

Acknowledgments

We thank the staff of the slaughterhouses for their help in collecting ticks from cattle. We thank Gregory Mollé, Laurence Thirion, Pierre Combe, and Cecile Baronti for the production and validation of diagnostic reagents.

This work was supported in part by the VHFMODRAD project (no. 823666, call H2020-JTI-IMI2-2015-08-single-stage) of the H2020 program by the Innovative Health Initiative (IHI) agency of the European Commission; by the European Commission European Virus Archive Global project (EVA GLOBAL, grant agreement no. 871029) of the Horizon 2020 Research and Innovation Programme; and by Centre de Coopération Internationale en Recherche Agronomique pour le Développement (provision of service no. E2F03F6F). The reagent material was provided by the European Virus Archive-Marseille (EVAM) under the label technological platforms of Aix-Marseille University.

About the Author

Ms. Kiwan is a PhD student at the Unité des Virus Emergents, University of Corsica Pascal Paoli and Aix-Marseille University, France. Her primary research interests focus on tickborne viruses via a One Health approach.

References

1. Lorenzo Juanes HM, Carbonell C, Sendra BF, López-Bernus A, Bahamonde A, Orfao A, et al. Crimean-Congo hemorrhagic fever, Spain, 2013–2021. *Emerg Infect Dis.* 2023;29:252–9. <https://doi.org/10.3201/eid2902.220677>
2. Sánchez-Seco MP, Sierra MJ, Estrada-Peña A, Valcárcel F, Molina R, de Arellano ER, et al.; Group for CCHFv Research. Widespread detection of multiple strains of Crimean-Congo hemorrhagic fever virus in ticks, Spain. *Emerg Infect Dis.* 2021;28:394–402. <https://doi.org/10.3201/eid2802.211308>
3. Grech-Angelini S, Lancelot R, Ferraris O, Peyrefitte CN, Vachieri N, Pédarrieu A, et al. Crimean-Congo hemorrhagic fever virus antibodies among livestock on Corsica, France, 2014–2016. *Emerg Infect Dis.* 2020;26:1041–4. <https://doi.org/10.3201/10.3201/eid2605.191465>
4. Cicculli V, Maitre A, Ayhan N, Mondoloni S, Paoli JC, Vial L, et al. Lack of evidence for Crimean-Congo hemorrhagic fever virus in ticks collected from animals, Corsica, France. *Emerg Infect Dis.* 2022;28:1035–8. <https://doi.org/10.3201/eid2805.211996>
5. Ninove L, Nougairède A, Gazin C, Thirion L, Delogu I, Zandotti C, et al. RNA and DNA bacteriophages as molecular diagnosis controls in clinical virology: a comprehensive study of more than 45,000 routine PCR tests. *PLoS One.* 2011;6:e16142. <https://doi.org/10.1371/journal.pone.0016142>
6. Estrada-Peña A, D'Amico G, Fernández-Ruiz N. Modelling the potential spread of *Hyalomma marginatum* ticks in Europe by migratory birds. *Int J Parasitol.* 2021;51:1–11. <https://doi.org/10.1016/j.ijpara.2020.08.004>
7. Tamura K, Stecher G, Peterson D, Filipowski A, Kumar S. MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. *Mol Biol Evol.* 2013;30:2725–9. <https://doi.org/10.1093/molbev/mst197>
8. Chamberlain J, Cook N, Lloyd G, Mioulet V, Tolley H, Hewson R. Co-evolutionary patterns of variation in small and large RNA segments of Crimean-Congo hemorrhagic fever virus. *J Gen Virol.* 2005;86:3337–41. <https://doi.org/10.1099/vir.0.81213-0>
9. Carroll SA, Bird BH, Rollin PE, Nichol ST. Ancient common ancestry of Crimean-Congo hemorrhagic fever virus. *Mol Phylogenet Evol.* 2010;55:1103–10. <https://doi.org/10.1016/j.ympev.2010.01.006>
10. Anses – National Agency for Food, Environmental and Occupational Health Safety. Crimean-Congo hemorrhagic fever: first detection of the virus in cattle farms in southern France [in French] [cited 2023 Dec 18]. <https://www.anses.fr/fr/content/fievre-hemorragique-crimee-congo-detection-virus-elevages-bovins>

Address for correspondence: Alessandra Falchi, Unité des Virus Emergents, IRD 190-Inserm 1207, University of Corsica Pascal Paoli and Aix-Marseille University, Laboratoire de Virologie, Faculté des Sciences, Campus Grimaldi, Corte 20250, France; email: falchi_a@univ-corse.fr

Deforestation and Bovine Rabies Outbreaks in Costa Rica, 1985–2020

Christie Jones, Amanda Vicente-Santos, Julie A. Clennon, Thomas R. Gillespie

Author affiliation: Emory University, Atlanta, Georgia, USA

DOI: <https://doi.org/10.3201/eid3005.230927>

In Latin America, rabies virus has persisted in a cycle between *Desmodus rotundus* vampire bats and cattle, potentially enhanced by deforestation. We modeled bovine rabies virus outbreaks in Costa Rica relative to land-use indicators and found spatial-temporal relationships among rabies virus outbreaks with deforestation as a predictor.

Costa Rica has benefited from effective vaccination campaigns to eliminate canine rabies virus infections. Still, the virus has endured, spread by vampire bats (*Desmodus rotundus*) to cattle, with rare but documented transfer from bats to humans (1,2). To determine how anthropogenic disturbance affects

rabies virus incidence and risk in this system, we investigated the relationship between land-use change and documented bovine rabies virus outbreaks in Costa Rica during 1985–2020.

Since 1985, the National Animal Health Service of Costa Rica (SENASA) has conducted rabies virus surveillance on domestic animals, confirming outbreaks of ≥ 1 cases by using fluorescent antibody testing (3). We mapped bovine rabies outbreaks during 1985–2020 reported by SENASA with neighboring land-use data by using QGIS 3.16.2 (QGIS, <https://qgis.org>). Ten outbreaks from the initial SENASA report ($n = 119$) were removed because of inaccurate location data, leaving 109 outbreaks for our study.

To evaluate outbreak probability and distribution, we used kernel density estimations with the QGIS default bandwidth to create spatial probability estimations on the basis of known outbreaks. We used a kernel function that smoothed and interpolated probabilities across the study area. We used a kernel radius of 10 km, the maximum vampire bat foraging range, limiting interpolation to the determined area (4). We applied a Kulldorff retrospective space-time scan with an elliptical spatial scan by

using SaTScan version 9.7 (SaTScan, <https://www.satscan.org>) to detect the number of outbreak locations in space and time (5).

We applied logistic regression by using a generalized linear mixed model R-package (<https://cran.r-project.org/web/packages/lme4/lme4.pdf>) to evaluate the effects of land-use factors on bovine rabies virus outbreak locations compared with random control locations ($n = 119$). We set the district as a random effect to account for spatial effects. We set the number of control points to match the true number of SENASA-reported outbreaks. We created control locations by using the random points function in QGIS and by using the 2005 and 2017 agricultural land-use data to bind nonoutbreak samples to areas that could house cattle. We matched controls temporally to outbreaks on the basis of the proportion of outbreaks before and after 2006.

For each outbreak and control location, we used district-level human population density and cattle population density as explanatory variables. We used the distance to and area of forest cover from each outbreak and control location within a 10-km buffer of the location. For outbreaks and control events up to

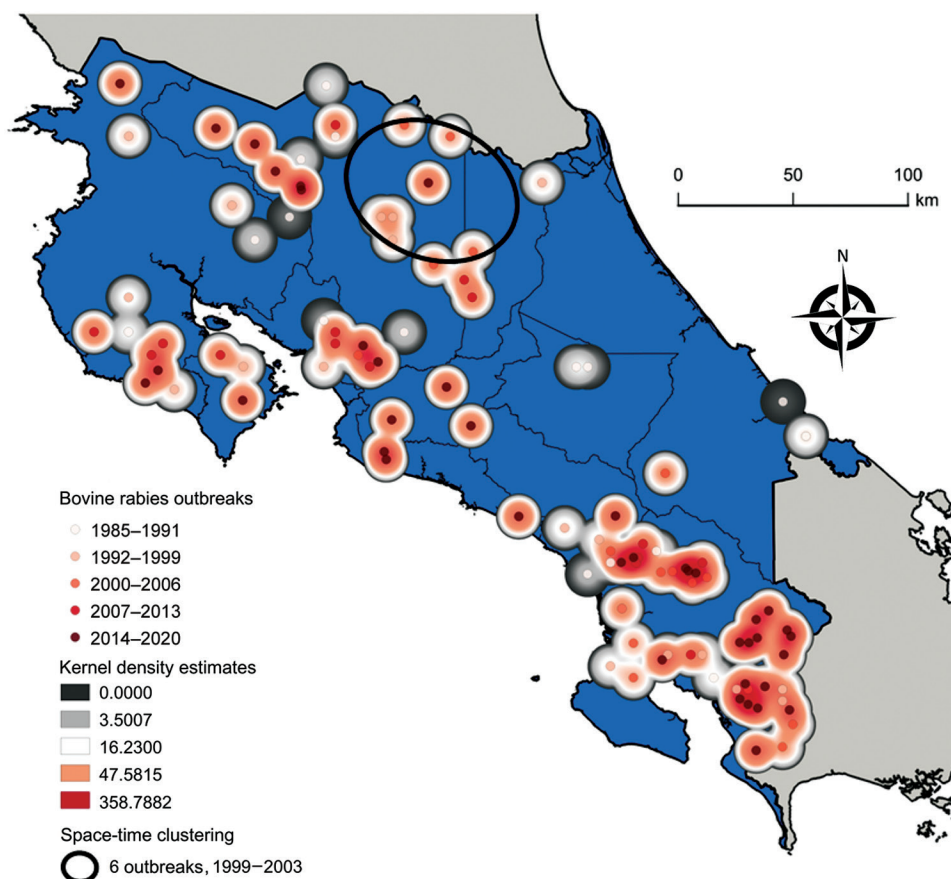


Figure. Kernel density estimations and Kulldorff space-time scan results for 109 bovine rabies outbreaks, Costa Rica, 1985–2020. Scan was limited to a 10-km distance from the epicenter of an outbreak to account for *Desmodus rotundus* vampire bat foraging ranges, enabling the detection of outbreak locations in space and time. Kernel density estimations were interpolated by using GeoDa version 1.18.0 (<http://geodacenter.github.io>), and the Kulldorff scan was implemented in SaTScan (<https://www.satscan.org>). The bovine rabies outbreak data is from the National Animal Health Service of Costa Rica. Map was created by using QGIS version 3.16.2 (<https://qgis.org>).

Table. Statistical relationship between bovine rabies virus outbreaks and relative variables of distance to forested areas, human density, and cattle density, Costa Rica, 1985–2020*

Relative variable	Estimate	SE	t-value	p value
Distance to forest	4.33×10^{-4}	3.32×10^{-1}	1.95	0.05†
Human density	-3.93×10^{-5}	1.93×10^{-5}	-1.53	0.13
Cattle density	-3.76×10^{-5}	3.23×10^{-5}	-0.93	0.35

*Results of a generalized linear mixed model regression analysis used to determine a statistical relationship between deforestation, human and bovine density, and bovine rabies outbreaks using data from the National Animal Health Service of Costa Rica, the 2014 Atlas of Costa Rica aerial photograph, the 2018 National Territorial Information System aerial photograph, population data, and population density estimates based on growth trends. †p-value ≤ 0.05 is considered statistically significant.

2014, we calculated forest cover by using the 2014 aerial photograph from the Atlas of Costa Rica (<http://www.kyriosoft.com/atlas>). For outbreaks after 2014, we used a 2018 aerial photograph from the National Territorial Information System (<https://www.snitcr.go.cr>). We used human population data from 2011 for all detections up to 2011. After 2011, we used a human population density estimate based on national growth trends (6). We used a similar approach for cattle density data based on a dataset from 2014 (7).

Outbreaks occurring in the northern provinces of Alajuela and Heredia clustered on the basis on their statistically significant closeness in both location and time of occurrence (6 outbreaks during 1999–2003; log likelihood ratio 7.52; $p = 0.035$) (Figure). The increased number of outbreaks in southern Puntarenas Province may be because of repeated emergence given the lack of space-time clustering (Figure).

We found a positive association between the distance to forested areas and bovine rabies virus outbreaks (generalized linear mixed model estimate 4.33×10^{-4} , SE 3.32×10^{-1} ; Z-value 1.95; $p = 0.05$) (Table). Each 1-km increase in distance from forested areas increased the probability of an outbreak by 4%. This finding aligns with our understanding of *D. rotundus* bat feeding preferences and rabies virus transmission risk. Decreased forested roosting site proximity appears to increase *D. rotundus* bat feeding behavior on cattle (8). Human and cattle densities were not associated with bovine rabies outbreaks (Table). Because human population data were unavailable until 2011 and cattle population data unavailable until 2014, the effect of those population densities may be skewed because agricultural intensification in Costa Rica has undergone major changes during the study period (9).

Our results show an association between deforestation and bovine rabies virus outbreaks, highlighting the importance of considering negative health effects in risk assessments for forest conversion proposals (10). Our results indicated the southern region of Costa Rica has the highest probability of bovine rabies outbreaks, indicating the need for localized, preventative interventions in the south. On the basis

of recent findings, we must caution against bat culling as a response to this threat, because disrupting bat dispersal in unexpected ways may increase the spread of the rabies virus (2). Because rabies virus remains endemic in Latin America, an increased focus on integrating spatial, dietary, and surveillance data for *D. rotundus* bats is needed to provide additional insights into land-use effects on the persistence and spread of the rabies virus.

Acknowledgments

We thank Mariano Arroyo of the National Animal Health Services of Costa Rica for data access.

About the Author

Ms. Jones is a recent graduate of the joint Environmental Sciences and Environmental Health Bachelor of Science–Masters of Public Health Program at Emory University, Atlanta, Georgia. Her primary research interests are the application of one health and planetary health approaches to mitigating zoonoses.

References

1. Badilla X, Pérez-Herra V, Quirós L, Morice A, Jiménez E, Sáenz E, et al. Human rabies: a reemerging disease in Costa Rica? *Emerg Infect Dis.* 2003;9:721–3. <https://doi.org/10.3201/eid0906.020632>
2. Viana M, Benavides JA, Broos A, Ibañez Loayza D, Niño R, Bone J, et al. Effects of culling vampire bats on the spatial spread and spillover of rabies virus. *Sci Adv.* 2023;9:eadd7437. <https://doi.org/10.1126/sciadv.add7437>
3. Hutter SE, Brugger K, Sancho Vargas VH, González R, Aguilar O, León B, et al. Rabies in Costa Rica: documentation of the surveillance program and the endemic situation from 1985 to 2014. *Vector Borne Zoonotic Dis.* 2016;16:334–41. <https://doi.org/10.1089/vbz.2015.1906>
4. Rocha F, Ulloa-Stanojlovic FM, Rabaquim VCV, Fadil P, Pompei JC, Brandão PE, et al. Relations between topography, feeding sites, and foraging behavior of the vampire bat, *Desmodus rotundus*. *J Mammal.* 2020;101:164–71. <https://doi.org/10.1093/jmammal/gyz177>
5. Kulldorff M, Heffernan R, Hartman J, Assunção RM, Mostashari F. Space-time permutation model: a space-time permutation scan statistic for the early detection of disease outbreaks. *PLoS Med.* 2005;2:216–24. <https://doi.org/10.1371/journal.pmed.0020059>
6. National Institute of Statistics and Censuses. District population estimates and projections 2000 to 2025. 2014

- [cited 2023 Jul 1] https://inec.cr/wwwisis/documentos/INEC/Estimaciones%20y%20Proyecciones/Estimaciones_Proyecciones_Distritales_2000-2025_2014.pdf
7. National Institute of Statistics and Censuses. VI National agricultural census: characteristics of farms and producers [in Spanish]. 2015 Jul [cited 2023 Jul 1] https://admin.inec.cr/sites/default/files/media/reagropeccenagro2014-ti-006_6.pdf
 8. Fleischer R, Jones C, Ledezma-Campos P, Czirják GÁ, Sommer S, Gillespie TR, et al. Gut microbial shifts in vampire bats linked to immunity due to changed diet in human disturbed landscapes. *Sci Total Environ*. 2024;907:167815. <https://doi.org/10.1016/j.scitotenv.2023.167815>
 9. Jadin I, Meyfroidt P, Lambin EF. International trade, and land use intensification and spatial reorganization explain Costa Rica's forest transition. *Environ Res Lett*. 2016; 11:035005. <https://doi.org/10.1088/1748-9326/11/3/035005>
 10. Gillespie TR, Jones KE, Dobson AP, Clennon JA, Pascual M. COVID-clarity demands unification of health and environmental policy. *Glob Change Biol*. 2021;27:1319–21. <https://doi.org/10.1111/gcb.15508>

Address for correspondence: Thomas Gillespie, Emory University, 400 Dowman Dr, Ste E510, Atlanta, GA, 30307, USA; email: thomas.gillespie@emory.edu

Novel Patterns in High-Resolution Computed Tomography in Whipple Pneumonia

Hui Li, Jiajia Wu, Xiaojun Mai, Wan Zeng, Shuping Cai, Xiuji Huang, Chunxia Zhou, Jin Li, Qin Jiang, Chunliu Lai, Canmao Xie

Author affiliation: The Seventh Affiliated Hospital, Sun Yat-sen University, Shenzhen, China

DOI: <https://doi.org/10.3201/eid3005.231130>

With the use of metagenomic next-generation sequencing, patients diagnosed with Whipple pneumonia are being increasingly correctly diagnosed. We report a series of 3 cases in China that showed a novel pattern of movable infiltrates and upper lung micronodules. After treatment, the 3 patients recovered, and lung infiltrates resolved.

Whipple pneumonia is a rare, chronic, multi-organ disease, with symptoms including arthritis, diarrhea, and weight loss. Diagnosis is traditionally confirmed by a histologic examination of a small bowel biopsy (1). The causative pathogen is *Tropheryma whipplei* bacteria, initially identified from the aortic valve of an endocarditis patient in 2000 (2). The bacterium was successfully cultured again in 2012 by using a sample of bronchoalveolar lavage fluid (BALF) from a pneumonia patient with an acute pulmonary infection (2). By using special culture systems, laboratories can grow positive staining or immunofluorescence detectable bacteria within a macrophage or fibroblast cell in 40–60 days. Metagenomic next-generation sequencing (mNGS) is a useful tool for diagnosis.

We report 3 patients in China diagnosed with *T. whipplei* pneumonia by using BALF mNGS (Vision Medicals Company, <http://www.visionmedical.com>) screening during July 2021–December 2022. The patients had unique radiologic patterns, including upper lung gathering of micronodules forming a galaxy sign, and slightly movable infiltrates before, during, and after treatment.

Patient 1 was a 46-year-old man with a productive cough and a 5-year history of lung abnormality. His lung lesions gradually increased over time, and we found gathering micronodules forming a galaxy sign on the right upper lung (Appendix Figure 1, <https://wwwnc.cdc.gov/EID/article/30/5/23-1130-App.pdf>). *T. whipplei* bacteria was the only pathogen we recovered from BALF screened by using mNGS.

Patient 2 was a 67-year-old man with progressive dyspnea, productive cough, poor appetite, and weight loss. Repeated high-resolution computed tomography (CT) showed gradual increase of diffused micronodules gathering on the upper right lung for 6 months before diagnosis (Figure, panel A). Lesions in the upper right lung also showed movement. After bronchoscopic examination, *T. whipplei* bacteria was the only pathogen we recovered from BALF. Our histologic examination of the lung biopsy showed increased foamy macrophages within the alveolar space and thickened alveolar septal (Figure, panel B); neutrophils were the predominant cell type seen.

Patient 3 was a 57-year-old man with complaints of cough and chest tightness. We found diffuse ground-glass micronodules in the left upper lung (Appendix Figure 2). We performed mNGS of BALF and found *Cryptococcus* spp. yeast and *T. whipplei* bacteria. We treated the patient with fluconazole. Six months later,