Bourbon Virus in Wild and Domestic Animals, Missouri, USA, 2012–2013

Katelin C. Jackson,1 Thomas Gidlewski, J. Jeffrey Root, Angela M. Bosco-Lauth, R. Ryan Lash, Jessica R. Harmon, Aaron C. Brault, Nicholas A. Panella, William L. Nicholson, Nicholas Komar

Author affiliations: Centers for Disease Control and Prevention, Fort Collins, Colorado, USA (K.C. Jackson, A.C. Brault, N.A. Panella, N. Komar); US Department of Agriculture, Fort Collins (T. Gidlewski, J.J. Root); Colorado State University, Fort Collins (A.M. Bosco-Lauth); Centers for Disease Control and Prevention, Atlanta, Georgia, USA (R.R. Lash, J.R. Harmon, W.L. Nicholson)

DOI: https://doi.org/10.32301/eid2510.181902

Since its recent discovery, Bourbon virus has been isolated from a human and ticks. To assess exposure of potential vertebrate reservoirs, we assayed banked serum and plasma samples from wildlife and domestic animals in Missouri, USA, for Bourbon virus–neutralizing antibodies. We detected high seroprevalence in raccoons (50%) and white-tailed deer (86%).

Bourbon virus (BRBV) was first isolated from a febrile patient with a history of tick bites in Bourbon County, Kansas, USA; the patient later died from severe illness in 2014 (1). Several additional human BRBV infections were reported subsequently from the midwestern and southern United States (2). BRBV belongs to the family Orthomyxoviridae, genus Thogotovirus, which is distributed worldwide and includes Araguaui, Aransas Bay, Dhori, Jos, Thogoto, and Upolu viruses (1,3). Thogoto and Dhori viruses have been associated with human disease (4–6). Viruses within the genus Thogotovirus have been associated with hard or soft ticks (7). Recent studies suggest that the lone star tick (Amblyomma americanum) is involved with BRBV transmission (2,3,8). These ticks feed primarily on mammals, which might play a role in BRBV ecology.

We evaluated banked animal serum and plasma for evidence of BRBV infection by using the plaque-reduction neutralization test (PRNT) to detect BRBV-reactive antibodies. We tested specimens of white-tailed deer (Odocoileus virginianus), raccoon (Procyon lotor), Virginia opossum (Didelphis virginiana), and various other mammals and birds from northwest Missouri, USA, for neutralizing antibodies against BRBV to identify naturally exposed host species and to implicate potential zoonotic amplifiers.

We collected specimens from wild and domestic vertebrates as described (9). We performed PRNTs on serum and plasma samples by using Vero cell culture as described (9). In brief, we initially screened samples by diluting them 1:5 and mixing them with an equal amount of BRBV suspension containing ≥100 PFUs/0.1 mL. Samples that showed ≥70% reduction of plaques were confirmed by serial 2-fold titration in duplicate from serum dilutions of 1:10–1:320. We considered 70% PRNT titers ≥10 as positive.

We screened serum and plasma samples from 301 birds and mammals for BRBV-neutralizing antibodies. A total of 48 (30.8%) of 156 mammalian serum samples were positive at the 70% neutralization level (Table). Mammals with evidence of past infection included domestic dogs, eastern cottontail, horse, raccoon, and white-tailed deer. None of 26 avian species were seropositive (Appendix Table, https://wwwnc.cdc.gov/EID/article/25/9/18-1902-App1.pdf).

BRBV is probably transmitted to humans and other vertebrates by the lone star tick, an abundant arthropod in the southeastern United States (8). This virus was cultured from these ticks in northwestern Missouri in 2013 and eastern Kansas in 2015 (2,8). Our results indicated that mammals are frequently exposed to BRBV. This finding was expected because lone star ticks feed primarily on mammals, and rarely on birds. Our study corroborates that birds are not involved in BRBV transmission, and our data establish that the vertebrate host range for infection now includes ≥5 mammalian species, 2 of which are domestic animals (dogs and horses). Of the wildlife species, the seropositivity rate for white-tailed deer was high (86%), whereas Virginia opossums, despite a moderate sample size (n = 28), showed no evidence of virus exposure. Deer and raccoons (seroprevalence 50%) could be useful wildlife sentinels for tracking the geographic distribution of BRBV. Dogs (seroprevalence 15%) and horses (seroprevalence 4%) merit further consideration among domestic animals for use as sentinels for either tracking virus activity or as an early warning system for mitigation of human risk. Because of limited sampling, we observed no statistically significant difference in seroprevalence between these 2 species.

A limitation of our study was small sample sizes, which reduces the accuracy of the seroprevalence measurements. Furthermore, serologic data provide indirect evidence of virus infection, rather than the detection of the virus itself or its parts (i.e., antigen or nucleic acid). However, a closely related congener could exist and generate cross-reactive antibodies to BRBV, causing false-positive results in our assay. Nevertheless, the PRNT is generally considered the standard for serologic assays.

In conclusion, we have demonstrated that nonhuman vertebrates are exposed to BRBV. These findings are useful for future public health efforts and to better understand the ecology of BRBV. Specifically, we identified 2 candidate wildlife sentinels and potential domestic sentinels for tracking...
and possible early warning of BRBV transmission risk. However, whether any of these mammalian species are competent amplifier hosts for BRBV remains to be determined.

Acknowledgments
We thank the property owners who granted access to their properties; the Missouri Departments of Health and Senior Services and Conservation, the Andrew County Health Department and participating veterinary clinics for providing assistance; David Ashley for providing laboratory space for specimen processing at Missouri Western State University; Luke Miller, Nathan Hubbard, and Sonja Weiss for assisting with mammal trapping; and Jason Velez for preparing Vero cell cultures for the neutralization assays performed.

About the Author
During this study, Ms. Jackson was an intern at the Division of Vector-Borne Diseases, National Center for Emerging and Zoonotic Infectious Diseases, Centers for Disease Control and Prevention, Fort Collins CO. She is currently a doctoral candidate at Washington State University, Pullman, WA. Her research interests include a One Health approach to zoonotic infectious diseases.

References
1. Kosoy OI, Lambert AJ, Hawkinson DJ, Pastula DM, Goldsmith CS, Hunt DC, et al. Novel thogotovirus associated with febrile illness and death, United States, 2014. Emerg Infect Dis. 2015;21:760–4. https://doi.org/10.3201/eid2105.150150

2. Savage HM, Burkhalter KL, Godsey MS Jr, Panella NA, Ashley DC, Nicholson WL, et al. Bourbon virus in field-collected ticks, Missouri, USA. Emerg Infect Dis. 2017;23:2017–22. https://doi.org/10.3201/eid2312.170532

3. Lambert AJ, Velez JO, Brault AC, Bell-Sakyi L, Bosco-Lauth AM, et al. Molecular, serological and in vitro culture-based characterization of Bourbon virus, a newly described human pathogen of the genus Thogotovirus. J Clin Virol. 2015;73:127–32. https://doi.org/10.1016/j.jcv.2015.10.021

4. Butenko AM, Leshchinskaia EV, Semashko IV, Donets MA, Mart'ianova LI. Dhori virus—a causative agent of human disease: 5 cases of laboratory infection [in Russian]. Vopr Virusol. 1987;32:724–9.

5. Filipe AR, Calisher CH, Lazuick J. Antibodies to Congo-Crimean hemorrhagic fever, Dhori, Thogoto and Bhanja viruses in southern Portugal. Acta Virol. 1985;29:324–8.

6. Frese M, Kochs G, Meier-Dieter U, Siebler J, Haller O. Human MxA protein inhibits tick-borne Thogoto virus but not Dhori virus. J Virol. 1995;69:3904–9.

7. Hubálek Z, Rudolf I. Tick-borne viruses in Europe. Parasitol Res. 2012;111:9–36. https://doi.org/10.1007/s00436-012-2910-1

8. Savage HM, Godsey MS Jr, Tatman J, Burkhalter KL, Hamm A, Panella NA, et al. Surveillance for Heartland and Bourbon viruses in eastern Kansas, June 2016. J Med Entomol. 2018;55:1613–6. https://doi.org/10.1093/jme/tjy103

9. Bosco-Lauth AM, Panella NA, Root JJ, Gidlewski T, Lash RR, Harmon JR, et al. Serological investigation of Heartland virus (Bunyaviridae: Phlebovirus) exposure in wild and domestic animals adjacent to human case sites in Missouri 2012–2013. Am J Trop Med Hyg. 2015;92:1163–7. https://doi.org/10.4269/ajtmh.14-0702

Address for correspondence: Nicholas Komar, Centers for Disease Control and Prevention, 3156 Rampart Rd, Fort Collins, CO 80521, USA; email: nck6@cdc.gov

Fatal Case of Lassa Fever, Bangolo District, Côte d’Ivoire, 2015

Mathieu Mateo, Caroline Picard, Yahaya Sylla, Emilie Kamo, Danielle Odegué, Alexandra Journeaux, Stéphane Kouassi Kan, Marcelle Money, David N’Golo Coulibaly, Eugène Koffi, Souleymane Meite, Véronique Akran, Hervé Kadjo, Edgard Adjogoua, Solange N’Gaza Kokou, Sylvain Baize, Mireille Dosso

Author affiliations: Institut Pasteur, Lyon, France (M. Mateo, C. Picard, A. Journeaux, S. Baize); Centre International de Recherche en Infectiologie, Lyon (M. Mateo, C. Picard, A. Journeaux, S. Baize); Institut Pasteur de Côte d’Ivoire, Abidjan, Côte d’Ivoire (Y. Sylla, E. Kamo, D. Odegué, S.K. Kan, M. Money, D. N’Golo Coulibaly, E. Koffi, S. Meite, V. Akran, H. Kadjo, E. Adjogoua, S. N’Gaza Kokou, S. Baize, M. Dosso)

DOI: https://doi.org/10.3201/eid2509.190239

Table. PRNT results for mammals tested for Bourbon virus–neutralizing antibodies, Missouri, USA, 2012–2013

| Common name | Species name | No. positive/no. tested | Titer | Proportion positive (95% CI) |
|-------------|--------------|------------------------|-------|-----------------------------|
| Domestic cat | Felis catus | 0/2 | <10 | 0 (0.0–0.66) |
| Domestic dog | Canis lupus familiaris | 2/13 | 10–320 | 0.15 (0.04–0.42) |
| Eastern cottontail | Sylvilagus floridanus | 2/9 | >320 | 0.22 (0.06–0.55) |
| Fox squirrel | Sciurus niger | 0/4 | <10 | 0 (0.0–0.49) |
| Horse | Equus caballus | 1/24 | 20 | 0.04 (0.007–0.20) |
| Raccoon | Procyon lotor | 31/62 | 10–320 | 0.50 (0.38–0.62) |
| Virginia opossum | Didelphis virginiana | 0/28 | <10 | 0 (0.0–0.12) |
| White-tailed deer | Odocoileus virginianus | 12/14 | 10–320 | 0.86 (0.60–0.96) |

PRNT, 70% plaque reduction neutralization titer.
# Appendix

**Appendix Table.** Bird species tested for antibodies to Bourbon virus, Missouri, USA, 2012–2013

| Common name             | Species name                | No. positive/no. tested |
|-------------------------|-----------------------------|-------------------------|
| American goldfinch      | Spinus tristis              | 0/16                    |
| Baltimore oriole        | Icterus galbulus            | 0/2                     |
| Black-capped chickadee  | Poecile atricapillus        | 0/13                    |
| Carolina wren           | Thryothorus ludovicianus    | 0/4                     |
| Chipping sparrow        | Spizella passerina          | 0/1                     |
| Common yellowthroat     | Geothlypis trichas          | 0/1                     |
| Downy woodpecker        | Dryobates pubescens         | 0/6                     |
| Eastern bluebird        | Sialia sialis               | 0/1                     |
| Eastern towhee          | Pipilo erythrphthalmus      | 0/1                     |
| Eastern wood-pewee      | Contopus virens             | 0/5                     |
| Field sparrow           | Spizella pusilla            | 0/1                     |
| Gray catbird            | Dumetella carolinensis      | 0/1                     |
| Hairy woodpecker        | Dryobates villosus          | 0/3                     |
| House sparrow           | Passer domesticus           | 0/14                    |
| House wren              | Troglodytes aedon           | 0/1                     |
| Indigo bunting          | Passerina cyanea            | 0/7                     |
| Least flycatcher        | Empidonax minimus           | 0/1                     |
| Mourning dove           | Zenaida macroura            | 0/15                    |
| Northern cardinal       | Cardinalis                  | 0/24                    |
| Red-bellied woodpecker  | Melanerpes carolinus        | 0/1                     |
| Red-eyed vireo          | Vireo olivaceus             | 0/4                     |
| Rose-breasted grosbeak  | Pheucticus ludovicianus     | 0/1                     |
| Summer tanager          | Piranga rubra               | 0/4                     |
| Tufted titmouse         | Baeolophus bicolor          | 0/7                     |
| White-breasted nuthatch | Sitta carolinensis          | 0/4                     |
| Wild turkey             | Gallopavo meleagris         | 0/7                     |
| Total                   | 26 species                  | 0/145                   |