Acknowledgments

We thank Olivier Bourry, José Gomez-Peña, and Hubert Bassene for their help during the conduct of field work, Jean-Paul Durand, and all the persons who contributed to the study, especially the French military veterinarians. We thank also the team of the virology laboratory of the Institut de recherche biomédicale des armées (William Daries, Patrick Gravier, and Olivier Merle) for processing the samples.

Financial support was provided in part by the French Defense Medical Service.

Bernard Davoust, Isabelle Leparc-Goffart, Jean-Paul Democheaux, Raphaël Tine, Mamadou Diarra, Grégoire Trombin, Oleg Mediannikov, and Jean-Lou Marié

Author affiliations: Groupe de Travail en Épidémiologie Animale du Service de Santé des Armées, Toulon, France (B. Davoust, J.-P. Democheaux, G. Trombin, J.L. Marié); Unité de Recherche sur les Maladies Infectieuses et Tropicales Émergentes (IRD 198), Dakar, Senegal (B. Davoust, O. Mediannikov); Centre National de Référence des Arbovirus—Institut de Recherche Biomédicale des Armées, Marseille, France (I. Leparc-Goffart); and Services Vétérinaires de la Gendarmerie Nationale, Dakar (R. Tine, M. Diarra).

DOI: http://dx.doi.org/10.3201/eid2008.130691

References


Address for correspondence: Bernard Davoust, Unité de Recherche en Maladies Infectieuses et Tropicales Emergentes (URMITE) CNRS UMR 7278 IRD 198 INSERM U1095 Aix–Marseille Université, Faculté de Médecine, 27 bd Jean Moulin, 13385 Marseille CEDEX 5, France; email: bernard.davoust@gmail.com

Another Dimension

EID publishes thoughtful essays, short stories, or poems on philosophical issues related to science, medical practice, and human health. Topics may include science and the human condition, the unanticipated side of epidemic investigations, or how people perceive and cope with infection and illness. This section is intended to evoke compassion for human suffering and to expand the science reader’s literary scope. Manuscripts are selected for publication as much for their content (the experiences they describe) as for their literary merit.

Severe Encephalitis Caused by Toscana Virus, Greece

To the Editor: In late June 2012, a previously healthy, 49-year-old woman was admitted to the emergency department of Trikala General Hospital in Trikala, Greece, with confusion and delirium. A few hours before admission, she had had a grand mal seizure; she had experienced gastrointestinal distress with fever (38°C) 5 days earlier. On admission, she was intubated and transferred to the intensive care unit, where she underwent mechanical ventilation and sedation.

The patient was a resident of Genesi village (350 m altitude), 22 km west of Trikala in the Thessaly region. She had not traveled abroad or to other area of Greece. Results of blood and cerebrospinal fluid (CSF) laboratory testing were unremarkable except slight leukocytosis (leukocytes 11,330 cells/mm³, 92% neutrophils) and slightly elevated serum lactate dehydrogenase level (240 U/L). Brain imaging showed edema markable except slight leukocytosis (leukocytes 11,330 cells/mm³, 92% neutrophils) and slightly elevated serum lactate dehydrogenase level (240 U/L). Brain imaging showed edema...

enteroviruses, and phleboviruses. PCR for phleboviruses (2) resulted in a PCR product of the expected size, and the sequence was most closely related to those of isolates belonging to the Sandfly fever Naples virus (SFNV) species (Figure). The sequence also had the highest homology (85%) with a Toscana virus (TOSV) strain belonging to lineage C that had been obtained from a patient with central nervous system infection in Croatia in 2008 (3). The TOSV sequence derived from the patient in this report was submitted to GenBank (accession no. KJ418710). On the basis of a partial sequence comparison (202 nt in the polymerase gene), we found that TOSV lineage C differs from lineages A and B by 29% and 30%, respectively.

Forty-one days after symptom onset, a second serum sample was taken from the patient and tested in parallel with the first serum sample by indirect immunofluorescence to detect IgM with the first serum sample by indirect immunofluorescence to detect IgM from the patient and tested in parallel. A second serum sample was taken and found to be positive for IgG and IgA against TOSV and SFNV, respectively, of the sandflies collected (9). Another phlebovirus, Adria virus (belonging to the Salehabad serocomplex), which was initially detected in sandflies collected in Albania, was detected in a febrile child with seizure in Thessaloniki in northern Greece (10). Concerning TOSV, however, although seroconversion has been previously observed in patients in Greece, RNA has not been detected.

For this patient, TOSV was detected by using phlebovirus generic primers. The TOSV sequence found in Greece differs greatly from other TOSV sequences, even from the genetically closer Croatian TOSV sequence (15%). To avoid false-negative results, the high genetic diversity among TOSV strains must be taken into consideration when using TOSV-specific primers.

In conclusion, a novel variant of TOSV has been detected in Greece. Further studies are needed to obtain a whole-genome sequence of the Greek TOSV strain and to identify the vector(s) of the virus. TOSV is a highly variable neurotropic phlebovirus, a characteristic that must be taken into account by laboratory scientists. Clinicians should be aware of the possibility of phlebovirus infections in Mediterranean countries and should include these viruses in the differential diagnosis of febrile illnesses observed during the warm seasons, especially in patients who exhibit neurologic symptoms.

The National Reference Centre for Arboviruses and Hemorrhagic Fever viruses in Thessaloniki, Greece, is financially supported by the Hellenic Center for Disease Control and Prevention.

Anna Papa, Theoniki Paraforou, Ioannis Papakonstantinou, Kiriaki Pagdatoglou, Anastasia Kontana, and Triantafilia Koukoubani
There is evidence that lineage 2 circulates in some regions of Europe (e.g., Italy, Austria, and Greece) (2,3). Further examinations were undertaken, and samples were sent to the Arboviruses and Viral Hemorrhagic Fever Laboratory of the Pasteur Institute of Iran in Teheran. For an IgG ELISA, wells in test plates were coated overnight with mouse hyperimmune ascitic fluid. Native antigen was added, and wells were incubated and washed. Test samples and peroxidase-labeled anti-human or anti-animal immunoglobulin were added. After incubation for 10 min, optical densities were read (6).

Viral RNA was extracted by using the QIAmp Viral RNA Mini Kit (QIA-GEN, Hilden, Germany) from serum of the patient. A reverse transcription PCR was conducted by using a One-Step RT-PCR Kit (QIAGEN). Samples were subjected to 1 cycle at 50°C for 30 min to synthesize cDNA; 95°C for 15 min; and 95°C for 30 s, 54°C for 30 s, and 72°C for 60 s; and a final extension at 72°C for 5 min (6). The serum sample was positive for IgG against WNV. Molecular tests showed positive results for WNV.

The PCR product was sequenced by using the Big Dye Terminator V3.1 Cycle Sequencing Kit (Applied
Severe Encephalitis Caused by Toscana Virus, Greece

Technical Appendix

Technical Appendix Figure. Computed tomography scan image of the brain of a 49-year-old female patient at admission to the emergency department of Trikala General Hospital, Trikala, Greece, June 2012. The woman had confusion and delirium. The scan shows dilated lateral ventricles and increased attenuation of the subarachnoid spaces due to edema. A Toscana virus strain was later detected in the patient.