and α2,6-SA (9). The equilibrium dissociation constant for 3′-Sialyl-N-acetyllactosamine is 12.2 (SD ± 0.7 nmol/L) and for 6′-Sialyl-N-acetyllactosamine is 43.3 (SD ± 2.8 nmol/L) (Appendix). These values show that A/common gull/Saratov/1676/2018 has prevalent affinity for the avian-like receptor with lower, but increased, affinity for the human-like receptor, compared with H5N1 strain A/rook/Chany/32/2015 clade 2.3.2.1.C.

Analysis of homology of A/common gull/Saratov/1676/2018 with H5N6 strains available from GISAID showed that all 8 gene segments clustered with human H5N6 strains isolated in southeast China in 2018. We noted 99% homology with human strain A/Guangxi/32797/2018 for all genes, a genetic similarity that raises the question of which pathway led to the spread of the virus. We believe A/common gull/Saratov/1676/2018 was transferred to eastern Russia through northeast Siberia, where HPAI H5N8 clade 2.3.4.4.A was detected in 2018 (10), the same pathway through which H5N8 virus was transferred from Southeast Asia to Europe. These viral pathogens could be spread by migratory birds over long distances along flyways from southern China to southwestern Russia during a migration season. Our study indicates that emerging H5N6 viruses are a potential threat to public health.

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About the Author
Dr. Susloparov is a senior researcher at the Zoonosis Infections and Influenza Department, State Research Center of Virology and Biotechnology Vector, Koltsovo, Russia. His research interests include the molecular genetics, epidemiology, and host–pathogen interaction of avian influenza viruses.

References

Address for correspondence: Ivan M. Susloparov, State Research Center of Virology and Biotechnology Vector, 630559, Koltsovo, Novosibirsk Region, Russia; email: imsous@vector.nsc.ru

Human Parasitism by Amblyomma parkeri Ticks Infected with Candidatus Rickettsia paranaensis, Brazil

Ana Beatriz P. Borsoi, Karla Bitencourth, Stefan V. de Oliveira, Marinete Amorim, Gilberto S. Gazêta

Author affiliations: Instituto Oswaldo Cruz, Rio de Janeiro, Brazil (A.B.P. Borsoi, K. Bitencourth, M. Amorim, G.S. Gazêta); Universidade Federal de Uberlândia, Uberlândia, Brazil (S.V. de Oliveira); Ministério da Saúde do Brasil, Brasília, Brazil (S.V. de Oliveira)

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Spotted fever is the main rickettsial disease in Brazil. We report 12 cases of human parasitism by *Amblyomma parkeri* in the Atlantic rainforest, an area of Brazil to which spotted fever is endemic. Nine of the ticks were infected with *Candidatus* Rickettsia paranaensis.

Spotted fever is considered the main tickborne disease in South America (1). In Brazil, spotted fever has been reported since the 1920s and is known to show great clinical diversity and ecopathological scenario complexity, involving *Rickettsia rickettsii* transmitted by *Amblyomma sculptum* and *A. aureolatum* ticks and *Rickettsia parkeri* strain Atlantic rainforest vectored by *A. ovale* ticks (2). However, several studies have identified different *Rickettsia* species infecting a variety of tick species in Brazil, indicating the possibility of newly emerging spotted fever scenarios in Brazil (1–3).

In southern Brazil, in addition to the scenario already established for the Atlantic forest region, studies indicate the possibility of a unique cycle developing in the Pampa biome, in which *R. parkeri* sensu stricto might be associated with spotted fever cases involving an *A. tigrinum* tick vector (3). Accordingly, to expand the understanding of the spotted fever scenario in Brazil, we conducted a molecular study of *Rickettsia* in *A. parkeri* ticks as parasites of humans in an area of Brazil to which spotted fever is endemic.

During 2013–2018, in an investigation and surveillance of spotted fever cases in urban areas near Atlantic rainforest fragments in the Parana, Santa Catarina, and Rio Grande do Sul states in southern Brazil, we collected 12 tick nymphs parasitizing humans and morphologically identified these ticks as *A. parkeri* (4). We individually processed 11 specimens for DNA extraction (5), subjected this DNA to PCR for molecular confirmation of tick species (6), and isolated *gltA*, *htrA*, *ompA*, and *ompB* gene fragments (Appendix Table, https://wwwnc.cdc.gov/EID/article/25/12/19-0988-App1.pdf). We purified PCR products, sequenced them, and compared them with rickettsial sequences available in GenBank. We subjected concatenated aligned rickettsial sequences to maximum-likelihood analysis.

We identified *A. parkeri* ticks with containing rickettsia in all 3 states studied. Nine samples amplified fragments from ≥1 of the 4 rickettsia gene markers studied. All sequences for *ompB* and *ompA* gene fragments showed 100% similarity with *Candidatus* Rickettsia paranaensis (GenBank accession nos. KX018050, JN126322, and...
The pathogenicity of Candidatus R. paranaensis is unknown. However, Peckle et al. (7) placed it close to the Old World species R. africae and R. sibirica, both of which are proven pathogenic species (1). A. parkeri nymphs infected by Candidatus R. paranaensis are not uncommon (7) and might have high frequencies of infection. Luz et al. (8) reported that 75% of passariform birds in southeastern Brazil were infected with ticks, a value similar to that obtained in this study (81.81%) for humans in the southern region. Thus, circulation of Candidatus R. paranaensis in the Atlantic Forest biome might be closely associated with the presence of A. parkeri immature tick stages and passeriform birds.

Although reports of human parasitism by tick species of the genus Amblyomma are increasing, A. parkeri ticks have been rarely reported from humans, although there are reports of parasitism in the Atlantic rainforest area of southeastern Brazil, including a high prevalence of this ixodid (nymphs) on humans in Rio Grande do Sul State (9,10). Although these reports were for a region to which spotted fever is endemic, there was no study of the associated rickettsia. However, our results show 12 humans parasitized by A. parkeri nymphs in the 3 states that comprise the southern region of Brazil, indicating that the parasitism of humans by such ticks is more common than that reported. Examples of Candidatus R. paranaensis in A. parkeri parasitizing humans in an area to which spotted fever is endemic, with milder clinical characteristics (2), highlight the need to investigate the role of vector and rickettsia in spotted fever in southern Brazil. This investigation should help in formulating appropriate public health responses by existing surveillance programs.

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About the Author
Ms. Borsoi is a PhD student at the Oswaldo Cruz Institute, Rio de Janeiro, Brazil. Her primary research interests are tick taxonomy and rickettsia, with an emphasis on tick–human interactions.

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Address for correspondence: Karla Bitencourth, Laboratório de Referência Nacional em Vetores das Riquetsioses, Anexo Posterior ao Pavilhão Lauro Travassos, Sala 8, Instituto Oswaldo Cruz, Fundação Oswaldo Cruz, Av. Brasil, 4.365, Manguinhos, Rio de Janeiro RJ 21040-900, Brazil; email: karlabitencourth@gmail.com
Human Parasitism by *Amblyomma parkeri* Ticks Infected with *Candidatus Rickettsia paranaensis*, Brazil

**Appendix**

**Appendix Table.** Primers used for PCRs to study human parasitism by *Amblyomma parkeri* ticks infected with *Candidatus Rickettsia paranaensis*, Brazil

<table>
<thead>
<tr>
<th>Gene</th>
<th>PCR characteristic</th>
<th>Primer</th>
<th>Nucleotide sequence, 5'→3'</th>
<th>Fragment, bp</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>glt</em></td>
<td>NA</td>
<td>CS2-78</td>
<td>GCAAGTATCGGTGAGGATGTAAT</td>
<td>401</td>
<td>(1)</td>
</tr>
<tr>
<td>NA</td>
<td>CS2-323</td>
<td>GCTTCTTACACGTGCTG</td>
<td>834</td>
<td>(2)</td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td>CS-1069</td>
<td>CAGGGTCTCGGTGCTTCT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>htr</em></td>
<td>Nested, primary round</td>
<td>17k-5</td>
<td>GCTTTACAAAATTCTAAAAACCATATA</td>
<td>549</td>
<td>(2)</td>
</tr>
<tr>
<td>NA</td>
<td>CS4</td>
<td>GCTCTTCTCATCCTATGGCTATTAT</td>
<td>434</td>
<td>(3)</td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td>CS4</td>
<td>CAGGGTCTTCGTGCATTTCTT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>omp</em></td>
<td>Nested, primary round</td>
<td>ompB-OF</td>
<td>GTAACCGAAGTAATGTCCTGTA</td>
<td>511</td>
<td>(4)</td>
</tr>
<tr>
<td>Nested, primary round</td>
<td>ompB-OR</td>
<td>CTTTATAACCGCTAAACCC</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Secondary round</td>
<td>ompB SFG-IF</td>
<td>GCTTTACAAAATTCTAAAAACCATATA</td>
<td>425</td>
<td>(4)</td>
<td></td>
</tr>
<tr>
<td>Secondary round</td>
<td>ompB SFG/TG-IR</td>
<td>GCTTTACAAAATTCTAAAAACCATATA</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td>M59</td>
<td>CCAGGGTCTTCGTGCATTTCTT</td>
<td>862</td>
<td>(5)</td>
<td></td>
</tr>
<tr>
<td>NA</td>
<td>Rr</td>
<td>ATGGCGAATATTTCTCCAAAA</td>
<td>532</td>
<td>(6)</td>
<td></td>
</tr>
</tbody>
</table>

*NA, not applicable.
†References for oligonucleotides and respective amplification protocols used.

**References**


