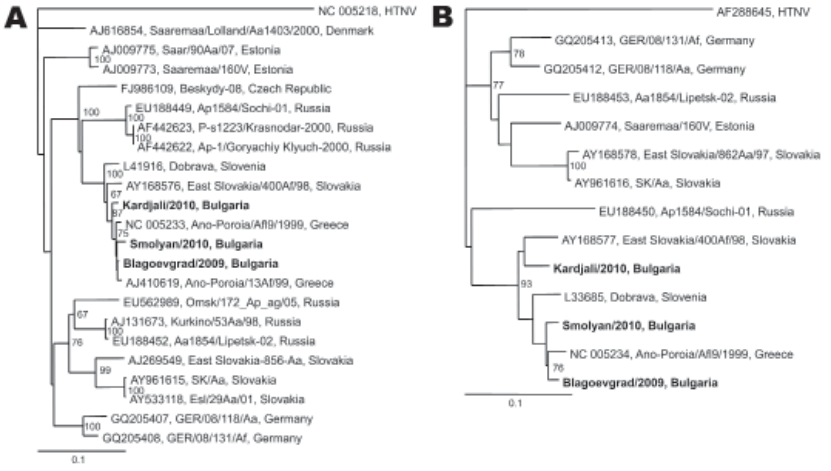

Figure. Phylogenetic trees based on a 560-bp fragment of the small RNA segment (A) and a 224-bp fragment of the medium RNA segment (B) of hantaviruses. Hantaan virus (HTNV) was used as the outgroup. The numbers at the nodes indicate percentage bootstrap replicates of 100; values <60% are not shown. Horizontal distances are proportional to the nucleotide differences. Sequences in the tree are indicated as GenBank accession number, strain name, country. Strains from this study are shown in **boldface**. Scale bars indicate 10% nucleotide sequence divergence.

of febrile cases accompanied by acute nephropathy. Further studies on patients and small mammals in Bulgaria will elucidate the hantavirus epidemiology in this Balkan region.

Acknowledgments

We thank N. Kalvatchev and E. Papadimitriou for excellent technical assistance.

Anna Papa and Iva Christova

Author affiliations: Medical School, Aristotle University of Thessaloniki, Thessaloniki, Greece (A. Papa); and National Centre of Infectious and Parasitic Diseases, Sofia, Bulgaria (I. Christova)

DOI: 10.3201/eid1702.101275

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Address for correspondence: Anna Papa, Department of Microbiology, Medical School Aristotle University of Thessaloniki, 54124, Thessaloniki 999149, Greece; email: annap@med.auth.gr

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To the Editor: Chikungunya virus (CHIKV) first emerged in Indian Ocean islands off the eastern coast of Africa in 2005 and was responsible for large-scale epidemics on the islands of Réunion, Comoros, Mayotte, Mauritius, Madagascar, and Seychelles (*1–4*). On Réunion Island, a French overseas territory of 810,000 inhabitants, herd immunity reached 38% in October 2006 (*5*). Molecular epidemiology of the strain responsible for these outbreaks indicated that it had originated in Kenya (*6*). The epidemic on Réunion Island was associated with a mutation in the envelope protein gene (E1-A226V) that improves replication and transmission efficiency in *Aedes albopictus* mosquitoes (*7*).

Since 2006, the Regional Office of the French Institute for Public Health Surveillance in the Indian Ocean has conducted epidemiologic and biological surveillance for CHIKV infection. Case definitions have been described (*8*). During December 2006–July 2009, no confirmed case was detected

on Réunion Island and Mayotte, but new outbreaks were reported in Madagascar (*9*). In August 2009, a cluster of cases was identified on the western coast of Réunion Island (*8*).

We report an outbreak of CHIKV infection that occurred on Réunion Island in 2010. The first case was detected on March 17, 2010. As of July 6, a total of 100 confirmed and 32 probable cases had been identified (online Appendix Figure, www.cdc.gov/EID/content/17/2/307-appF.htm). Median age of case-patients was 39 years (range 6 months–80 years), and the ratio of male to female case-patients was 0.81:1. In addition to fever (95%), case-patients had arthralgia (95%), headache (78%), and myalgia (75%). Seven (5%) were admitted to hospitals. No severe illness or death was reported. The outbreak remained largely restricted to residents of Saint Paul (75%) on the western coast. Sporadic cases in other cities also were detected.

Sequence comparison based on partial envelope gene or complete genome showed a high level (>99.6%) of nucleotide and amino acid identity of 2010 isolates from Réunion Island with the strains of the 2009 sporadic cases on Réunion Island, as well as with the Malagasy strains circulating since 2006. All isolates sequenced bore the A226V substitution within the E1 protein. Altogether, these results support the hypothesis of a continuous circulation of A226V strains in the southwestern Indian Ocean since 2006 and the possible reintroduction of CHIKV on Réunion Island, most probably from Madagascar. Once again, human travel may have contributed to the rapid spread of the virus between islands because imported and autochthonous cases on Réunion Island occurred after a holiday period for residents on Réunion Island who often traveled to Madagascar. Migration and birth rate on Réunion Island might have contributed to a decrease in the immunity of the population. Furthermore, seroprevalence in 2007 was not homogenous throughout the territory. A hypothesis would be that a lower immunity of the population in the Saint Paul area and environmental and vectorial characteristics contributed to the emergence of this CHIKV disease cluster.

On Réunion Island, *Ae. albopictus* mosquitoes have been described as the main vector responsible for transmitting CHIKV (*10*). The austral winter may contribute to moderate vector activity and transmission. We cannot exclude a continuous transmission until next austral summer, followed by an increase of cases and an extension to the whole island, as occurred in 2005 (*1*). Epidemiologic and entomologic surveillance has been reorganized to prevent this risk. Medical staff, the general population, and travelers have been informed about the situation through the news media and meetings organized by health authorities, and recommendations have been issued about destroying mosquito breeding sites and preventing mosquito bites.

In recent years, the area of circulation and the epidemic potential of CHIKV have increased, and CHIKV has emerged as a major public health problem. This outbreak could be a new warning to Réunion Island health authorities about the need for preparation not only for CHIKV but also for dengue virus (DENV). With the extent of human travel to and from areas with active CHIKV and DENV circulation, viremic returning travelers constitute an ongoing risk for introduction of such viruses on Réunion Island. In May 2010, two locally acquired DENV-3 cases were also detected, illustrating this threat. These cases occurred during an outbreak of DENV-3 in Comoros Island. Public health efforts to control *Ae. albopictus* mosquitoes have not been completely effective.

This outbreak of CHIKV infection, the detection of autochthonous cases of DENV infection, and the influenza season on Réunion Island emphasize the difficulty of making the appropriate clinical diagnosis. Clinicians and biologists should be aware of the cocirculation of CHIKV, DENV, and influenza viruses. The reemergence of CHIKV on Réunion Island illustrates the permanent threat of circulation of exotic pathogens in the Indian Ocean and the need for strong epidemiologic and laboratory surveillance. Human travel and the geographic expansion of *Ae. albopictus* mosquitoes raise concern for the spread of CHIKV in Europe and North America.

Acknowledgments

We are grateful to the Vector Control Team and to Dominique Polycarpe of the Health Agency of Indian Ocean who collected epidemiologic data. We also thank all the physicians and biologists for their participation in the surveillance of CHIKV and DENV infections on Réunion Island, Jean-Michel Heraud (Institut Pasteur, Madagascar) for sending Chikungunya strains from Madagascar and Valérie Caro (Institut Pasteur, Paris, France) for the sequence genome analysis.

Eric D'Ortenzio,[1] Marc Grandadam, Elsa Balleydier, Marie-Christine Jaffar-Bandjee, Alain Michault, Elise Brottet, Marie Baville, and Laurent Filleul

Author affiliations: Regional Office of French Institute for Public Health Surveillance, Saint-Denis, Réunion Island (E. D'Ortenzio, E. Balleydier, E. Brottet, L. Filleul); Institut Pasteur, Paris, France (M. Grandadam); Regional Hospital, Saint-Denis (M.-C. Jaffar-Bandjee); Regional Hospital, Saint-Pierre, Réunion Island (A. Michault); and Health Agency of Indian Ocean, Saint-Denis (M. Baville)

DOI: 10.3201/eid1702.101056

[1]Current affiliation: Institut Pasteur, Noumea, New Caledonia.

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Address for correspondence: Eric D'Ortenzio, Epidemiology Unit—Institut Pasteur de Nouvelle-Calédonie, 9-11 Avenue Paul Doumer—BP 61 98845 Noumea, New Caledonia; email: edortenzio@pasteur.nc

Segniliparus rugosus–associated Bronchiolitis in California Sea Lion

To the Editor: Until now, *Segniliparus rugosus* has not been isolated from nonhuman animals or the environment (*1*). On April 14, 2010, a rescue team from Pacific Marine Mammal Center impounded an emaciated and unresponsive subadult female California sea lion (*Zalophus californicus*) stranded on the beach at San Onofre, California, USA. Physical examination showed the animal to be obtunded and emaciated (third-stage malnutrition), with moderate bradycardia, hypoventilation, and hypothermia. Euthanasia was elected because of a poor prognosis. Immediately before euthanasia, a blood sample was taken for a complete blood count and serum chemistry evaluation. A postmortem examination was conducted immediately after euthanasia.

The postmortem examination showed marked subcutaneous and visceral adipose tissue depletion, as well as moderate skeletal muscle loss, especially in the axial skeleton. In the lungs, a frothy, greenish, mucoid material exuded from several dozen bronchioles. Samples of the exudate were submitted for cytologic examination and bacterial culturing (IDEXX Laboratories, Irvine, CA, USA). Selected tissues were sampled and fixed in 10% neutral buffered formalin for histopathologic examination.

Complete blood count and serum chemistry analysis showed moderate anemia; relative neutrophilia and monocytosis; mild to moderate relative lymphopenia; moderate to markedly reduced albumin, globulin, and total protein levels; and elevated creatine kinase and alkaline phosphatase levels. Such values are common in California sea lions with severe malnutrition (starvation).

Cytologic examination of the bronchiolar exudate indicated large amounts of mucin with erythrocytes; occasional epithelial cells; and small to moderate numbers of eosinophils, neutrophils, monocytes, and lymphoid cells, characteristic of a mild to moderate, subacute, mixed bronchiolitis. Histologic examinations of 3 sections of lung showed 33 bronchioalveolar foci containing varying numbers of adult *Parafilaroides decorus* nematodes, without associated inflammation. Eleven other foci showed moderate to marked chronic inflammation, with nematodes in only 2 foci. Gram stain did not show bacteria in any of these foci. Lesions were not found in sections of liver, kidney, bladder, spleen, and heart.

A commercial veterinary laboratory (IDEXX Laboratories) isolated an acid-fast organism from the lung swab. This organism was referred to National Jewish Medical and Research Center (Denver, CO, USA) for species identification and sensitivity analysis. By 16S rDNA sequencing, the organism was identified as *S. rugosus*. Sensitivity testing showed that it was susceptible to rifabutin, cycloserine, clofazimine, moxifloxacin, ciprofloxacin, and clarithromycin and resistant to rifampin, streptomycin, amikacin, kanamycin, capreomycin, ethambutol, and ethionamide.

As in humans, this isolation of *S. rugosus* was associated with pathologic changes in the respiratory tract. Whether the relationship was causal or simply a fortuitous isolation of a previously unrecognized part of the normal respiratory flora is uncertain. However, a recent report by Sikorski et al. stated that "Environmental screens and metagenomic surveys did not detect a single phylotype… of the members of the genus *Segniliparus*" (*2*). In contrast, this case report begs the question of whether *S. rugosus* could be free-living in the oceans or part of the flora of any number of ocean-dwelling vertebrates or invertebrates.