

Genotyping revealed the *S. hematobium*-*S. bovis* hybrid parasite described in previous outbreaks, suggesting ongoing transmission rather than reintroduction. The parasite's emergence in another river cannot be explained by the persistence of infected snails (9) but could be explained by reseeding of the river by a mammalian host.

Animal reservoirs have been discussed as a possible explanation for ongoing transmission; however, evidence of a major role is lacking. No infection has been detected in livestock in the region, and the only infected animals found were 2 rats (10). Even if we cannot rule out the influence of an undetected animal reservoir (e.g., *Ovis aries musimon*, wild sheep native to Corsica, have never been tested), the most likely explanation is that 1 or several infected persons continue to infest the water.

In summary, this case highlights that transmission of schistosomiasis in Corsica is ongoing and is no longer restricted to the Cavu River. The parasite appears to be of the same strain detected previously on the island. The infection was acquired at a frequented tourist site, suggesting that more persons might have been infected. Further screening of residents and tourists is urgently needed.

About the Author

Dr. Rothe is deputy head of the division of tropical medicine at the LMU Hospital Munich. Her main areas of interest are clinical tropical medicine, neglected tropical diseases, emerging infections, travel medicine, and migrant medicine.

References

- Berry A, Moné H, Iriart X, Mouahid G, Aboo O, Boissier J, et al. Schistosomiasis haematobium, Corsica, France. *Emerg Infect Dis*. 2014;20:1595-7. <https://doi.org/10.3201/eid2009.140928>
- Moné H, Holtfreter MC, Allienne JF, Mintsu-Nguéma R, Ibikounlé M, Boissier J, et al. Introgressive hybridizations of *Schistosoma haematobium* by *Schistosoma bovis* at the origin of the first case report of schistosomiasis in Corsica (France, Europe). *Parasitol Res*. 2015;114:4127-33. <https://doi.org/10.1007/s00436-015-4643-4>
- Ramalli L, Mulero S, Noël H, Chiappini JD, Vincent J, Barré-Cardi H, et al. Persistence of schistosomal transmission linked to the Cavu River in southern Corsica since 2013. *Euro Surveill*. 2018;23. <https://doi.org/10.2807/1560-7917.ES.2018.23.4.18-00017>
- Boissier J, Grech-Angelini S, Webster BL, Allienne JF, Huyse T, Mas-Coma S, et al. Outbreak of urogenital schistosomiasis in Corsica (France): an epidemiological case study. *Lancet Infect Dis*. 2016;16:971-9. [https://doi.org/10.1016/S1473-3099\(16\)00175-4](https://doi.org/10.1016/S1473-3099(16)00175-4)
- Holtfreter MC, Moné H, Müller-Stöver I, Mouahid G, Richter J. *Schistosoma haematobium* infections acquired in Corsica, France, August 2013. *Euro Surveill*. 2014;19:20821. <https://doi.org/10.2807/1560-7917.ES2014.19.22.20821>
- Berry A, Paris L, Boissier J, Caumes E. Schistosomiasis screening of travelers to Corsica, France. *Emerg Infect Dis*. 2016;22:159. <https://doi.org/10.3201/eid2201.151290>
- Noël H, Ruello M, Maccary A, Pelat C, Sommen C, Boissier J, et al. Large outbreak of urogenital schistosomiasis acquired in Southern Corsica, France: monitoring early signs of endemicization? *Clin Microbiol Infect*. 2018;24:295-300. <https://doi.org/10.1016/j.cmi.2017.06.026>
- Ramalli L, Noël H, Mulero S, Vincent J, Malfait P, Allienne JF, et al. Persistence of urogenital schistosomiasis in Southern Corsica since 2013 [in French]. *Med Mal Infect*. 2018;48:S126.
- Mulero S, Rey O, Arancibia N, Mas-Coma S, Boissier J. Persistent establishment of a tropical disease in Europe: the preadaptation of schistosomes to overwinter. *Parasit Vectors*. 2019;12:379. <https://doi.org/10.1186/s13071-019-3635-0>
- Oleaga A, Rey O, Polack B, Grech-Angelini S, Quilichini Y, Pérez-Sánchez R, et al. Epidemiological surveillance of schistosomiasis outbreak in Corsica (France): are animal reservoir hosts implicated in local transmission? *PLoS Negl Trop Dis*. 2019;13:e0007543. <https://doi.org/10.1371/journal.pntd.0007543>

Address for correspondence: Camilla Rothe, LMU University Hospital Munich, 4th Medical Department, Division of Infectious Diseases and Tropical Medicine, Leopoldstr. 5, D-80802 Munich, Germany; email: rothe@lrz.uni-muenchen.de

Relapsing Fever Group *Borrelia* in Human-Biting Soft Ticks, Brazil

Sebastián Muñoz-Leal,¹ Álvaro A. Faccini-Martínez, Bruno M. Teixeira, Maria Marlene Martins, Maria Carolina A. Serpa, Glauber M.B. Oliveira, Felipe R. Jorge, Richard C. Pacheco, Francisco B. Costa, Hermes R. Luz, Marcelo B. Labruna

Author affiliations: Universidade de São Paulo, São Paulo, Brazil (S. Muñoz-Leal, M.C.A. Serpa, G.M.B. Oliveira, M.B. Labruna); Asociación Colombiana de Infectología, Bogotá, Colombia (Á.A. Faccini-Martínez); Centro Universitario INTA-UNINTA, Sobral, Brazil (B.M. Teixeira, F.R. Jorge); Universidade Federal de

¹Current affiliation: Universidad de Concepción, Chillán, Ñuble, Chile.

Uberlândia, Uberlândia, Brazil (M.M. Martins); Universidade Federal de Mato Grosso, Cuiabá, Brazil (R.C. Pacheco); Universidade Estadual do Maranhão, São Luís, Brazil (F.B. Costa); Universidade Federal do Maranhão, São Luís (H.R. Luz)

DOI: <https://doi.org/10.3201/eid2701.200349>

We conducted a molecular survey for *Borrelia* spp. in *Ornithodoros* ticks previously reported as biting humans. We collected specimens in natural ecosystems and inside human dwellings in 6 states in Brazil. Phylogenetic analyses unveiled the occurrence of 4 putatively new species of relapsing fever group borreliae.

Tick-borne relapsing fever (TBRF) is a vectorborne disease caused by spirochetes of the genus *Borrelia* that thrive in enzootic cycles and are transmitted mainly by soft ticks of the genus *Ornithodoros* (1). Humans bitten by infected ticks can become ill and present a typical recurrent febrile syndrome (1). In the New World, research on TBRF persists mainly in North America, where *Borrelia turicatae*, *B. parkeri*, and *B. hermsii* infect humans (1). Meanwhile, the knowledge on relapsing fever spirochetes in South America has remained comparatively incomplete. In Brazil, *Ornithodoros brasiliensis*, *O. fonsecai*, *O. mimon*, *O. rietcorraei*, and *O. rostratus* ticks have been reported to parasitize humans (2,3), yet their role as vectors of *Borrelia* spp. is unknown. Recently, in Brazil, *B. venezuelensis*, the agent of South American TBRF during the first half of the 20th century, was isolated from the anthropophilic tick *O. rudis* (4). This finding highlighted the occurrence of pathogenic relapsing fever group borreliae (RFGB) and called attention to study human-biting *Ornithodoros* ticks as possible vectors of these microorganisms.

During December 2018–October 2019, we conducted collections of soft ticks in the Brazilian states of Ceará (CE), Goiás (GO), Mato Grosso (MT), Mato Grosso do Sul (MS), Maranhão (MA), and Rondônia (RO) (Appendix Figure, <https://wwwnc.cdc.gov/EID/article/27/1/20-0349-App1.pdf>). Collections in MS were implemented using dry ice as an attractor; in CE, GO, MA, and RO, we collected soft ticks inside caves, abandoned nests or between rocks in rural areas. In MT, specimens were collected on the walls of an inhabited house in an urban area. Collections of ticks were authorized by Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio permits 65137-1 and 36413-1).

A total of 665 specimens (236 males, 145 females, 284 nymphs) belonging to 8 species of the genus *Ornithodoros* were submitted to individual or pooled

DNA extractions (Appendix Table 1). We screened extractions with a *Borrelia*-specific real-time PCR with primers Bor16S3F and Bor16S3R and probe Bor16S3P, using 2 µL of genomic DNA, to amplify a fragment of the 16S rRNA gene (5). Samples with cycle threshold values <32 were tested with a battery of PCRs targeting the 16S rRNA and the *flaB* and *glpQ* borrelial genes.

Four species of ticks were positive by *Borrelia*-specific real-time PCR. We generated sequences of *Borrelia* 16S rRNA, *flaB*, and *glpQ* genes for these specimens (Appendix Table 1). Two haplotypes of 16S rRNA gene were sequenced from each of the 2 positive *O. mimon* ticks, and the obtained sequences of *flaB* and *glpQ* were identical for both specimens. One haplotype for each gene was obtained for *O. hasei* and the *Ornithodoros* sp. ticks from CE, and only a 16S rDNA sequence was obtained from *O. rietcorraei* ticks (Appendix Table 2). With high support values, Bayesian phylogenetic analyses showed that the *Borrelia* spp. characterized from *O. mimon*, *O. rietcorraei*, and the *Ornithodoros* sp. ticks from CE form a monophyletic clade related to RFGB occurring in the Old World. In turn, the *Borrelia* sp. harbored by *O. hasei* ticks clustered within New World RFGB (Figure). These results add further evidence that Old and New World RFGB do not necessarily have defined geographic distributions but rather correspond to arbitrary groups.

Five species of *Ornithodoros* ticks have been reported to parasitize humans in Brazil (2,3). We have added 2 more species to this list, as *O. hasei* and the *Ornithodoros* sp. ticks from CE avidly bit us during collections in the field (data not shown). Although with low prevalence, these 2 species, together with *O. mimon* and *O. rietcorraei*, harbored DNA of putatively new *Borrelia* spp. phylogenetically related to the relapsing fever group. The implications of these new spirochetes as human pathogens are still unknown. *O. mimon* and *O. rietcorraei* ticks are associated with human parasitism in urban and rural dwellings in Brazil (2,3), so vector roles of both species should not be overlooked.

TBRF courses with febrile episodes and should be considered as a differential diagnosis within the spectrum of diseases that cause an undifferentiated febrile syndrome (UFS) (6). Although specific data are vague for the states where tick collections were performed in this study, UFS is common in Brazil; mosquito-borne viruses and malaria are the main etiologic agents (7,8). Nevertheless, febrile illnesses still remain underdiagnosed in a substantial proportion of the cases in the country (7,8). The results of this study are a contribution to the knowledge of RFGB in human-biting *Ornithodoros* ticks, and stress the

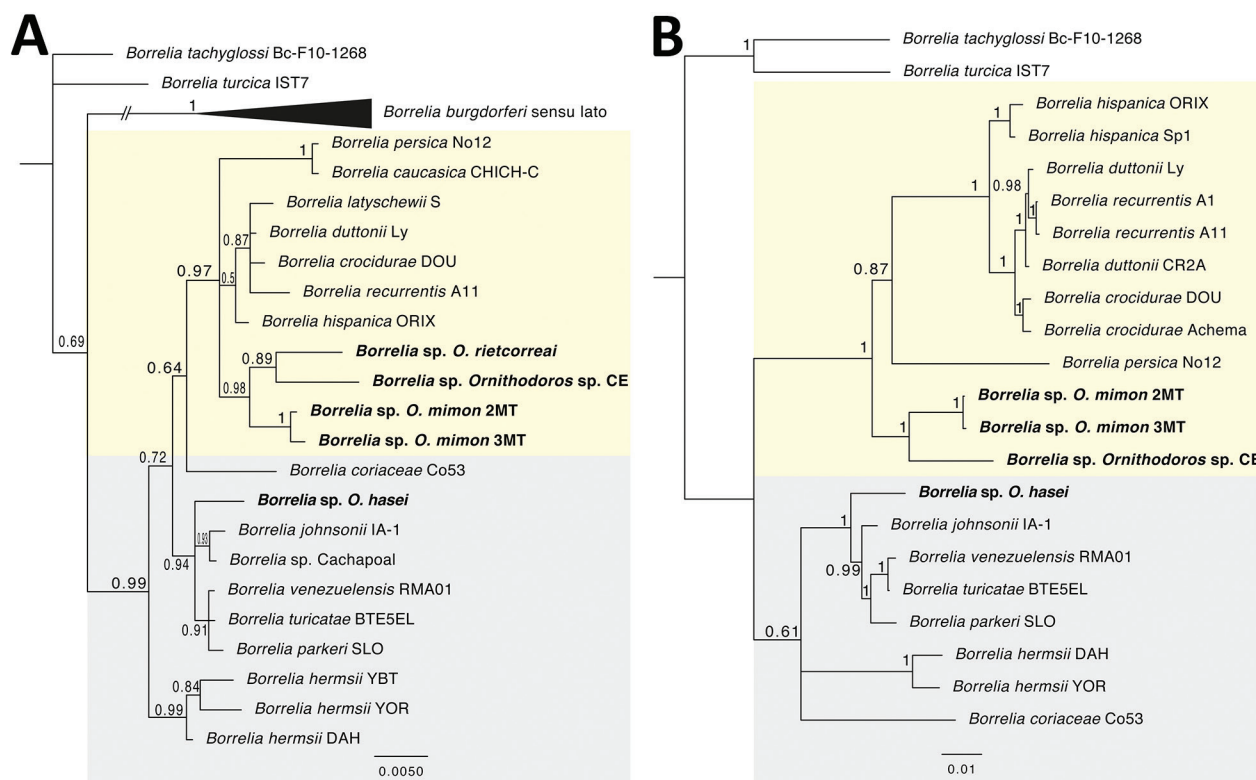


Figure. Bayesian phylogenetic trees inferred for the *Borrelia* spp. characterized in study of relapsing fever group borreliae in human-biting soft ticks, Brazil. A) Ambiguous alignments of single 16S rRNA gene (1,274 bp); B) concatenated 16S rRNA-*flaB*-*glpQ* genes (2,435 bp). Bold indicates borreliae from this study. Trees are drawn to scale. Four independent Markov chain runs for 1,000,000 metropolis-coupled MCMC generations were implemented for the analyses, sampling a tree every 100th generation. The first 25% of the trees represented burn-in, and the remaining trees were used to calculate Bayesian posterior probability values. Both trees were inferred using the Hasegawa-Kishino-Yano model with gamma distribution. Numbers above or below tree branches represent Bayesian posterior probabilities. Light yellow and gray backgrounds denote Old World and New World relapsing fever group *Borrelia* spp., respectively. Scale bar indicates nucleotide substitutions per site.

investigation of TBRF as a possible cause of UFS in Brazil. It is known that antibodies of patients exposed to RFGB infection cross-react in serologic tests for the diagnosis of Lyme borreliosis (9). This cross-reactivity is particularly relevant in Brazil because serologic evidence for an alleged Lyme-like disease in humans has been reiteratively published, yet refuted (10), and TBRF has not yet been considered as a possible cause of such disease.

Acknowledgments

We thank Gilson Miranda from Serra das Almas Natural Reserve for his assistance during our visit to this location; Jerônimo Carvalho Martins from Jericoacoara National Park for gently allowing us the use of their facilities at Jericoacoara village; and Adriane Suzin, Ana Carolina Souza, Flavio A. Terassini, Ivaneide N. Costa, Luís Marcelo A. Camargo, Romilson S. Lopes Jr., Simone D. Tojal, and Vinicius S. Rodrigues for their valuable assistance in field work.

Laboratory work was funded by Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP), and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq, grant no. 09262/2018-8). S.M.L. was funded by FAPESP (grant nos. 2018/02521-1 and 2019/17960-3).

About the Author

Dr. Muñoz-Leal is a veterinarian affiliated with Universidad de Concepción, Chillán, Chile, who specializes in tickborne diseases. His research is focused on soft ticks and tickborne relapsing fever in South America.

References

1. Lopez J, Krishnavahjla A, Garcia M, Bermudez S. Tick-borne relapsing fever spirochetes in the Americas. *Vet Sci.* 2016;3:16. <https://doi.org/10.3390/vetsci3030016>
2. de Oliveira SV, Bitencourth K, Borsoi ABP, de Freitas FSS, Coelho GCB, Amorim M, et al. Human parasitism and toxicosis by *Ornithodoros rietcorrei* (Acari: Argasidae) in

- an urban area of Northeastern Brazil. *Ticks Tick Borne Dis.* 2018;9:1494–8. <https://doi.org/10.1016/j.ttbdis.2018.07.011>
3. Labruna MB, Marcili A, Ogrzewalska M, Barros-Battesti DM, Dantas-Torres F, Fernandes AA, et al. New records and human parasitism by *Ornithodoros mimon* (Acari: Argasidae) in Brazil. *J Med Entomol.* 2014;51:283–7. <https://doi.org/10.1603/ME13062>
 4. Muñoz-Leal S, Faccini-Martínez AA, Costa FB, Marcili A, Mesquita ETKC, Marques EP, et al. Isolation and molecular characterization of a relapsing fever *Borrelia* recovered from *Ornithodoros rudi*s in Brazil. *Ticks Tick Borne Dis.* 2018;9:864–71. <https://doi.org/10.1016/j.ttbdis.2018.03.008>
 5. Parola P, Diatta G, Socolovschi C, Mediannikov O, Tall A, Bassene H, et al. Tick-borne relapsing fever borreliosis, rural Senegal. *Emerg Infect Dis.* 2011;17:883–5. <https://doi.org/10.3201/eid1705.100573>
 6. Chikeka I, Dumler JS. Neglected bacterial zoonoses. *Clin Microbiol Infect.* 2015;21:404–15. <https://doi.org/10.1016/j.cmi.2015.04.022>
 7. Doltario AB, Menon LJB, Bollela VR, Martinez R, de Almeida E Araújo DC, da Fonseca BAL, et al. Malaria and other febrile diseases among travellers: the experience of a reference centre located outside the Brazilian Amazon Region. *Malar J.* 2016;15:294. <https://doi.org/10.1186/s12936-016-1347-x>
 8. Moreira J, Bressan CS, Brasil P, Siqueira AM. Epidemiology of acute febrile illness in Latin America. *Clin Microbiol Infect.* 2018;24:827–35. <https://doi.org/10.1016/j.cmi.2018.05.001>
 9. Rath PM, Fehrenbach FJ, Rögler G, Pohle HD, Schönberg A. Relapsing fever and its serological discrimination from Lyme borreliosis. *Infection.* 1992;20:283–6. <https://doi.org/10.1007/BF01710797>
 10. de Oliveira SV, Faccini-Martínez AA, Cerutti Junior C. Lack of serological evidence for Lyme-like borreliosis in Brazil. *Travel Med Infect Dis.* 2018;26:62–3. <https://doi.org/10.1016/j.tmaid.2018.04.005>

Address for correspondence: Sebastián Muñoz-Leal, Departamento de Patología y Medicina Preventiva, Facultad de Ciencias Veterinarias, Universidad de Concepción, Av. Vicente Méndez 595-Casilla 537, Chillán-Región de Ñuble, Chile; email: sebamunoz@udec.cl

Etiology of Severe Acute Respiratory Infections, Bangladesh, 2017

Md R. Rahaman, Karen A. Alroy, Chris A. Van Beneden, Michael S. Friedman, Erin D. Kennedy, Mahmudur Rahman, Arunmozhi Balajee, A.K.M. Muraduzzaman, Tahmina Shirin, Meerjady S. Flora, Eduardo Azziz-Baumgartner

Author affiliations: The University of Adelaide, Adelaide, South Australia, Australia (M.R. Rahaman); Institute of Epidemiology, Disease Control, and Research, Dhaka (M.R. Rahaman, A.K.M. Muraduzzaman, T. Shirin, M.S. Flora); Centers for Disease Control and Prevention, Atlanta, Georgia, USA (K.A. Alroy, C.A. Van Beneden, M.S. Friedman, E.D. Kennedy, A. Balajee, E. Azziz-Baumgartner) icddr,b, Dhaka, Bangladesh (M.R. Rahman);

DOI: <https://doi.org/10.3201/eid2701.201347>

In April 2017, surveillance detected a surge in severe acute respiratory infections (SARI) in Bangladesh. We collected specimens from SARI patients and asymptomatic controls for analysis with multipathogen diagnostic tests. Influenza A(H1N1)pdm09 was associated with the SARI epidemic, suggesting that introducing vaccines and empiric antiviral drugs could be beneficial.

In April 2017, the Institute of Epidemiology Disease Control and Research (IEDCR) in Bangladesh noted an 89% increase in severe acute respiratory infections (SARI) compared with April 2016 through the National Influenza Surveillance Bangladesh (NISB) at 10 tertiary-care hospitals. During April 10–June 21, 2017, we conducted a case-control study to ascertain the cause of the outbreak and its associated risk factors.

We defined a SARI case as acute respiratory illness in a patient within 10 days of onset, with history of fever and cough, and requiring hospitalization (1). We sought to enroll all adults ≥ 18 years of age who were admitted to NISB hospitals with SARI. Staff screened patients for eligibility, obtained written informed consent, surveyed participants about demographics, and took combined nasal and throat swab samples. Patients who died in hospital wards before enrollment were ineligible. Within 2 days of case-patient enrollment, staff enrolled 2 asymptomatic controls, identified by convenience from the same hospitals' outpatient clinics, surveyed them, and took combined nasal and throat swab samples. Patients who had fever or respiratory symptoms in the previous 14 days were ineligible to serve as controls. Swab

Relapsing Fever Group *Borreliae* in Human-Biting Soft Ticks, Brazil

Appendix

Supplemental Methods

Collected ticks were morphologically identified following original descriptions and redescrptions of South American species (1–5), and by comparisons with specimens deposited in the tick collection Coleção Nacional de Carrapatos Danilo Gonçalves Saraiva, São Paulo, Brazil. DNA extraction was performed using a phenol chloroform-based protocol (6). Successful extractions were confirmed by amplifying tick mitochondrial 16S rRNA gene with primers 16S+1 and 16S–1 in all the samples (7). Amplicons of this locus were sequenced only for soft ticks collected in new localities for the country, confirming their morphological identities by a phylogenetic analysis (data not shown). Amplicons of expected size for tick mitochondrial 16S rRNA gene and *Borrelia* 16S rRNA, *flab*, and *glpQ* genes were treated with Illustra ExoproStar (GE Healthcare, <https://www.gehealthcare.com>) and sequenced using an ABI 3500 genetic analyzer with a BigDye Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, <https://www.thermofisher.com>). Sequences were assembled and analyzed using Geneious R9 (8). Quality values (Q) for base calls were scaled between Q20 (error probability of 1 in 100) and Q40 (error probability of 1 in 10,000). Scores <Q20 in the 5' and 3' ends of each sequence were automatically trimmed. Values of quality, coverage, and length are shown in Appendix Table 2. BLASTn analyses were performed to infer most identical sequences in GenBank (<https://blast.ncbi.nlm.nih.gov/Blast>). Bayesian phylogenetic trees (<http://mrbayes.csit.fsu.edu/>) were inferred for single (*Borrelia* 16S rRNA) and concatenated (*Borrelia* 16S rRNA-*flaB*-*glpQ*) gene alignments constructed with MAFFT (<https://mafft.cbrc.jp/alignment/server>).

References

1. Labruna MB, Nava S, Marcili A, Barbieri ARM, Nunes PH, Horta MC, et al. A new argasid tick species (Acari: Argasidae) associated with the rock cavy, *Kerodon rupestris* Wied-Neuwied

- (Rodentia: Caviidae), in a semiarid region of Brazil. *Parasit Vectors*. 2016;9:511. [PubMed](#)
<https://doi.org/10.1186/s13071-016-1796-7>
2. Labruna MB, Nava S, Terassini FA, Onofrio VC, Barros-Battesti DM, Camargo LMA, et al. Description of adults and nymph, and redescription of the larva, of *Ornithodoros marinkellei* (Acari: Argasidae), with data on its phylogenetic position. *J Parasitol*. 2011;97:207–17.
<https://doi.org/10.1645/GE-2620.1>
 3. Labruna M, Venzal J. *Carios fonsecai* sp. nov. (Acari, Argasidae), a bat tick from the central-western region of Brazil. *Acta Parasitol*. 2009;54:355–63 <https://doi.org/10.2478/s11686-009-0051-1>
 4. Barros-Battesti D, Onofrio V, Nieri-Bastos F, Soares J, Marcili A, Famadas K, et al. *Ornithodoros brasiliensis* Aragão (Acari: Argasidae): description of the larva, redescription of male and female, and neotype designation. *Zootaxa*. 2012;3178:22. <https://doi.org/10.11646/zootaxa.3178.1.2>
 5. Cooley RA, Kohls GM. The Argasidae of North America, Central America and Cuba. The American Midland Naturalist Monograph No. 1. South Bend (IN): Notre Dame University Press; 1944. 152 p.
 6. Sangioni LA, Horta MC, Vianna MCB, Gennari SM, Soares RM, Galvão MAM, et al. Rickettsial infection in animals and Brazilian spotted fever endemicity. *Emerg Infect Dis*. 2005;11:265–70.
[PubMed](#) <https://doi.org/10.3201/eid1102.040656>
 7. Mangold AJ, Bargues MD, Mas-Coma S. Mitochondrial 16S rDNA sequences and phylogenetic relationships of species of *Rhipicephalus* and other tick genera among Metastriata (Acari: Ixodidae). *Parasitol Res*. 1998;84:478–84. [PubMed](#) <https://doi.org/10.1007/s004360050433>
 8. Kearse M, Moir R, Wilson A, Stones-Havas S, Cheung M, Sturrock S, et al. Geneious Basic: an integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics*. 2012;28:1647–9. [PubMed](#)
<https://doi.org/10.1093/bioinformatics/bts199>
 9. Schwan TG, Raffel SJ, Schruppf ME, Policastro PF, Rawlings JA, Lane RS, et al. Phylogenetic analysis of the spirochetes *Borrelia parkeri* and *Borrelia turicatae* and the potential for tick-borne relapsing fever in Florida. *J Clin Microbiol*. 2005;43:3851–9. [PubMed](#)
<https://doi.org/10.1128/JCM.43.8.3851-3859.2005>
 10. Stromdahl EY, Williamson PC, Kollars TMJ Jr, Evans SR, Barry RK, Vince MA, et al. Evidence of *Borrelia lonestari* DNA in *Amblyomma americanum* (Acari: Ixodidae) removed from humans. *J Clin Microbiol*. 2003;41:5557–62. [PubMed](#) <https://doi.org/10.1128/JCM.41.12.5557-5562.2003>

Appendix Table 1. Results of molecular survey for *Borrelia* spp. in 8 *Ornithodoros* species from different parts of Brazil

Species	Site of collection	Locality/state	No. tested specimens (N, M, F)	Pools/individual ticks	Real-time PCR positive samples (%)	GenBank accession no., 16S	GenBank accession no., <i>flaB</i>	GenBank accession no., <i>gfpQ</i>
<i>O. rudis</i>	Abandoned bird nests in hollow palm trees	Araguapaz/GO	82 (15N, 35M, 32F)	Pools: 15N; 17M; 18M; 15F; 17F	0	None	None	None
<i>O. mimon</i> *	Abandoned bird nests in hollow palm trees	Formosa da Serra Negra/MA	22 (11N, 7M, 4F)	Pools: 11N; 7M; 4F	0	None	None	None
	House	Cuiabá/MT	19 (8N, 9M, 2F)	Individual ticks	1F, 1N (10.5)	MT013211, MT013212	MT076262	MT076265
<i>O. hasei</i> *	Cave	Jericoacoara National Park/CE	129 (48N, 51M, 20F)	Pools: 25N; 23N; 25M; 26M; 20F	23N pool (0.05†)	MT013210	MT076261	MT076264
<i>O. rietcorrei</i> *	Between rocks	Serra das Almas Natural Reserve/CE	111 (75N, 23M, 13F)	Pools: 40N; 35N; 23M; 13F	13F pool (0.1†)	MT013213	None	None
<i>Ornithodoros</i> sp.*	Between rocks	Serra das Almas Natural Reserve/CE	39 (6N, 27M, 6F)	Pools: 6N; 27M; 6F	6F pool (0.2†)	MT013214	MT076263	MT076266
<i>O. rostratus</i>	Sandy soil	Fazenda São José/MS	171 (121N, 40M, 10F)	Individual ticks	0	None	None	None
<i>O. marinkellei</i>	Cave	Porto Velho/RO	68 (22M, 46F)	Individual ticks	0	None	None	None
<i>O. fonsecai</i> *	Cave	Furna do Araticum/CE	34 (22M, 12F)	Individual ticks	0	None	None	None

*Tick with mitochondrial 16S rDNA sequence generated in this study (GenBank accession nos. MT021429, MT021430, MT021431, MT021432, MT021433, MT021434, MT021435). F, female; M, male; N, nymph.

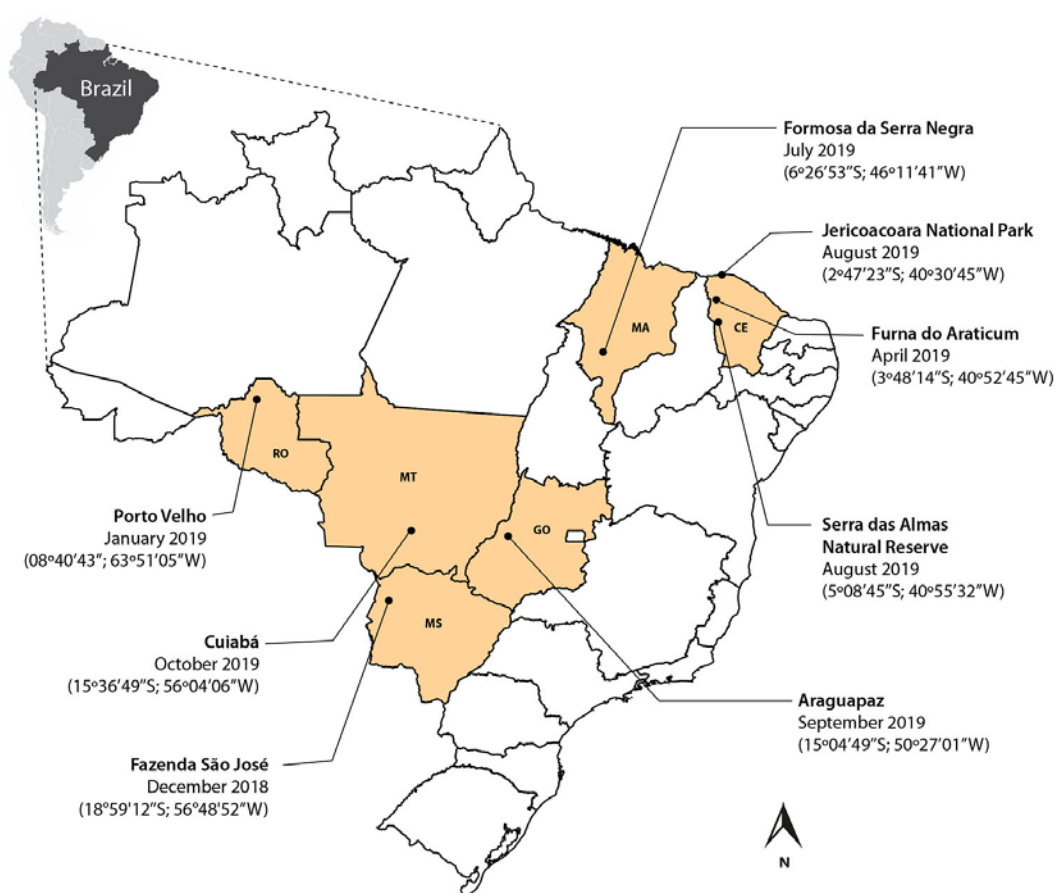
†Minimal infection rate.

The following primers (forward/reverse) were used for PCR (‡) and sequencing (§) *Borrelia* genes: 16S rRNA gene: FD3/T50‡§, 16S-1/16S-2§, Rec4/Rec9§ (9); *flaB* gene: FLA LL/FLA LS‡§ (10); *gfpQ* gene: gfpQ F+1/ Rev-2‡§, Rev-1/2gfpQ F-1§ (9).

Appendix Table 2. Coverage (mean $\pm$ standard deviation), quality values (% of base calls matching Q20, Q30, and Q40 values, discounting trimmed bases), and length (base pairs, bp) for the sequences of *Borrelia* 16S rRNA, *flaB*, and *glpQ* genes obtained in this study

Genes obtained in this study			Quality			
Tick species/ <i>Borrelia</i> sp.	Gene	Coverage	Q20	Q30	Q40	Trimmed length
<i>Ornithodoros mimon</i>						
<i>Borrelia</i> sp. Omi2MT	16S rRNA*	2.7 ± 0.8	92.9	90.6	87.6	1,342 bp
	flaB	5.5 ± 1.2	93.2	90.3	88.1	504 bp
	glpQ	3.3 ± 0.6	96.7	93.5	85.5	447 bp
<i>Borrelia</i> sp. Omi3MT	16S rRNA*	2.8 ± 0.8	94.1	92.4	89.1	1,342 bp
	flaB	5.5 ± 1.2	93.2	90.3	88.1	504 bp
	glpQ	3.3 ± 0.6	96.7	93.5	85.5	447 bp
<i>Ornithodoros rietcorraei</i>						
<i>Borrelia</i> sp. OrietCE	16S rRNA	2.8 ± 0.6	94.3	92.4	90.5	1,326 bp
<i>Ornithodoros hasei</i>						
<i>Borrelia</i> sp. JericoCE	16S rRNA	2.1 ± 0.9	95.5	90.8	85.9	1,345 bp
	flaB	3.1 ± 1.2	92.5	90.1	88.9	674 bp
	glpQ	2.9 ± 0.8	94.3	92.3	86.3	715 bp
<i>Ornithodoros</i> sp. CE						
<i>Borrelia</i> sp. Tabajara CE	16S rRNA					
	flaB	2.2 ± 1.1	98.6	96.8	95.5	614 bp
	glpQ	3.9 ± 0.8	96.4	94.7	92.4	598 bp
*Haplotypes for 16S rRNA gene of <i>Borrelia</i> sp. Omi2MT and <i>Borrelia</i> sp. Omi3MT differed in a single nucleotide with unambiguous base calls in each sequence (1341/1342 bp, 99.93% of identity).						

*Haplotypes for 16S rRNA gene of *Borrelia* sp. Omi2MT and *Borrelia* sp. Omi3MT differed in a single nucleotide with unambiguous base calls in each sequence (1341/1342 bp, 99.93% of identity).



Appendix Figure. Map of Brazil showing the dates and localities where collection of ticks was performed. CE, Ceará State; GO, Goiás State; MA, Maranhão State; MS, Mato Grosso do Sul State; MT, Mato Grosso State; RO, Rondônia State.