Prevalence of serum antibodies to hantavirus in a rural population from the Southern State of Santa Catarina, Brazil

Gregório Wrublevski Pereira1,2,3, André Martins Teixeira1,3, Mirela Silva de Souza1,3, Alixandre Dias Braga1,3, Gilberto Sabino dos Santos Junior4,5, Glauçiane Garcia de Figueiredo4,5, Luiz Tadeu Moraes Figueiredo4,6 and Alessandra Abel Borges1,7

ABSTRACT

Introduction: Rodent-borne hantaviruses cause severe human diseases. We completed a serological survey of hantavirus infection in rural inhabitants of Turvo County, in the southern State of Santa Catarina, Brazil, in which seropositivity for hantavirus was correlated to previous disease in the participants. Methods: The levels of IgG antibodies to hantavirus Araraquara in the sera of 257 individuals were determined using an immunoenzymatic assay. Results: IgG antibodies to hantavirus were found in 2.3% of the participants. All seropositive participants reported previous disease with symptoms suggestive of hantavirus cardiopulmonary syndrome. Conclusions: Human infections causing unreported cardiopulmonary syndrome probably occur in the southern State of Santa Catarina. Keywords: Hantavirus. Seroepidemiological studies. Hantavirus pulmonary syndrome.

RESUMO


1. Faculdade de Medicina, Universidade do Sul de Santa Catarina, Tubarão, SC. 2. Grupo de Pesquisa em Pneumologia e Semiologia, Universidade do Sul de Santa Catarina, Tubarão, SC. 3. Unidade de Pesquisa em Virologia, Universidade do Sul de Santa Catarina, Tubarão, SC. 4. Centro de Pesquisa em Virologia, Faculdade de Medicina, Universidade de São Paulo, Ribeirão Preto, SP. 5. Programa de Pós-Graduação em Imunologia Básica e Aplicada, Faculdade de Medicina, Universidade de São Paulo, Ribeirão Preto, SP. 6. Departamento de Clínica Médica, Faculdade de Medicina, Universidade de São Paulo, Ribeirão Preto, SP. 7. Laboratório de Pesquisas em Virologia e Imunologia, Instituto de Ciências Biológicas e da Saúde, Universidade Federal de Alagoas, Maceió, AL.

Address to: Dra. Alessandra Abel Borges. Lab. Pesquisas Virologia e Imunologia/ICBS/UFAL. Pça Afânio Jorge s/n, Prado, 57010-020 Maceió, AL, Brasil. Phone: 55 82 3336-3444 r: 223; Fax: 55 82 3221-2501 e-mail: alessandra.a.borges@gmail.com

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Hantaviruses are zoonotic diseases transmitted to humans by the excreta of rodents, the natural reservoirs of these viruses. In the Americas, hantaviruses are associated with the Sigmodontinae subfamily of Muridae rodents1,2. Different species of hantavirus have been identified as the etiological agents responsible for two human diseases: hemorrhagic fever with renal syndrome (HFRS) in Asia and Europe and hantavirus pulmonary syndrome (HPS) in the Americas3. HPS was first reported in 1993, and through May 2010, a total of 1,253 cases of HPS have been reported in Brazil, with a case-fatality rate of 39%4,5.

In the State of Santa Catarina (SC), located in the South Region of Brazil, 211 cases of HPS have been recorded since 1999, with 54 human deaths (case-fatality rate of 25.4%)3. The incidence of HPS in SC has increased each year. Currently, SC ranks second among Brazilian states with respect to the number of HPS cases4. Hantavirus infections in SC have been documented in the western and central regions of the state, and cases have not been reported in the southern region. However, the antibody levels to hantavirus in the population in the southern region are unknown, and there is no evidence that hantavirus infections do not occur in the southern part of SC5.

Herein, we report the results of a serological survey for hantavirus among individuals living in the rural area of Turvo County in the southern region of SC. This study was approved by the Research Ethics Committee of the University of South of Santa Catarina, process number 08.057.4.01.III. A total of 257 participants voluntarily consented to participate in the study after a brief explanation about its aim. The participants were interviewed individually to obtain information about their past medical history. Data for one participant were also obtained from medical records. In total, 149 (57.9%) men and 108 (42%) women, mostly adults younger than 60 years old, participated in the study. Among them, 223 reported exposure to rodents or their excreta at work. After the interview, the blood of the participants was collected, and their sera were tested for hantavirus-specific IgG antibodies.

Sera were tested using an indirect IgG ELISA using the N recombinant (rN) protein of the Araraquara hantavirus (ARAV) as the antigen. This technique has been previously described by Figueiredo et al. Briefly, polystyrene microtiter plates (Greiner Bio-one, Germany) were coated overnight in a wet chamber at 4°C with 2μg/mL of ARAV rN protein (lines A to D) or with negative
TABLE 1 - Data for the 6 individuals seropositive for hantavirus from Turvo, State of Santa Catarina, Brazil.

<table>
<thead>
<tr>
<th>Features</th>
<th>Participant n=16</th>
<th>Participant n=17</th>
<th>Participant n=18</th>
<th>Participant n=19</th>
<th>Participant n=52</th>
<th>Participant n=88</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>54</td>
<td>51</td>
<td>55</td>
<td>38</td>
<td>36</td>
<td>61</td>
</tr>
<tr>
<td>Sex</td>
<td>M</td>
<td>F</td>
<td>M</td>
<td>M</td>
<td>M</td>
<td>M</td>
</tr>
<tr>
<td>Current occupation</td>
<td>government employee</td>
<td>maintenance assistant</td>
<td>farmer</td>
<td>farmer</td>
<td>maintenance assistant</td>
<td>farmer</td>
</tr>
<tr>
<td>Past occupations</td>
<td>farmer and bricklayer</td>
<td>Farmer</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>High-risk activities: cleaning attics, barns, sheds, abandoned houses or deposits; storage of bales of wool, wood or grass; plowing; livestock farming; unclogging drains</td>
<td>yes*</td>
<td>yes*</td>
<td>yes*</td>
<td>yes*</td>
<td>yes*</td>
<td>yes*</td>
</tr>
<tr>
<td>Symptoms of past illnesses: fever, myalgia, headache, malaise, dyspnea, oliguria, chest pain, abdominal pain, vomiting and hypotension</td>
<td>yes**</td>
<td>yes**</td>
<td>yes**</td>
<td>yes**</td>
<td>yes**</td>
<td>yes**</td>
</tr>
</tbody>
</table>

M: male, F: female. *The participant reported performing all of these high-risk activities, **The participant reported having had almost all of these symptoms during an illness episode in the past.

control antigen (purified plasmid pET200D with no insert - lines E to H). Then the microplate wells were washed six times with PBST (phosphate-buffered saline [PBS]-1% Tween 20) and incubated with a blocking solution (10% skim milk powder in PBST) at 37°C for 2h. After washing the plates six times with wash buffer, the samples were diluted 1:100 in blocking solution and added to the plates at a volume of 50uL/well. The plates were incubated for 1h at 37°C and washed, and then peroxidase-labeled affinity-purified goat-human IgG antibody (Sigma, USA) diluted 1:2,000 in blocking solution was added (50uL/well). After incubation for 1h at 37°C, the plates were washed, and 50uL/well of 2,2'-azinobis (3-ethylbenzthiazoline-6-sulfonic acid) (ABTS) substrate (KPL, EUA) was added. After appearance of the color, the plates were read in a spectrophotometer (Flow, Finland) at 405nm. Values were expressed as the optical density (OD) obtained with ARAV rN antigen minus the OD values for the control antigens. The cut-off value of the test was determined by the mean plus three standard deviations of the ODs obtained from at least four negative control serum samples. Seropositive samples were tittered from dilutions of 1:100 to 1:25,600.

Six participants had IgG antibodies to hantavirus (2.3% seropositivity). This prevalence is similar to that observed in previous studies in areas of Brazil with HPS reported cases8,9. The titers of the positive sera were 800, 1,600 and 3,200. These serum titers can be considered high, although they did not allow the determination of the time elapsed since infection. In a similar study performed in São Paulo, the highest serum titers for hantavirus were 40010.

A previous infection by hantavirus was observed in 2.3% of the participants of the survey, showing that these viruses probably produce human infections in the southern region of SC, where HPS has not yet been reported. At least one of the seropositive participants had a previous unconfirmed disease compatible with HPS. Thus, it is important to inform local clinicians about the epidemiology, signs and symptoms of HPS so that this disease is included in the differential diagnosis of pneumonia with acute respiratory failure.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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