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CASE REPORT

Francisella philomiragia Bacteremia in a Patient with Acute Respiratory Insufficiency and Acute-on-Chronic Kidney Disease

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***Francisella philomiragia* is a very uncommon pathogen of humans. Diseases caused by it are protean and have been reported largely in near-drowning victims and those with chronic granulomatous disease. We present a case of *F. philomiragia* pneumonia with peripheral edema and bacteremia in a renal transplant patient and review the diverse reports of *F. philomiragia* infections.**

CASE REPORT

A 63-year-old female from Indiana presented to an Indianapolis hospital with worsening shortness of breath, nonproductive cough, and increasing bilateral peripheral edema. The patient was afebrile, normotensive, and normocardic but was tachypnic (40 bpm) and denied having fevers, chills, or other symptoms of an infectious process while at home. Significant medical history obtained at the time of presentation included a renal transplant secondary to polycystic kidney disease 14 years prior for which she receives chronic immunosuppressive therapy (tacrolimus and prednisone). The patient did not report recent travel outside Indiana, exposure to wild animals or recreational water sources, or exposure to sick individuals. Because of the possibility of acute transplant rejection, the patient was admitted to the intensive care unit for extensive evaluation. A chest X-ray performed at the time of admission revealed bilateral perihilar and upper-lobe infiltrates consistent with bilateral bronchopneumonia, prompting the collection of a set of blood cultures from the left arm and of another set from the right arm and initiation of empirical broad-spectrum antimicrobial therapy with vancomycin and piperacillin-tazobactam. Aside from blood cultures, no other microbiology testing was performed. Laboratory studies conducted at the time of admission revealed leukocytosis (11,600 cells/μl) with 93% neutrophils, anemia (3.32 million cells/μl), kidney failure (elevated levels of urea nitrogen [48 mg/dl] and creatinine [3.70 mg/dl]), and hyperglycemia (127 mg of glucose/dl). Elevated levels of procalcitonin (1.86 ng/ml), hematuria (25 cells/μl), and proteinuria (500 mg/dl) were also noted. Because of the possibility of acute-on-chronic kidney disease, a renal biopsy was subsequently performed, and it revealed acute allograft rejection.

Following approximately 24 h of incubation in a continuous-monitoring blood culture instrument (BD Bactec 9240; BD Diagnostic Systems, Sparks, MD), the aerobic bottles from both sets of blood cultures signaled positively. Gram stains of broth from both bottles revealed pleomorphic, Gram-negative coccobacilli (Fig. 1A). Subcultures of the blood culture broth grew medium-sized (~5-mm diameter), glossy, convex colonies resembling a member of the *Enterobacteriaceae* on sheep blood and chocolate agars after 48 h of incubation at 35°C in 5% CO_2. A Gram stain of the colonies revealed organisms with morphologies identical to those seen in the blood culture broth smear. The isolate tested positive for cytochrome oxidase and catalase. Together, these observations ruled out possible select agents, including *Francisella tularensis* and *Brucella* spp., so enhanced biosafety precautions were not implemented. Conventional tubed biochemical testing of the isolate did not result in an identification, and automated phenotypic testing (Vitek 2 GNI card; bioMérieux) yielded disparate identifications, including *Aggregatibacter aphrophilus*, *Pseudomonas aeruginosa*, and *Sphingomonas paucimobilis*. Subsequently, triplicate analysis of the isolate by both matrix-assisted laser desorption ionization–time of flight (MALDI-TOF) mass spectrometry (research-use-only [RUO] library iteration 4613, spring 2013; Bruker Daltonics, Billerica, MA) and fatty acid methyl ester analysis by gas chromatography (Sherlock MIS; MIDI, Inc., Newark, DE) yielded an identification of *Francisella philomiragia*. For the former, top score values above 2.200 were obtained for each of three analyses using standard techniques, and all top identification matches corresponded to all 6 mass spectral profiles (MSP) of *F. philomiragia* isolates incorporated into the MSP library. Sequencing of a 1,448-bp region of the 16S rRNA gene as previously described (1) confirmed the identification as *F. philomiragia* (Fig. 1B). Following isolate identification, the patient was started on a 2-week course of doxycycline therapy, which was based upon treatment recommendations for tularemia, since the clinicians were unfamiliar with the treatment options for *F. philomiragia* infections. Upon completion of antimicrobial chemotherapy, the patient was well enough to be discharged to home. Prior to discharge, however, the patient received a tunneled dialysis catheter

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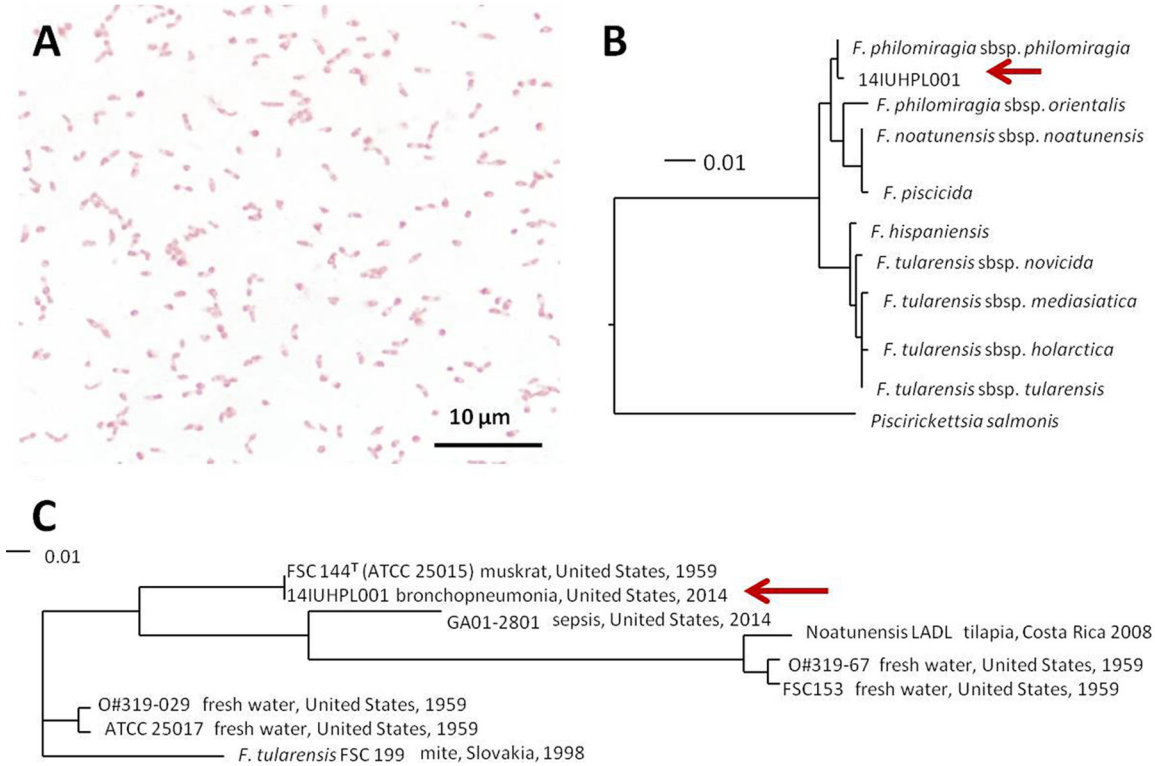

FIG 1 Imaging and phylogeny of *F. philomiragia* isolate 14IUHPL001. (A) Gram-stained smear of positive blood culture bottle demonstrating small, pleomorphic Gram-negative coccobacilli (bar, 10 μm). Neighbor-joining trees were generated based on 16S rRNA gene sequence or concatemers of MLST targets. (B) A tree generated using 16S rRNA gene input sequence from all *Francisella* species (outgroup = *Piscirickettsia salmonis*) indicates that isolate 14IUHPL001 (arrow) groups with the type strain (FSC 144^T) of *F. philomiragia* subspecies *philomiragia*, indicating a species identification of *F. philomiragia*. (C) A second tree generated using a concatemerized sequence of MLST targets from 6 strains of *F. philomiragia* subsp. *philomiragia* and a single strain of *F. noatunensis* (outgroup = *F. tularensis* subsp. *tularensis*) indicates that isolate 14IUHPL001 (arrow) branches closely with the type strain FSC144^T and is appropriately identified as *F. philomiragia* subsp. *philomiragia* at the subspecies level.

and underwent hemodialysis to compensate for decreased renal allograft function. Follow-up blood cultures were negative, and, to date, the patient has not reported infection recrudescence. The source of the patient's infection remains unknown, as none of the previously reported routes of exposure (see below) were noted. In addition, the patient denied receipt of medical treatment involving aerosolized instillation of aqueous medications or recreational water exposure.

Fine-scale phylogenetic analysis of the isolate was performed by multilocus sequence typing (MLST) using gene targets *gyrB*, *metG*, the AKIII gene, and *glpK* as follows: (i) for the AKIII gene, GAAGAAATTATAGAACAGGT and AGCATCTGAACCAATAA ACCCT; (ii) for *gyrB*, AGCTCTATCAGAAGTCAGAA and AGA TCTTCATAATCCTTAGT; (iii) for *metG*, AGCTCAAGCGTATG ATCCTG and GTTTTTGTCACAGTAATCTCT; (iv) for *metG*, TAAACCAACCTATGCTTTAG and ATTGGAATTGCATCAA GAAA ; (v) for *tyr*, ATGCTTAGTATCATACAAAG and CCTTA AAAGTAAAAGTTACAGG; (vi) for *adh*, CAGATTGTTGGTGT TGATAC and CTATCAATATCAATACGACC. An identification of *F. philomiragia* subspecies *philomiragia* was indicated by MLST (Fig. 1C). Reference sequences were obtained from GenBank (2), and phylogenetic trees were generated using Clustal Omega (3) and visualized using iTOL 2.0 (4). Conventional biochemical analysis and antimicrobial susceptibility testing (AST) by broth microdilution of this clinical isolate (14IUHPL001) and the type strain of the species, *F. philomiragia* subsp. *philomiragia* FSC144^T, were performed in parallel, and the results are summarized in Table 1.

This report describes a case of *F. philomiragia*-associated pneumonia in a patient receiving chronic immunosuppressive therapy secondary to renal allograft, as well as acute-on-chronic kidney disease and acute allograft rejection in the absence of known risk factors (e.g., exposure to an aquatic environment). Fortunately, the pathogen was rapidly identified and antimicrobial chemotherapy, which was based upon treatment recommendations for *Francisella tularensis* infections, was initiated. To our knowledge, *F. philomiragia* pulmonary infection and bacteremia in a renal transplant patient has never been reported until now.

Francisella philomiragia, a close relative of *Francisella tularensis*, is a rarely encountered opportunistic bacterial pathogen of humans. However, it is an important cause of francisellosis in wild and farmed fish. Because human diseases caused by this organism are not nationally notifiable, trends in its incidence and prevalence are unknown. Identification of this organism requires methods beyond the scope of many community hospital microbiology laboratories, and such facilities are encouraged to refer isolates to state health department laboratories or commercial reference laboratories. *F. philomiragia* is well represented in the Bruker MALDI Biotyper RUO library, so laboratories employing this technology

TABLE 1 Biochemical and antimicrobial susceptibility testing of *F. philomiragia* isolate 14IUHPL001 and the type strain, *F. philomiragia* FSC144^T

Biochemical reaction or antimicrobial agent(s)[a]	Result or MIC (μg ml^{-1})	
	FSC114^T	14IUHPL001
Biochemical reaction		
Catalase	+	+
Oxidase	+	+
Indole	−	−
Methyl red	−	−
Voges-Proskauer	−	−
Citrate utilization	−	−
Nitrate reduction	−	−
Gelatin hydrolysis	−	−
Urease	−	−
Motility	−	−
Arginine hydrolysis	−	−
Lysine decarboxylation	−	−
Ornithine decarboxylation	−	−
Malonate	−	−
DNase	−	−
β-Lactamase (nitrocefin disk)	+	+
H_2S production (TSI medium)	+	+
Acid from D-glucose	+	+
Acid from glycerol	−	−
Acid from lactose	−	−
Acid from maltose	−	−
Acid from sucrose	−	−
Antimicrobial agent(s)		
Amikacin	≤0.5	≤0.5
Amoxicillin-clavulanic acid	1	1
Ampicillin	32	32
Aztreonam	4	2
Cefepime	4	1
Ceftazidime	≤0.5	≤0.5
Ceftriaxone	≤0.5	≤0.5
Cefazolin	2	16
Ciprofloxacin	≤0.25	≤0.25
Colistin	>8	>8
Doripenem	≤0.25	≤0.25
Doxycycline	≤1	≤1
Ertapenem	≤0.25	≤0.25
Erythromycin	1	2
Gentamicin	≤0.5	≤0.5
Imipenem	≤0.25	≤0.25
Levofloxacin	≤2	≤2
Meropenem	≤0.25	≤0.25
Moxifloxacin	≤0.25	≤0.25
Oxacillin	≤0.25	>16
Polymyxin B	>4	>4
Ticarcillin-clavulanic acid	≤4	≤4
Tigecycline	≤0.25	≤0.25
Tobramycin	≤0.5	≤0.5
Trimethoprim-sulfamethoxazole	>4	>4

[a] Antimicrobial susceptibility testing was performed by broth microdilution using cation-adjusted Mueller-Hinton broth incubated in an ambient atmosphere at 35°C for 24 h. TSI, triple sugar iron.

can obtain a confident identification without further testing. Seibold et al. have reported the reliability and robustness of mass spectrometry-based identification of francisellae, including *F. philomiragia* (5); thus, as this technology becomes more commonly adopted, many laboratories will be able to confidently identify *F. philomiragia* and other rarely isolated bacteria. However, laboratories should remain diligent with regard to appropriate biosafety and biosecurity practices for isolates that cannot be ruled out as being *F. tularensis* or another select agent. Up-to-date guidance, including biosafety practices and laboratory methods, from the Laboratory Response Network for ruling out select agents should always be followed.

The taxonomy of *F. philomiragia* and of francisellae in general has been contentious, and, as a result, review of case literature is challenging. Members of the genus *Francisella* were originally classified as part of the genus *Yersinia* (6); therefore, the initial *F. philomiragia* case reports were published using the epithet *Yersinia philomiragia*. Upon publication of the genus *Francisella*, at least one distinct species was condensed into a subspecies of *F. tularensis* (7), though this recommendation has yet to be validly published and adopted by the International Committee for the Systematics of Prokaryotes. Valid publication of the novel subspecies *F. philomiragia* subspecies *noatunensis* forced the renaming of existing strains to *F. philomiragia* subspecies *philomiragia* (8). This continual revision of nomenclature indicates that cases associated with this organism have been described in the literature with one of three epithets: *Y. philomiragia*, *F. philomiragia*, or *F. philomiragia* subspecies *philomiragia*. These discrepancies prompted us to prepare a thorough review of the clinical literature to generate a complete view of the spectrum of clinical presentations associated with this organism (Table 2).

Human infections with *F. philomiragia* subspecies *philomiragia* most often present as pneumonia, but peritonitis, sepsis, and meningitis have also been documented (9–12). Most commonly, *F. philomiragia* is isolated from patients with underlying immunosuppressive conditions, especially children and young adults with chronic granulomatous disease (CGD). However, several cases of sepsis have been documented in otherwise healthy individuals who were victims of near-drowning in marine and estuary waters. In addition, isolates from both clinical (sepsis, meningitis) and subclinical infections of mammals have been reported. In contrast to *F. tularensis*, very little is understood about the pathogenesis of *F. philomiragia*. Since it is most commonly isolated from individuals who have had recent exposure to water, it is postulated that contact with, ingestion of, and/or inhalation of contaminated water may put individuals at risk for infection. However, the patient in this report has no known history of water exposure. Research has demonstrated that *F. philomiragia* is capable of forming robust biofilms in association with aquatic amoebae and, in prolonged coincubation, is capable of infecting amoebae (13). Such association may permit both the persistence of the organism in the environment and its dissemination to hosts that come in contact with *F. philomiragia*-infected amoebae via waterway exposure. *F. philomiragia* subspecies *noatunensis* appears, at this point, to be exclusively a pathogen of fish.

Clinical outcomes vary widely, ranging from complete resolution with no residual pathology to rapid death. Numerous antimicrobial treatment regimens have been reported with differing rates of resolution. A reported case that included fever and pleuritis resolved without antimicrobial therapy; however, all other reported cases featured treatment with at least two and as many as four antibiotics (9, 11). There is not a standard recommended therapy for *F. philomiragia* infection, though beta-lactams should be avoided because beta-lactamase production appears to be com-

TABLE 2 Clinical features of reported *F. philomiragia* cases

Patient sex, age (yrs)[a]	Patient location/treatment date or report date	Preexisting illness	Clinical presentation	Source of isolate	Clinical outcome	Reference
M, 18	California, USA/1974	CGD	Fever, pneumonia	Lung biopsy specimen	Resolution	9
M, 39	Colorado, USA/1976	Chronic pleural effusions	Fever	Pleural fluid	Resolution	9
M, ?	New York, USA/1977	Near-drowning	Unknown	Blood	Unknown	9
M, 39	California, USA/1978	Near-drowning	Fever, pneumonia, brain abscess	Blood	Death	9
M, 6	Switzerland/?	CGD	Fever, sepsis	Blood, bone marrow, ascitic fluid	Death	9
F, 68	Pennsylvania, USA/1980	Agnogenic myeloid metaplasia	Fever	Blood	Resolution	9
F, 86	Connecticut, USA/1980	Near-drowning	Fever, pneumonia	Blood	Resolution	9
M, 75	Connecticut, USA/1980	Near-drowning	Pneumonia	Blood	Resolution	9
M, 5	New York, USA/1981	CGD	Fever	Blood	Unknown	9
F, 12	California, USA/1981	CGD	Pneumonia	Lung biopsy specimen	Unknown	9
F, 34	New Mexico, USA/1984	None	Peritonitis	Ascitic fluid	Resolution	9
M, 28	Virginia, USA/1985	Near-drowning	Sepsis	Blood	Unknown	9
F, 47	New York, USA/1986	Hodgkin disease	Fever, sepsis	Pericardial fluid, blood	Resolution	9
M, 16	Massachusetts, USA/1986	CGD	Meningitis	Cerebrospinal fluid	Resolution	9
M, 19	Maryland, USA/1997	CGD	Fever, sepsis	Blood	Resolution	10
M, 10	Nova Scotia, Canada/2003	CGD	Adenitis, pulmonary nodules	Submandibular node	Resolution	11
M, 25	Turkey/2003	CGD	Pneumonia	Blood	Death	12
F, 63	Indiana, USA/2014	Immunosuppression secondary to renal allograft, chronic kidney disease	Pneumonia	Blood	Resolved	This report

[a] F, female; M, male.

mon (9). It is apparent that disease presentation and prognosis of *F. philomiragia* infection is case and context dependent, driven by variables that have included antimicrobial agent selection, disease status at the onset of treatment, and underlying conditions. For the first time, we report a case of francisellosis in a renal transplant patient.

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