

Hantaviruses in São Paulo State, Brazil

To the Editor: Hantavirus pulmonary syndrome (HPS) is an emerging health problem in Brazil. This syndrome was first reported in 1993 in three persons living in a rural area of Juquitiba County; two of them died of acute respiratory failure (1). Although Juquitiba County is part of the metropolitan area of greater São Paulo City, patients lived in a recently deforested region. From 1993 through 2002, approximately 200 HPS cases were reported in Brazil, with a 40% case-fatality ratio (Ministry of Health of Brazil, Report on Hantavirus cases 1993–2002, unpub. data).

The wild rodent *Bolomys laziurus* is believed to be the most important hantavirus reservoir in the State of São Paulo, based on high levels of specific antibodies observed in serum from captured specimens (L.E. Pereira, Adolpho Lutz Institute, pers. comm., 2001). The economy of the inland region of Ribeirão Preto in the State of São Paulo, with its 3.5 million inhabitants, is based on the sugar cane agroindustry. The region has been almost completely deforested, with important consequences to the environment and wild rodent ecology. Twenty HPS cases were reported in Ribeirão Preto in the last 5 years, with a 60% case-fatality ratio. Review of medical records showed that a prodromic fever occurred in all 14 case-patients studied; dyspnea, cough, hypotension, and tachycardia occurred in about two thirds of patients; and hemorrhagic phenomena (hematuria, melena, and hypermenorrhea) in about one third. Thrombocytopenia was observed in all the patients, elevated hematocrit in about three fourths, and leukocytosis with neutrophilia and a left shift in the differential count in about two thirds. Serum creatinine levels were also

increased (average level 2 mg/dL). Chest radiographs showed diffuse alveolar flocculant infiltrates in most cases (2,3). Laboratory diagnosis of HPS was made by serologic testing (enzyme-linked immunosorbent assay [ELISA]) in 18 cases and by reverse transcription–polymerase chain reaction (RT-PCR) in 11 cases; for 7 cases, both techniques were used. We performed a nucleotide sequence analysis of the N gene of hantavirus (residues 236–477) obtained from the blood of 11 of the 20 patients. This analysis showed that the infections were caused by Araraquara virus, a previously known hantavirus that had been detected by RT-PCR in the serum of an HPS patient living in a nearby county (4). Thus, Araraquara virus is the causative agent of a severe form of HPS, with a high death rate. This high death rate could also be related to the lack of adequate initial therapy provided by clinicians who probably did not immediately suspect HPS and may have not recommended hospitalization in intensive-care units. In addition, some hospitalized patients were in shock when first seen and were rehydrated with massive quantities of fluids, which may have aggravated pulmonary edema and contributed to death.

The occurrence of 10% of the Brazilian HPS reported cases in Ribeirão Preto indicates that this region is suitable for studying the epidemiology of hantavirus infections. A serologic survey conducted in the region in 1999, which included 567 primary-care patients from Ribeirão Preto, Guariba, and Jardinópolis Counties, found that 7 (1.23%) of them had immunoglobulin (Ig) G antibodies to Sin Nombre virus by ELISA and that 5 of those lived in Jardinópolis (population 30,000), a county where a fatal case of HPS occurred in 1999 (5). Thus, Jardinópolis County was chosen for a population-based survey. In May 2001, we obtained personal informa-

tion and collected fingerprick blood samples from 818 participants, 15–70 years of age, living in urban and rural areas of the county. IgG antibodies to the N recombinant protein of Andes virus were detected by ELISA in the blood samples of 14.3% of the participants (5). Even though all HPS cases in Ribeirão Preto were associated with rural activity and rodent exposure, these serologic data suggest that hantavirus infections are common in Jardinópolis County, independent of sex, profession, or history of contact with rodents. None of the 14.3% participants with IgG antibodies to hantavirus had a history of HPS-like disease, and the ELISA test showed cross-reactions with most of the South American hantaviruses, including Araraquara. Persons living in the urban area had higher levels of antibodies to hantavirus than those from rural areas. In Ribeirão Preto, the physical boundaries of cities have expanded to incorporate other areas, encroaching upon rural areas with many popular subsidized housing complexes. Work-related and recreational rural activities in that region are also frequent, which makes it difficult to interpret these data. These results suggest that in this region of southeast Brazil, hantaviruses may be causing undiagnosed asymptomatic or clinically minor infections in addition to typical HPS. This finding evokes important questions. Is more than one hantavirus circulating in this region, causing mostly benign infections? Is Araraquara virus widespread, causing mostly inapparent infections and only rarely causing HPS? Would HPS be associated with some predisposing condition in the infected person? If more than one hantavirus is circulating in the region, could urban rodents be reservoirs?

Further studies are necessary to better understand the epidemiology and clinical signs and symptoms of hantavirus infection in the region of Ribeirão Preto. Such studies should

emphasize determining the reservoirs, the modes of virus transmission to people, and the possible distinct clinical forms of hantavirus infections.

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References

1. Iversson LB. Hantavirus pulmonary syndrome in the rural area of Juquitiba, São Paulo Metropolitan Area, Brazil. *Rev Inst Med Trop Sao Paulo* 1997;39:237–8.
2. Figueiredo LTM, Moreli ML, Kashima S, Almeida VSO, Félix PR, Bruno JC, et al. Hantavirus pulmonary syndrome (HPS) in Guariba, SP, Brazil. Report of 2 cases. *Rev Inst Med Trop Sao Paulo* 1999;41:131–7.
3. Figueiredo LTM, Campos GM, Rodrigues FB. Síndrome pulmonar e cardiovascular por hantavirus: aspectos epidemiológicos, clínicos, do diagnóstico laboratorial e do

tratamento. [in Portuguese]. *Rev Soc Bras Med Trop* 2001;34:15–27.

4. Johnson AM, de Souza LT, Ferreira IB, Pereira LE, Ksiazek TG, Rollin PE, et al. Genetic investigation of novel hantaviruses causing fatal HPS in Brazil. *J Med Virol* 1999;59:527–35.
5. Holmes RR, Boccanera R, Figueiredo LTM, Mançano SR, Pane C. Seroprevalence of human hantavirus infection in Ribeirão Preto Region of São Paulo State, Brazil. *Emerg Infect Dis* 2000;6:560–1.

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Israeli Spotted Fever *Rickettsia* in Sicilian *Rhipicephalus sanguineus* Ticks

To the Editor: Mediterranean spotted fever (MSF) is endemic in Italy, where it is a reportable disease. From 1992 to 1998, the Italian Ministry of Health was notified of approximately 8,500 cases of human rickettsioses presumed to be MSF. MSF occurs more commonly in some central (Lazio) and southern (Sardinia, Sicily, and Calabria) regions (1,2); in 1998, an average of 8.8 cases occurred for every 100,000 persons in Sicily, compared with the national average of 1.6 cases per 100,000 persons. *Rickettsia conorii* has been thought to be the only pathogenic *Rickettsia* of the spotted fever group in Sicily (3,4) or the western Mediterranean area. Recently, three different spotted fever group rickettsiae, including *R. helvetica*, were detected in *Ixodes ricinus* ticks from central and northern Italy. This finding suggests that bacteria other than *R. conorii* are involved in rickettsial diseases in Italy (5).

To investigate whether unusual tick-transmitted rickettsiae are also present in Sicily, we used molecular-sequence-based identification techniques to study two strains isolated from the hemolymph of *Rhipicephalus sanguineus* ticks collected in 1990 in western Sicily. These isolates had been previously identified by serologic tests as belonging to the spotted fever group rickettsiae. We obtained bacterial DNA and performed polymerase chain reaction (PCR) for *ompA* gene and restriction analysis under conditions previously described by Roux et al. (6). Our observation of a peculiar *PstI* profile allowed a presumptive identification of one of the two tick isolates as belonging to the Israeli spotted fever rickettsiae, while the other showed a restriction profile corresponding to that of *R. conorii* strain Seven. To confirm the identification of the Israeli spotted fever *Rickettsia* isolate, we sequenced the PCR-amplified fragment of *ompA* gene (MWG-Biotech AG, Ebersberg, Germany) and aligned sequence data with homologous sequences of reference strains of the spotted fever group rickettsiae retrieved from the GenBank database. Sequence analysis showed

100% similarity with the homologous sequence of Israeli spotted fever *Rickettsia* reference strain ISTT CDC1 (GenBank accession no. U43797). The Israeli spotted fever *Rickettsia* belongs to the *R. conorii* complex (7,8) and was first isolated in 1974 from ticks and humans. Initially, Israeli spotted fever rickettsiae distribution appeared to be restricted to Israel (9), but more recently the organism has also been isolated from patients with MSF in Portugal (10). Our finding of Israeli spotted fever *Rickettsia* infection in a *R. sanguineus* tick, the main vector for MSF in Sicily, also suggests that the geographic distribution of Israeli spotted fever might be wider than previously thought, including not only Israel and the Iberian Peninsula but also Italy.

Molecular analysis of spotted fever group *Rickettsia* isolates from Sicilian MSF patients is under way to verify this hypothesis. Because initial signs and symptoms of Israeli spotted fever are particularly uncharacteristic, awareness of the presence of Israeli spotted fever *Rickettsia* in our geographic area may hasten provision of the appropriate treatment. The Sicilian *ompA* gene sequence described in this study has been