Published in final edited form as:

N Engl J Med. 2023 December 21; 389(25): 2355–2362. doi:10.1056/NEJMoa2306448.

Locally Acquired Melioidosis Linked to Environment — Mississippi, 2020–2023

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Summary

Melioidosis, caused by *Burkholderia pseudomallei*, is a rare but potentially fatal bacterial disease endemic to tropical and subtropical regions worldwide. It is typically acquired through contact with contaminated soil or fresh water. Before this investigation, *B. pseudomallei* was not known to have been isolated from the environment in the continental United States. Here, we report on three patients living in the same Mississippi Gulf Coast county who presented with melioidosis within a 3-year period. They were infected by the same Western Hemisphere *B. pseudomallei* strain that was discovered in three environmental samples collected from the property of one of the patients. These findings indicate local acquisition of melioidosis from the environment in the Mississippi Gulf Coast region.

Melioidosis is a potentially fatal disease caused by the environmental bacterium *Burkholderia pseudomallei*, which is mainly distributed in tropical regions worldwide between the latitudes of 20 degrees north and 20 degrees south.[1] Infection occurs by means of exposure to contaminated soil or water by percutaneous inoculation, inhalation, or ingestion. Symptoms typically appear within 21 days after exposure. The clinical manifestations of melioidosis are diverse and often mimic those of other diseases.[2]

Melioidosis is rare in the United States; the Centers for Disease Control and Prevention (CDC) has received reports of approximately 12 cases each year that were determined to have been predominately associated with travel to regions in which melioidosis was endemic.[8] Excluding the cases described in this report, 12 cases of melioidosis that were not associated with travel were identified in the contiguous United States from 1950 through 2022[1,3,10–17]; half these cases occurred in the past 5 years.[3,16,17] Of the 10 cases that were not associated with travel and in which whole-genome sequencing was available, only 2 cases were associated with infecting strains that originated in the Western Hemisphere. These 2 cases involved patients who resided in the same south Texas county; the sources of their infections were never determined.[3–5]

Despite several investigations that included environmental sampling,[3–6] *B. pseudomallei* has not been isolated from the environment in the continental United States. However, environmental suitability modeling suggests that parts of the southern United States are suitable environments for *B. pseudomallei*.[7,9]

Here we report on non–travel-associated melioidosis in three patients from the same Mississippi Gulf Coast county. Clinical *B. pseudomallei* isolates obtained from these three patients were clonal to each other, a finding that suggests a common source of infection.

Case Reports

Patient 1, a 39-year-old man, presented to a Mississippi hospital in July 2020 (Fig. 1) with fever (temperature, 40.8°C), in acute respiratory distress, and with multisystem organ failure after 1 week of fatigue, weakness, loss of appetite, worsening dyspnea at rest, and

productive cough with hemoptysis. He was admitted to the intensive care unit with bilateral multilobar pneumonia (Fig. 2A) and sepsis. Positive blood cultures growing gram-negative rods were detected on admission and persisted until hospital day 6. On hospital day 7, *B. pseudomallei* was presumptively identified from the initial blood culture and later confirmed by the Mississippi State Department of Health (MSDH) and the CDC. Patient 1 also tested positive for influenza A virus on admission. The patient's medical history was notable for type 2 diabetes mellitus, alcohol use disorder (1000 g of alcohol per day), fatty liver disease, and obesity; he smoked tobacco cigarettes (20 pack-years). He reported no travel outside the continental United States in his lifetime.

Patient 2, a 62-year-old man, presented to the hospital in April 2022 (Fig. 1) with left upper lobe pneumonia with associated mediastinal lymphadenopathy (Fig. 2B) and sepsis after 10 days of progressive weakness, productive cough, and subsequent fever, chills, night sweats, and diarrhea. On hospital day 4, *B. pseudomallei* was presumptively identified from a blood culture obtained on admission. Treatment with oral levofloxacin at a dose of 750 mg per day was started, and the patient was discharged home. He was readmitted 1 week later, still symptomatic, after *B. pseudomallei* was confirmed by the MSDH, and acute-phase therapy for melioidosis was started. The patient's medical history included excessive alcohol use, hypertensive heart disease, and prediabetes (glycated hemoglobin level, 6.1); he smoked tobacco cigarettes (>10 pack-years). He reported no travel to a melioidosis-endemic country in his lifetime.

Patient 3, a 64-year-old man, presented to the emergency department in January 2023 (Fig. 1) with progressively worsening midback pain that had begun approximately 3 weeks earlier after he had had an influenza-like illness. A spinal epidural abscess (Fig. 2C) was diagnosed on magnetic resonance imaging, resulting in an emergency thoracic laminectomy with washout. *B. pseudomallei* infection was confirmed from blood and spinal-abscess cultures. Right-lobe pneumonia was also diagnosed on admission, but cultures of pleural fluid remained negative. His medical history included hypertension and arthritis. He reported no travel outside the continental United States in his lifetime.

Patients 1 and 2 ultimately recovered after completing antimicrobial therapy (a description of the complete clinical course is provided in the Supplementary Appendix, available with the full text of this article at NEJM.org). Patient 3 was still recovering at the time of this report, and his case continues to be investigated.

Methods

CDC and MSDH Investigations

Two separate investigations were conducted by the CDC and the MSDH after case notifications regarding Patients 1 and 2 (Fig. 1) and are described below. A third investigation involving Patient 3 is currently under way and is not described here.

2020 Investigation—Extensive interviews with Patient 1 and his close contacts were conducted in September 2020 to determine potential exposures. In September 2020, investigators collected 59 environmental samples from a dry-docked tugboat in Louisiana

where Patient 1 worked and where the nonpotable water system on board was believed to have been his most likely source of exposure (Table S1 in the Supplementary Appendix).

2022 Investigation—We interviewed Patient 2 in May and June 2022 and reinterviewed Patient 1 on June 27, 2022, using an open-ended exposure questionnaire. We focused the questions on the 30 days preceding the onset of symptoms and covered a wide range of potential exposures. We assessed the patients' domestic travel history over the preceding year and all international travel in the patients' lifetimes. We reviewed and compared medical records for Patients 1 and 2 to identify clinical risk factors and potential exposure timelines, events, and locations that informed our environmental sampling strategy.

On June 26, 27, and 28, 2022, we obtained 109 samples, including 18 household-product samples from the household of Patient 2 and 79 environmental samples from the patients' properties and the areas they frequented in their Mississippi county. Water was collected from a nearby river, a lake, a pond, and other water sources associated with the patients' properties. Other environmental samples included plant material, dead fish, and surface swabs of water drains and pipes. All the soil samples were collected at a depth of 30 cm when possible. Domestic products that were sampled from the household of Patient 2 were focused on liquid or moist products that he reported using in the 30 days before the onset of symptoms.

The CDC processed the samples for *B. pseudomallei* testing according to consensus guidelines (see the Supplementary Appendix).[18] Isolates from clinical cases and environmental investigations that were suggestive of *B. pseudomallei* were confirmed with the use of the Laboratory Response Network algorithm, including biochemical and polymerase-chain-reaction (PCR) assays. DNA from isolates was extracted for whole-genome sequencing with the use of the Promega Maxwell RSC with a Cultured Cells extraction kit (Promega).

Libraries of DNA were generated with the use of the Illumina Nextera FLEX kit or the Illumina DNA Prep (M) Tagmentation Illumina Purification Bead kit and were processed on an iSeq 100 instrument with a 2×151 bp cartridge (Illumina). Draft genomes of the recovered isolates were analyzed with traditional multilocus sequence typing and compared with all publicly available *B. pseudomallei* genomes included in the National Center for Biotechnology Information (NCBI) RefSeq database (as of December 23, 2022) with the use of Parsnp, version 1.5.2, for detection of single-nucleotide polymorphisms (SNPs) and FastTree, version 2.1.11, for maximum-likelihood phylogenetic inferences.[19–22] Dendrograms were generated with the use of iTol, version 6.6, and FigTree, version 1.4.4, and were refined with InkScape software, version 1.0.1.[23,24] Genome sequences from isolates were submitted to the NCBI under BioProject accession number PRJNA942243. ClonalFrameML, version 1.12, was used for recombination detection.[25]

Results

Patients

Patients 1 and 2 lived 7 miles apart and were longtime residents of the same Mississippi Gulf Coast county. They did not know each other or have any connection to each other's properties. Patient 1 had lived in Mississippi his entire life and had never traveled internationally. Patient 2 moved to Mississippi as a child and was a retired veteran with no travel history to melioidosis-endemic countries. Both patients had occupational histories as maritime professionals working in intracoastal areas of the Lower Mississippi River Delta region in Louisiana and Mississippi. Patient 1 was employed in the industry at the time of the onset of his symptoms, but Patient 2 had been retired for several years. Both patients fished recreationally on the same river but at different locations. Patient 2 reported that he would routinely go fishing during rainstorms. Patient 1 reported that he did not do yard work or have any direct contact with soil or water on his property; the property, in a subdivision built on previously uninhabited swamp land, was constructed after Hurricane Katrina and approximately 10 years before this investigation. Patient 1 lived on the property for approximately 10 months before the onset of his symptoms. Patient 2 reported spending most of his time at home during the 3 weeks preceding the onset of his symptoms, and he did not perform gardening or yard work except for lawn mowing during that period of time. He had lived on his property, which abutted the river where both patients recreationally fished, for approximately 10 years.

Investigations

One surface-water sample collected from a puddle in the front yard and two soil samples collected from the front and back yard on the property belonging to Patient 1 in June 2022 tested positive for *B. pseudomallei* by PCR assay and InBios Active Melioidosis Detect Rapid Test Kit (InBios) on enrichment. Isolates obtained from enrichment broths were confirmed to be *B. pseudomallei*. All other samples collected during the two investigations in 2020 and 2022, including those obtained from the property belonging to Patient 2, were negative for *B. pseudomallei*.

The genome sequences from the clinical isolates obtained from the three patients and the isolates recovered from the three environmental samples were all sequence type 92 (determined with the use of traditional multilocus sequence typing), which is a sequence type associated with strains of Western Hemisphere origin. A higher-resolution analysis of SNPs showed that all isolates were clonal to each other (3 to 15 SNPs apart) but did not match any previously sequenced examples, a finding that indicates a new strain. A comparison of this strain with publicly available *B. pseudomallei* genomes indicates that it is within the major clade of Western Hemisphere strains and that it groups with strains associated with South America (Fig. 3).

Discussion

Our investigation identified an environmental isolation of *B. pseudomallei* in the continental United States. The clonality of the isolates from the three patients and from the

environmental samples, along with the patients' close geographic proximity and lack of travel history, suggest local acquisition of melioidosis from the environment in Mississippi. All three patients lived within a 20-mile radius in the same Mississippi Gulf Coast county but did not know each other. The genomic sequences of the isolates comprising this new strain, which we are calling GCS2020 (Gulf Coast Strain 2020), are greater than 1000 SNPs distant from any other genome available, which means that it is not a match to any previously encountered isolate and thus is distinct from other strains such as ATS2021, which was implicated in a multistate outbreak caused by an aromatherapy spray,[17] and strain 1026b, which was the source of a melioidosis outbreak among nonhuman primates at the Tulane National Primate Research Center in Louisiana.[26] Our analysis groups this new strain with others from South America, but it lacks the resolution to conclude timing, source, or geographic origin for the introduction of this strain to the Mississippi Gulf Coast region. This lack of resolution is due mainly to the limited number of genome sequences that are available from strains in the Americas.

The location of the environmental detection of *B. pseudomallei*, at 30 degrees north latitude, is outside the typical range for this bacterium but within a predicted geographic area, as determined with environmental suitability modeling.[1,9] Although *B. pseudomallei* was not detected in the other sampled locations, including the property belonging to Patient 2, this factor does not rule out the presence of the bacterium in those areas. A similar outcome was seen in the 2018 Texas investigation, in which the local environment remains the likely source of exposure in the two patients involved.[3]

Although the exact mode of transmission and exposure event could not be identified for the patients in this study, inhalation exposure[2,27,28] aligns with the mediastinal lymphadenopathy that was observed on computed tomography in Patient 2 and the patient's report of fishing during rainstorms.[29] Patients 1 and 2 had clinical risk factors (diabetes and excessive alcohol use) that are associated with melioidosis, but Patient 3 did not.[2,30] Patient 1 had a coinfection with influenza A virus, a finding that had been documented in other case reports of melioidosis.[31,32]

Because melioidosis was not initially suspected in these patients, appropriate antimicrobial therapy[33,34] was delayed. Additional locally acquired cases may have been missed owing to lack of awareness, misidentification, or misdiagnosis, especially during the coronavirus disease 2019 pandemic. Clinicians should consider melioidosis in patients with a compatible illness who reside in or have traveled to the Gulf Coast region of the southern United States or to areas where *B. pseudomallei* has historically been endemic. Because culture is the reference standard for diagnosis, clinicians should repeat cultures from all clinically relevant sites, even if initial cultures are negative, in patients in whom there is a high clinical suspicion for melioidosis. Misidentification of the pathogen on automated platforms is possible[35] and can delay diagnosis and treatment and potentially worsen outcomes.[36,37]

Clinical laboratory scientists should consult with a reference laboratory regarding infection suggestive of *B. pseudomallei*. Because melioidosis is now a nationally notifiable condition, the Council of State and Territorial Epidemiologists recommends that it be added to the reportable disease list in every U.S. jurisdiction to aid in timely investigation and increase

our epidemiologic knowledge of this disease in the United States.[38] Environmental sampling and seroprevalence studies are needed to determine how widespread *B. pseudomallei* is in the environment in Mississippi and other Gulf Coast states where environmental conditions are predicted to be suitable for this bacterium.[7,9]

The investigation of non–travel-related melioidosis cases in Mississippi led to the isolation of *B. pseudomallei* in soil and water and ultimately linked to infection in three patients. These findings indicate that melioidosis may be endemic to the Mississippi Gulf Coast region.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

We thank Robyn A. Stoddard, D.V.M., Ph.D., Lindy Liu, M.P.H., Brian Richardson, M.P.H., Katherine DeBord, M.P.H., and Caroline Schrodt, M.D., M.S.P.H., of the CDC, for laboratory and epidemiology assistance; Marie De Perio, M.D., of the CDC National Institute for Occupational Safety and Health, Mia Mattioli, Ph.D., E.I.T., of the CDC Division of Foodborne, Waterborne, and Environmental Diseases, and Bart Currie, F.R.A.C.P., F.A.F.P.H.M., of the Menzies School of Health Research, for consultation; and the patients and their families for their cooperation and for welcoming the investigation team onto their properties.

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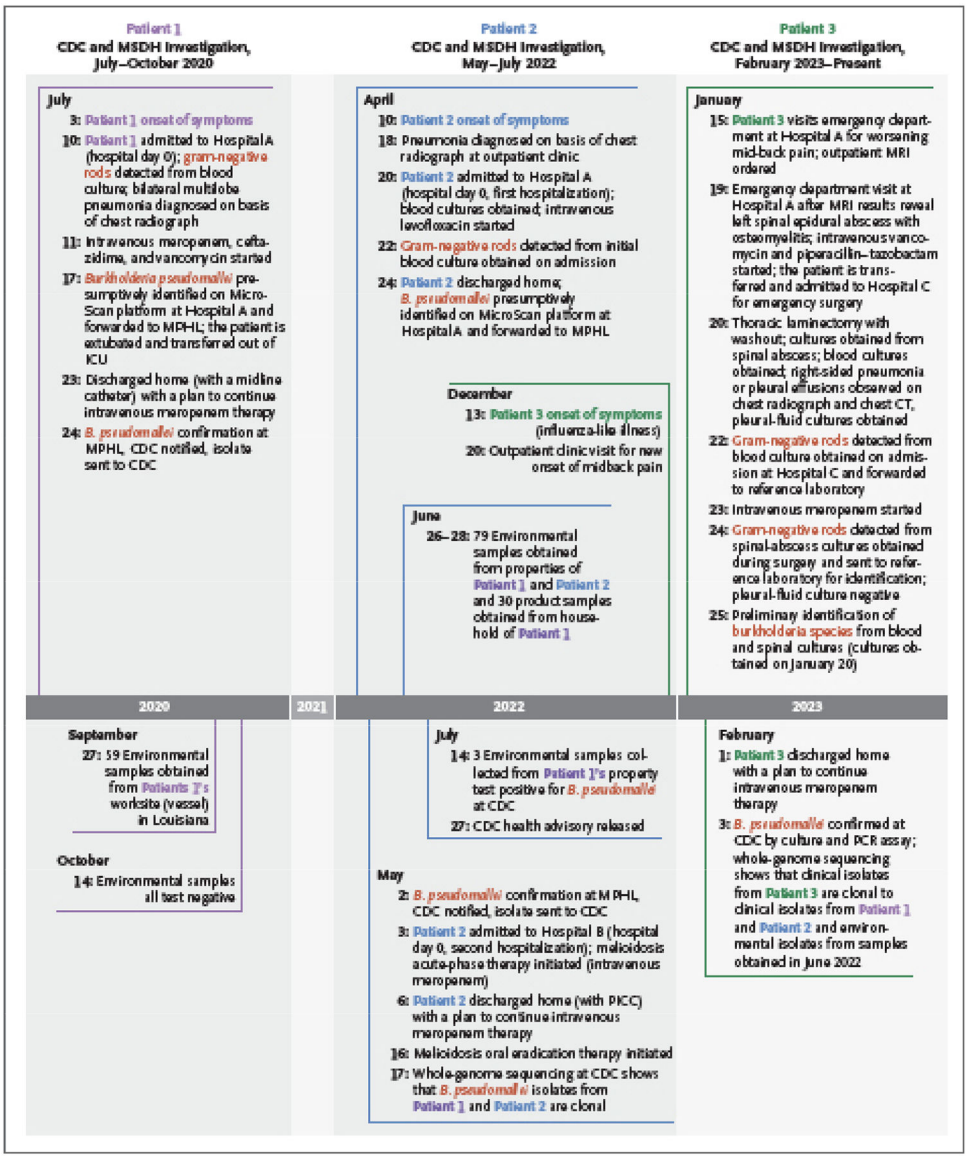

Figure 1. Clinical Course and Investigation Timeline for Patients 1, 2, and 3.

Shown is the timeline of events starting with symptom onset in Patient 1 in July 2020 and progressing to the ongoing Centers for Disease Control and Prevention (CDC) and Mississippi State Department of Health (MSDH) investigation with regard to melioidosis in Patient 3. CT denotes computed tomography, ICU intensive care unit, MPHL Mississippi Public Health Laboratory, MRI magnetic resonance imaging, PCR polymerase chain reaction, and PICC peripherally inserted central catheter.

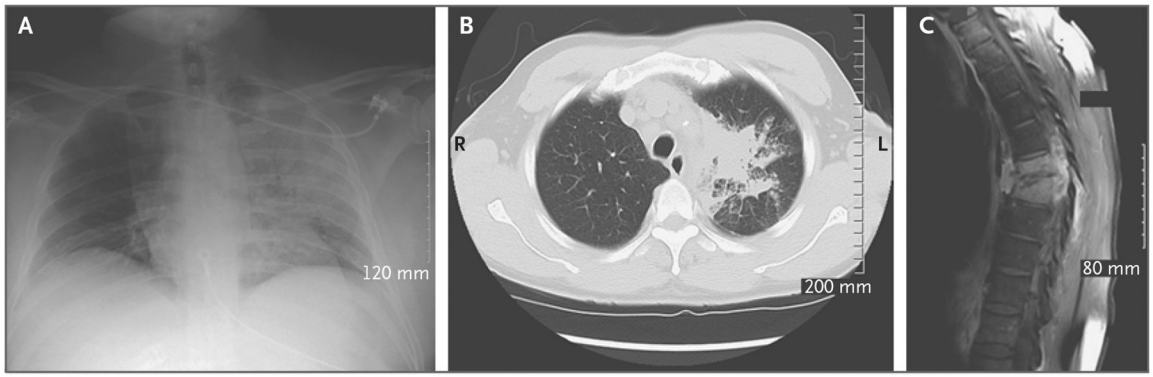

Figure 2. Thoracic Imaging in Patients 1, 2, and 3.

In Patient 1, a chest radiograph (Panel A) on hospital day 0 showed multilobar pneumonia and areas of subsegmental atelectasis. In Patient 2, CT without contrast material (Panel B) on hospital day 0 of the first hospitalization showed a 6.5-cm left apical, pleural-based, masslike consolidation with associated bulky hilar and mediastinal lymphadenopathy that aroused concern for necrotizing pneumonia or cancer. (R denotes right, and L denotes left.) In Patient 3, MRI of the thoracic spine (Panel C) showed T7–T8 discitis or osteomyelitis with prevertebral and left epidural abscess resulting in mass effect on the thecal sac and cord (the site from which an abscess swab was obtained for culture and tested positive for *B. pseudomallei*).

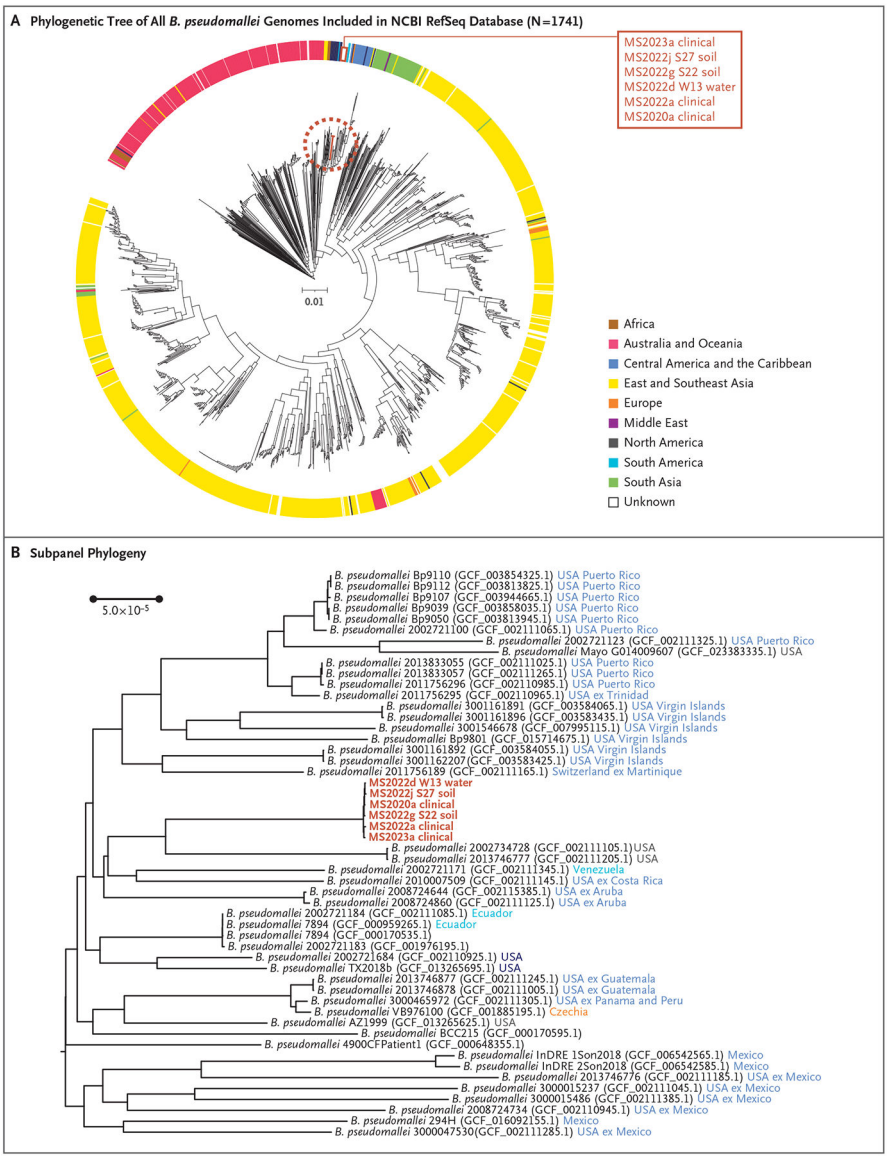

Figure 3. Whole-Genome Sequencing Analysis.

Panel A shows a phylogenetic tree of all *B. pseudomallei* genomes included in the National Center for Biotechnology Information (NCBI) RefSeq database (1741 genomes) with the addition of six new isolate genomes from this study (shown as a red node within the dashed red circle). For isolates with known geographic origin, the country was associated with the respective genome, and geographic regions were linked according to definitions listed in *The World Factbook* (https://www.cia.gov/the-world-factbook) as of December 30, 2022. Scale units denote substitutions per core single-nucleotide polymorphism (SNP) site. Panel B depicts a subpanel phylogeny of refined (recombination sites removed) mutation-only core SNP sites of genomes related to the six isolates from this study (labeled in red), which includes the clinical isolates of the three patients (shown as MS2020a clinical, MS2022a clinical, and MS2023a clinical) and the isolates recovered from the environmental samples (shown as MS2022d W13 water, MS2022g S22 soil, and MS2022j S27 soil). Tree “leaves”

list the strain or isolate identifier first, followed by the RefSeq accession in parentheses and geographic information. The notation “ex” in geographic information denotes travel history. Genomes with known geographic data are shown in the colors identical to those used in the large comprehensive panel. Scale units denote mutation-only core SNP substitutions (164,171 sites) per genome alignment site (5,706,452). ClonalFrameML posterior means for R/theta (relative rate of recombination to mutation), 1/delta (inverse mean DNA import length), and nu (mean divergence of imported DNA) were 0.949388, 0.00130988, and 0.00534356, respectively.