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

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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 ecoepidemiologic 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 *glt*A, *htr*A, *omp*A, and *omp*B 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 *omp*B and *omp*A gene fragments showed 100% similarity with *Candidatus* Rickettsia paranaensis (GenBank accession nos. KX018050, JN126322, and

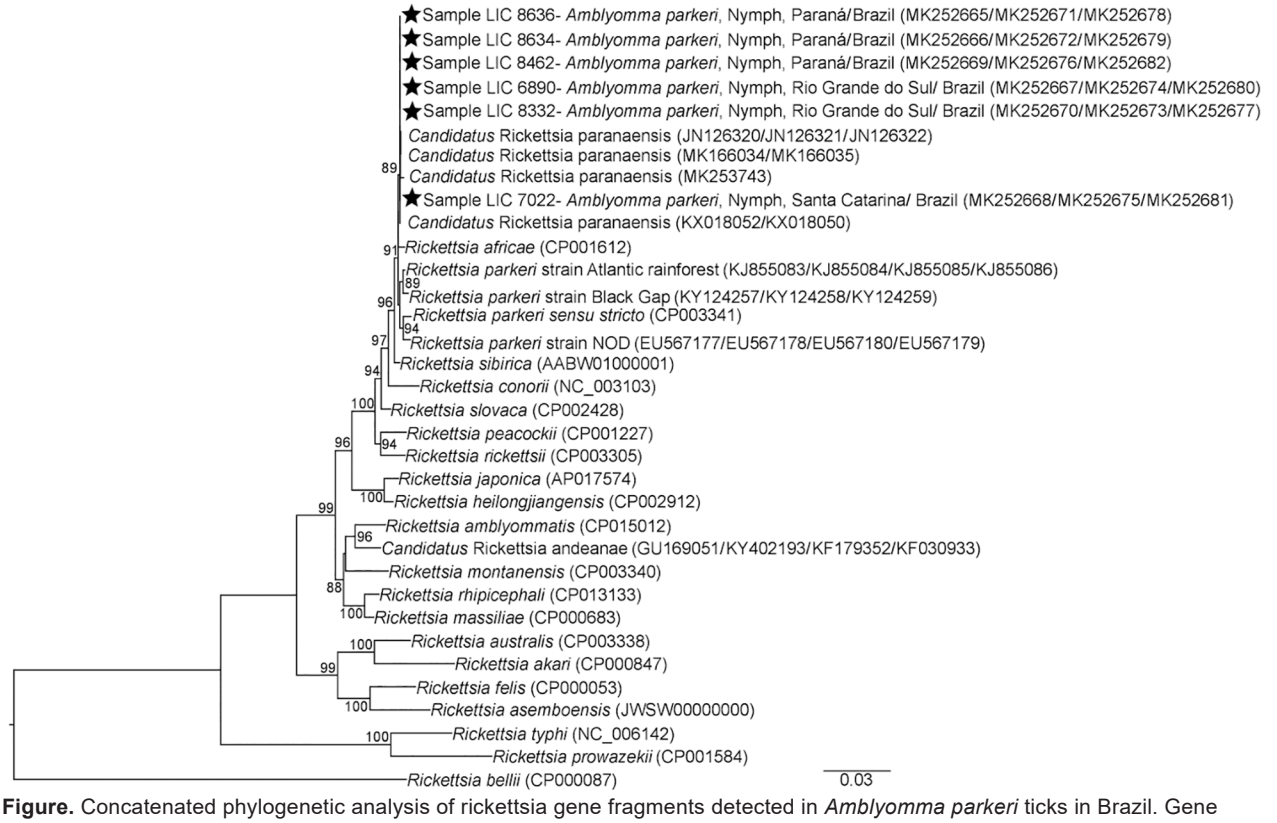

Figure. Concatenated phylogenetic analysis of rickettsia gene fragments detected in *Amblyomma parkeri* ticks in Brazil. Gene fragments *glt*A (1,013 bp), *htr*A (370 bp), *omp*A (494 bp), and *omp*B (822 bp) were inferred by maximum-likelihood analysis with the evolution model T92 + G (Tamura model). Values on the branches indicate bootstrap values (cutoff value 70%). Stars indicate sequences obtained in this study. GenBank accession numbers are given in parentheses. Scale bar indicates nucleotide substitutions per site.

JN126321). The *htr*A and *glt*A sequences had 100% similarity to many of the spotted fever group rickettsia, including *Candidatus* R. paranaensis (GenBank accession nos. KX018052 and JN126320). Phylogenetic analysis showed that bacteria detected in *A. parkeri* ticks from southern Brazil were in the same clade as *Candidatus* R. paranaensis (Figure).

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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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*

Gene	PCR characteristic	Primer	Nucleotide sequence, 5'→3'	Fragment, bp	Reference†
*glt*A	NA	CS2-78	GCAAGTATCGGTGAGGATGTAAT	401	(*1*)
	NA	CS2-323	GCTTCCTTAAAATTCAATAAATCAGGAT		
	NA	CS4-239	GCTCTTCTCATCCTATGGCTATTAT	834	(*2*)
	NA	CS4-1069	CAGGGTCTTCGTGCATTTCTT		
*htr*A	Nested, primary round	17k-5	GCTTTACAAAATTCTAAAAACCATATA	549	(*2*)
	Nested, primary round	17k-3	TGTCTATCAATTCACAACTTGCC		
	Secondary round	17Kd1	GCTCTTGCAACTTCTATGTT	434	(*3*)
	Secondary round	17Kd2	CATTGTTCGTCAGGTTGGCG		
omp B	Nested, primary round	ompB-OF	GTAACCGGAAGTAATCGTTTCGTAA	511	(*4*)
	Nested, primary round	ompB-OR	CTTTATAACCAGCTAAACCACC		
	Secondary round	ompB SFG-IF	GTTTAATACGTGCTGCTAACCAA	425	(*4*)
	Secondary round	ompB SFG/TG-IR	GGTTTGGCCCATATACCATAAG		
	NA	120-M59	CCGCAGGGTTGGTAACTGC	862	(*5*)
	NA	120-807	CCTTTTAGATTACCGCCTAA		
omp A	NA	Rr 190.70p	ATGGCGAATATTTCTCCAAAA	532	(*6*)
	NA	Rr 190.602n	AGTGCAGCATTCGCTCCCCCT		

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

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