

UCLA

UCLA Previously Published Works

Title

Co-Circulating Monkeypox and Swinepox Viruses, Democratic Republic of the Congo, 2022.

Permalink

<https://escholarship.org/uc/item/0x86d84w>

Journal

Emerging Infectious Diseases, 30(4)

Authors

Kalonji, Thierry

Malembi, Emile

Matela, Jean

et al.

Publication Date

2024-04-01

DOI

10.3201/eid3004.231413

Peer reviewed

Co-Circulating Monkeypox and Swinepox Viruses, Democratic Republic of the Congo, 2022

Thierry Kalonji, Emile Malembi, Jean Paul Matela, Toutou Likafi, Eddy Kinganda-Lusamaki, Emmanuel Hasivirwe Vakaniaki, Nicole A. Hoff, Amuri Aziza, Francisca Muyembe, Joelle Kabamba, Tine Cooreman, Béatrice Nguete, Danae Witte, Ahidjo Ayouba, Nicolas Fernandez-Nuñez, Stijn Roge, Martine Peeters, Sydney Merritt, Steve Ahuka-Mundeke, Eric Delaporte, Elisabeth Pukuta, Joachim Mariën, Eugene Bangwen, Steven Lakin, Charles Lewis, Jeffrey B. Doty, Laurens Liesenborghs, Lisa E. Hensley, Andrea McCollum, Anne W. Rimoin, Jean Jacques Muyembe-Tamfum, Robert Shongo, Didine Kaba, Placide Mbala-Kingebeni

In September 2022, deaths of pigs manifesting pox-like lesions caused by swinepox virus were reported in Tshuapa Province, Democratic Republic of the Congo. Two human mpox cases were found concurrently in the surrounding community. Specific diagnostics and robust sequencing are needed to characterize multiple poxviruses and prevent potential poxvirus transmission.

Until recently, the Democratic Republic of the Congo (DRC), located in an mpox-endemic region, has had the highest global burden of mpox (1). Mpox is caused by the monkeypox virus (MPXV), which belongs to the *Orthopoxvirus* genus of the Poxviridae family. Within Poxviridae, 10 other genera are known to infect animals and reptiles (2), including *Suipoxvirus* (3). The only known species of *Suipoxvirus* causes swinepox, a pox-like disease in pigs.

Two genetically distinct clades of MPXV cause disease in humans. Clade I (formerly Congo Basin clade) is endemic in Central Africa, and outbreaks are thought to result mainly from zoonotic spillover;

squirrels or other small mammals are suspected reservoirs or hosts (4; J. Mariën et al., unpub. data, <https://doi.org/10.21203/rs.3.rs-414280/v2>). Clade II (formerly West African clade) is endemic in West Africa; suspected reservoirs and transmission routes are historically similar to those of clade I. In 2022, MPXV subclade IIb spread beyond West Africa, causing an unprecedented global outbreak, leading the World Health Organization to declare mpox as a Public Health Emergency of International Concern (5).

During 2011–2015, the incidence of MPXV cases in DRC's Tshuapa Province increased compared with the incidence during 1981–1986 in similar geographic areas (6). This observation is not unique, suggesting expanding spillover rates of MPXV clade I into the human population and highlighting the possibility of increasing human-to-human virus transmission (1,4).

Confirmation of suspected MPXV cases in DRC is hindered by logistical difficulties, such as complications involving sample collection and lack of

Author affiliations: Program National Lutte Contre MPX-VHF, Kinshasa, Democratic Republic of the Congo (T. Kalonji, E. Malembi, R. Shongo); Province Health Division, Tshuapa, Democratic Republic of the Congo (J.P. Matela, T. Likafi); Institute National de Recherche Biomedical, Kinshasa (E. Kinganda-Lusamaki, E.H. Vakaniaki, A. Aziza, F. Muyembe, S. Ahuka-Mundeke, E. Pukuta, J.J. Muyembe-Tamfum, P. Mbala-Kingebeni); Cliniques Universitaires de Kinshasa, Kinshasa (E. Kinganda-Lusamaki, F. Muyembe, S. Ahuka-Mundeke, J.J. Muyembe-Tamfum, P. Mbala-Kingebeni); University of Montpellier, IRD, INSERM, Montpellier, France (E. Kinganda-Lusamaki, A. Ayouba, N. Fernandez-Nuñez, M. Peeters, E. Delaporte); University

of California, Los Angeles, California, USA (N.A. Hoff, S. Merritt, A.W. Rimoin); Centers for Disease Control and Prevention, Atlanta, Georgia, USA (J. Kabamba, J.B. Doty, A. McCollum); University of Antwerp, Antwerp, Belgium (T. Cooreman, J. Mariën); Kinshasa School of Public Health, Kinshasa (B. Nguete, D. Kaba); University of Florida, Gainesville, Florida, USA (D. Witte); Institute of Tropical Medicine, Antwerp (S. Roge, E. Bangwen, L. Liesenborghs); US Department of Agriculture, Manhattan, Kansas, USA (S. Lakin, C. Lewis, L.E. Hensley)

DOI: <https://doi.org/10.3201/eid3004.231413>

available diagnostic laboratories in remote regions (4). Reported increases in mpox cases without well-defined origins suggest that a One Health investigation might be a more informative strategy. As part of this approach, investigations by DRC's National Program for Mpox and Viral Hemorrhagic Fevers (PNLMPX-VHF) and its partners, including the US Centers for Disease Control and Prevention (CDC) and Belgium Institute of Tropical Medicine, have begun sampling mammals in areas with mpox cases and animals manifesting pox-like lesions (7). Previous sampling focused on wildlife and small animals but has recently been expanded to include

domestic animals because animal husbandry is widespread throughout DRC (8). Yet, areas with livestock have not been regularly surveilled for health, hygiene, or suspected viruses, including MPXV. Close proximity of humans and animals provides an environment conducive to virus spillover (9). We report on the co-circulation of MPXV and swinepox virus in the same locality within DRC.

The Study

In early September 2022, provincial authorities in Tshuapa, a heavily forested province in northwest DRC, alerted PNLMPX-VHF regarding the deaths of

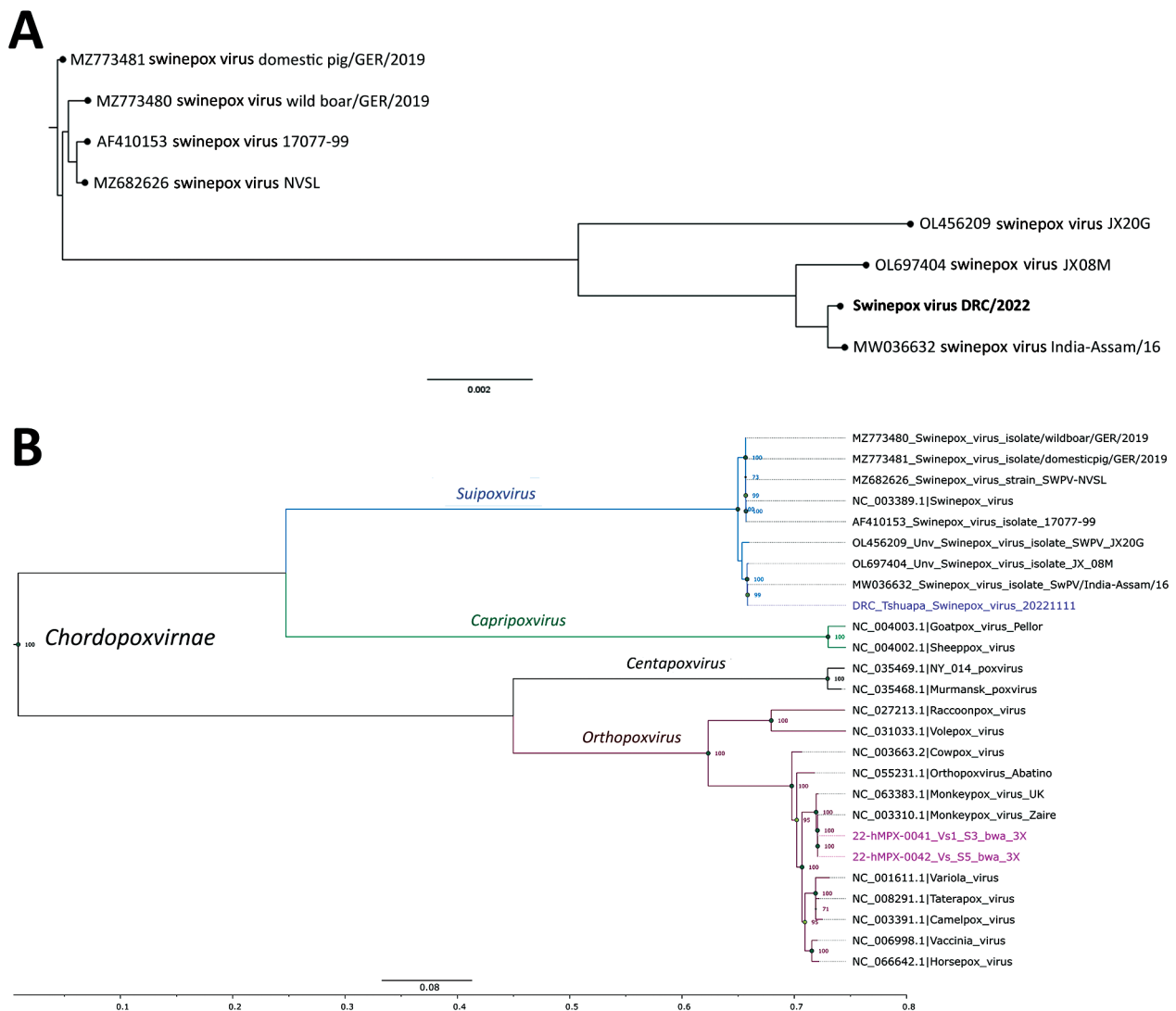


Figure 1. Phylogenetic analysis of swinepox virus (A) and monkeypox virus (B) in study of co-circulating viruses, Democratic Republic of the Congo, 2022. Poxvirus sequences for comparison were obtained from GenBank. Trees were constructed by using the maximum-likelihood method and the general time reversal substitution model with gamma distribution and proportion of invariable sites. In panel A, bold text indicates the swinepox sequence from this study; in panel B, magenta-colored text indicates monkeypox virus sequences and blue text indicates swinepox virus sequence from this study. The monkeypox virus samples belong to clade I and correspond to those previously described (11). Scale bars indicate nucleotide substitutions per site. DRC, Democratic Republic of the Congo.

pigs in the Boende health zone that manifested vesicular, pox-like lesions. Authorities also reported several suspected mpox cases among farm owners and residents in the area; clinical samples were not collected from those persons. Tshuapa is considered an MPXV-endemic area and has a robust surveillance program through PNLMPX-VHF, Kinshasa School of Public Health, and CDC. Through this program, suspected mpox case samples are sent to DRC's reference laboratory, the Institute National de Recherche Biomedical (INRB), for testing and, when requested, sequencing (6).

After the alert, during September 9–19, two pigs with pox-like lesions were identified at a domestic farm where pigs roamed freely. The first diseased pig had died on September 1; no samples had been collected. On September 10, the second pig, which had predominant umbilicated nodular dermatitis between the ears and on the face and chest, died. We collected a vesicle swab sample from the cadaver for analysis at INRB. MPXV PCR (10) results were negative, and we sequenced the sample by using an Illumina platform (Illumina, <https://www.illumina.com>) probe enrichment-based protocol and a custom-made virus research panel (Twist Biosciences, <https://www.twistbioscience.com>). We used the open source bioinformatics pipeline, GeVarLi (<https://forge.ird.fr/transvihmi/nfernandez/GeVarLi>), to clean and align sequences and assign virus lineages. We completed de novo sequence assembly by using SPAdes Genome Assembler (<https://github.com/ablab/spades>). Sequencing results and phylogenetic analysis indicated that the second pig had been infected with swinepox virus (Figure 1, panel A) (11).

After identifying a swinepox-positive pig, we launched an expanded poxvirus investigation on

October 2. In the Boende health zone, we identified 16 suspected human mpox cases, all occurring within 400 m of the farm that had a confirmed swinepox case. None of the persons with suspected mpox reported having mpox vaccinations, nor did they have scars indicating past mpox or variola vaccination. We collected skin swab, oropharyngeal lesion swab, and venous blood samples from case-patients with suspected active ($n = 2$) and convalescent ($n = 4$) MPXV infections and analyzed those samples at INRB. The 2 suspected active case-patients (both children) were MPXV positive. Sequencing confirmed infection with MPXV; however, we did not detect swinepox co-infection (Figure 1, panel B). We deposited the 2 MPXV sequences in the GISAID database (<https://www.gisaid.org>; accession nos. EPI_ISL_18857033–4). We determined that the farm owner, not realizing his pigs were sick, permitted his 2 children to directly contact the pigs during feeding without protection. Both children exhibited mild symptoms of fever, colds, headaches, and physical asthenia and developed a rash and papules 12 days after disease onset in the first pig. Both MPXV PCR-positive children recovered without complications; in addition, the family reported that they had previously recovered from PCR-confirmed MPXV. No epidemiologic link was established between the mpox-positive children and the swinepox-positive pig.

We returned to the area to identify additional pigs with symptoms of pox-like virus infections and found 4 suspected cases, of which 3 were in a neighboring farm. All pigs with suspected infections had pox-like skin lesions on the face, mouth, or tongue and had liver pathology similar to clinical manifestations of pox-like virus infections. A third pig from the original farm died on October 24; we collected lesion



Figure 2. Animals with mpox-like lesions in study of co-circulating monkeypox and swinepox viruses, Democratic Republic of the Congo, 2022. A, B) Mpox-type lesions observed on internal tissues (A) and on the skin (B) of a pig. C) Lesion on the skin of a rodent that was found in the same geographic area as the pig.

crust, vesicle, and rectal swab samples and internal organs where vesicular lesions were noted (Figure 2, panels A, B). Small mammals found in the vicinity of the original farm and that farmer's house also showed similar skin lesions (Figure 2, panel C). We collected lung, spleen, kidney, liver, oral, and rectal swab samples from 28 small mammals (shrews and black rats [*Rattus rattus*]) for analysis at INRB; 2 rats exhibited lesions. All samples tested negative for MPXV by PCR, and no additional sequencing was performed.

Concurrently, medical staff reported periodic human outbreaks of suspected MPXV infections and wild and domesticated animal deaths in other health zones. In addition, outbreaks of pox-like disease in pigs preceded confirmed human mpox cases in the Bolomba health zone of Equateur Province in 2008 and the Masi-Manimba health zone of Kwilu Province in 2022 (12). Although no samples were collected from sick animals during those outbreaks, the overall frequency of reports warrants future use of One Health approaches in all investigations of pox-like diseases, regardless of the initial species in which the lesions are observed.

Conclusions

In environments endemic for zoonotic pathogens such as MPXV, the role of animal reservoirs and hosts must be considered in surveillance and outbreak control. Reports of multiple poxviruses (vaccinia virus, pseudocowpox virus, MPXV) co-circulating within a community are rare and have been primarily confined to Brazil (2,13). In addition to mpox, and now swinepox, DRC has reported cases of other poxviruses, including molluscum contagiosum and tanapox (14). The appearance of multiple poxviruses in the same region indicates sensitive, specific diagnostics and robust sequencing capacity are needed to characterize those agents, better understand poxvirus epidemiology, and prevent potential poxvirus transmission.

Acknowledgments

We thank the Tshuapa provincial health division staff, especially those in Boende and Mompono health zones; all health care workers involved in mpox surveillance and animal health and safety in DRC; the staff from the INRB Biorepository, including Gabriel Kabamba, Raphael Lumembe, Andre Citenga, Fiston Cikaya, Sifa Kavira, Zadoc Yungwe, and Michel Kenye; staff from the INRB Department of Epidemiology and Virology, including Guy Midingi, Sophie Gabia, Akil Prince, Chloé Muswamba, and Ola Rilia, for their valuable involvement and support; and Yu Li for review of the genetic sequences.

This work was funded by the Research Foundation-Flanders (grant no. G096222N to L.L.); US Department of Defense, Defense Threat Reduction Agency, Monkeypox Threat Reduction Network (MPX-TRN) (to A.W.R.); US CDC (to D.K.); project PANAFPOX funded and coordinated by The French National Agency for Research on AIDS and Viral Hepatitis-Maladies Infectieuses Emergentes (ANRS-MIE) and Agence Française de Développement through the AFROSCREEN project (grant agreement no. CZZ3209) in partnership with Institut Pasteur and Institute de Recherche pour le Développement (to M.P. and E.D.); and a PhD grant from the French Foreign Affairs Office (to E.K.-L.).

About the Author

Dr. Kalonji is a disease surveillance officer with the National Viral Hemorrhagic Fevers and Mpox Control Program of the DRC. His research interests include mpox, COVID-19, Ebola virus disease, cholera, and vaccine-preventable diseases.

References

1. Bunge EM, Hoet B, Chen L, Lienert F, Weidenthaler H, Baer LR, et al. The changing epidemiology of human monkeypox – a potential threat? A systematic review. *PLoS Negl Trop Dis*. 2022;16:e0010141. <https://doi.org/10.1371/journal.pntd.0010141>
2. Oliveira GP, Rodrigues RAL, Lima MT, Drumond BP, Abrahão JS. Poxvirus host range genes and virus-host spectrum: a critical review. *Viruses*. 2017;9:331. <https://doi.org/10.3390/v9110331>
3. Afonso CL, Tulman ER, Lu Z, Zsak L, Osorio FA, Balinsky C, et al. The genome of swinepox virus. *J Virol*. 2002;76:783–90. <https://doi.org/10.1128/jvi.76.2.783-790.2002>
4. Rimoin AW, Mulembakani PM, Johnston SC, Lloyd Smith JO, Kisalu NK, Kinkela TL, et al. Major increase in human monkeypox incidence 30 years after smallpox vaccination campaigns cease in the Democratic Republic of Congo. *Proc Natl Acad Sci USA*. 2010;107:16262–7. <https://doi.org/10.1073/pnas.1005769107>
5. World Health Organization. WHO Director-General's statement at the press conference following IHR Emergency Committee regarding the multicountry outbreak of monkeypox – 23 July 2022 [cited 2023 Jun 20]. <https://www.who.int/director-general/speeches/detail/who-director-general-s-statement-on-the-press-conference-following-IHR-emergency-committee-regarding-the-multicountry-outbreak-of-monkeypox--23-july-2022>
6. Whitehouse ER, Bonwitt J, Hughes CM, Lushima RS, Likafi T, Nguete B, et al. Clinical and epidemiological findings from enhanced monkeypox surveillance in Tshuapa Province, Democratic Republic of the Congo during 2011–2015. *J Infect Dis*. 2021;223:1870–8. <https://doi.org/10.1093/infdis/jiab133>
7. Centers for Disease Control and Prevention. Mpox in animals and pets. January 4, 2023 [cited 2023 March 13]. <https://www.cdc.gov/poxvirus/mpox/veterinarian/mpox-in-animals.html>
8. Bosworth A, Wakerley D, Houlihan CF, Atabani SF. Monkeypox: an old foe, with new challenges. *Infect Prev*

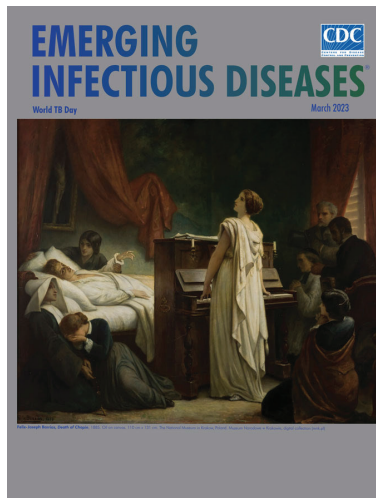
- Pract. 2022;4:100229. <https://doi.org/10.1016/j.infpip.2022.100229>
9. Vora NM, Hannah L, Walzer C, Vale MM, Lieberman S, Emerson A, et al. Interventions to reduce risk for pathogen spillover and early disease spread to prevent outbreaks, epidemics, and pandemics. *Emerg Infect Dis.* 2023;29:1–9. <https://doi.org/10.3201/eid2903.221079>
 10. Li Y, Zhao H, Wilkins K, Hughes C, Damon IK. Real-time PCR assays for the specific detection of monkeypox virus West African and Congo Basin strain DNA. *J Virol Methods.* 2010;169:223–7. <https://doi.org/10.1016/j.jviromet.2010.07.012>
 11. Berthet N, Descorps-Declère S, Besombes C, Curaudeau M, Nkili Meyong AA, Selekon B, et al. Genomic history of human monkey pox infections in the Central African Republic between 2001 and 2018. *Sci Rep.* 2021;11:13085. <https://doi.org/10.1038/s41598-021-92315-8>
 12. World Health Organization. RD Congo: rapport de mission d'investigation et riposte à l'épidémie de monkey pox à Djoa, zone de santé de Bolomba. 2008 [cited 2023 Jun 23]. <https://reliefweb.int/report/democratic-republic-congo/rd-congo-rapport-de-mission-dinvestigation-et-riposte-%C3%A0-l%C3%A9pid%C3%A9mie>
 13. Luques MN, Oliveira RL, Hir S, Nunes DDS, Higa LM, Mendonça AF, et al. Co-circulation of vaccinia and monkeypox viruses in rural areas of Brazil: importance of differential molecular diagnosis. *Travel Med Infect Dis.* 2023;53:102578. <https://doi.org/10.1016/j.tmaid.2023.102578>
 14. Monroe BP, Nakazawa YJ, Reynolds MG, Carroll DS. Estimating the geographic distribution of human Tanapox and potential reservoirs using ecological niche modeling. *Int J Health Geogr.* 2014;13:34. <https://doi.org/10.1186/1476-072X-13-34>

Address for correspondence: Nicole Hoff, Department of Epidemiology, UCLA Fielding School of Public Health, 650 Charles E. Young Dr, CHS 41-275, Los Angeles, CA 90095, USA; email: nhoff84@ucla.edu

March 2023

World TB Day

- Risk for Prison-to-Community Tuberculosis Transmission, Thailand, 2017–2020
- Multicenter Retrospective Study of Vascular Infections and Endocarditis Caused by *Campylobacter* spp., France
- Yellow Fever Vaccine–Associated Viscerotropic Disease among Siblings, São Paulo State, Brazil
- *Bartonella* spp. Infections Identified by Molecular Methods, United States
- COVID-19 Test Allocation Strategy to Mitigate SARS-CoV-2 Infections across School Districts
- Using Discarded Facial Tissues to Monitor and Diagnose Viral Respiratory Infections
- Postacute Sequelae of SARS-CoV-2 in University Setting
- Associations of *Anaplasma phagocytophilum* Bacteria Variants in *Ixodes scapularis* Ticks and Humans, New York, USA
- Prevalence of *Mycobacterium tuberculosis* Complex among Wild Rhesus Macaques and 2 Subspecies of Long-Tailed Macaques, Thailand, 2018–2022
- Increase in Colorado Tick Fever Virus Disease Cases and Effect of COVID-19 Pandemic on Behaviors and Testing Practices, Montana, 2020
- Clonal Dissemination of Antifungal-Resistant *Candida haemulonii*, China



- Comparative Effectiveness of COVID-19 Vaccines in Preventing Infections and Disease Progression from SARS-CoV-2 Omicron BA.5 and BA.2, Portugal
- Clonal Expansion of Multidrug-Resistant *Streptococcus dysgalactiae* Subspecies *equisimilis* Causing Bacteremia, Japan, 2005–2021
- Extended Viral Shedding of MERS-CoV Clade B Virus in Llamas Compared with African Clade C Strain
- SARS-CoV-2 Incubation Period during the Omicron BA.5–Dominant Period in Japan

- Seroprevalence of Specific SARS-CoV-2 Antibodies during Omicron BA.5 Wave, Portugal, April–June 2022
- Risk Factors for Reinfection with SARS-CoV-2 Omicron Variant among Previously Infected Frontline Workers
- Correlation of High Seawater Temperature with *Vibrio* and *Shewanella* Infections, Denmark, 2010–2018
- Tuberculosis Preventive Therapy among Persons Living with HIV, Uganda, 2016–2022
- Nosocomial Severe Fever with Thrombocytopenia Syndrome in Companion Animals, Japan, 2022
- *Burkholderia thailandensis* Isolated from the Environment, United States
- *Mycobacterium leprae* in Armadillo Tissues from Museum Collections, United States
- Reemergence of Lymphocytic Choriomeningitis Mammarenavirus, Germany
- *Emergomyces pasteurianus* in Man Returning to the United States from Liberia and Review of the Literature
- New Detection of Locally Acquired Japanese Encephalitis Virus Using Clinical Metagenomics, New South Wales, Australia

**EMERGING
INFECTIOUS DISEASES**

To revisit the March 2023 issue, go to:
<https://wwwnc.cdc.gov/eid/articles/issue/29/3/table-of-contents>