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CASE REPORTS

Legionella jordanis Lower Respiratory Tract Infection: Case Report and Review▽

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***Legionella jordanis* was first described in 1982 after isolation from environmental sources and is otherwise a very rare human pathogen. Here, we report the recovery of *L. jordanis* from a bronchoalveolar lavage specimen from a patient who presented with an indolent lower respiratory tract infection associated with constitutional symptoms. This case is the first culture-positive case of infection involving this species in Canada.**

CASE REPORT

A 53-year-old man was assessed for a chronic cough that had become increasingly productive with nonpurulent sputum over the past year. In the past 3 months, he had also noted constitutional symptoms, including headache, myalgia, malaise, and one episode of watery diarrhea, but denied experiencing any fever, night sweats, or weight loss. He had previously worked in the quality control chemistry laboratory of the regional paper mill but had been reassigned to administrative duties ~1 year prior to the onset of the cough. To his knowledge, there were no similar symptoms in coworkers, family members, or close contacts. He denied smoking, alcohol abuse, or risk factors for human immunodeficiency virus infection. An initial evaluation of his cough consisted of a chest radiograph, which was interpreted as normal. One month later, a computed tomodensitometry test of the chest revealed a bilateral "tree-in-bud" pattern, suggesting a chronic, indolent pulmonary infectious process. A bronchoalveolar lavage was then performed, and specimens were sent for Gram staining, routine bacteriologic culture, culture for *Legionella* spp., staining and culture for *Mycobacteria* spp., and staining and culture for fungi. Direct microscopy with Gram staining revealed heavy leukocytes with mixed organisms. Within 3 days, two organisms had been isolated: *Streptococcus pneumoniae* from the blood agar plate and a gram-negative bacillus producing ~15 small, clear colonies on the buffered charcoal-yeast extract (BCYE) agar incubated aerobically at 37°C, suggestive of *Legionella* spp. This strain was sent to a regional and then a federal reference center for further characterization.

Moxifloxacin at 400 mg orally once daily for 28 days was prescribed for the patient. After finishing the treatment, he reported complete resolution of his constitutional symptoms. Although the level of sputum production significantly decreased, his cough persisted. Despite routine culture of all bronchoscopy specimens for *Legionella* spp., no other clinical isolates of *Legionella* spp. were identified in the treating medical center, L'Hôpital Regional George Dumont, Moncton, New Brunswick, Canada, in the 7 years preceding and the 8 months subsequent to this case.

Microbiological identification. The isolate obtained from the bronchoalveolar lavage specimen was forwarded to the Laboratoire de Santé Publique de Quebec, the regional reference center, where it was confirmed to be a member of the *Legionella* genus. The organism was subsequently sent to the National Microbiology Laboratory, the reference center for Canada, for species identification. Standard and extended microbiological testing procedures (14) were carried out at both institutions and are described herein. The isolate (NML 060502) grew in a candle jar atmosphere on BCYE with L-cysteine at 25, 35, and 42°C, as well as on BCYE with L-cysteine and a polymyxin-anisomycin-vancomycin supplement at 35°C. No growth occurred on BCYE with L-cysteine at 50°C, BCYE without L-cysteine at 35°C, and sheep blood agar at 35°C after 7 days. The isolate demonstrated growth with browning on tyrosine-supplemented buffered yeast extract agar, as described previously (4). The organism demonstrated a dull autofluorescence under long-wave UV light. Gram staining revealed short to long, faintly staining, gram-negative asporogenous bacilli. The isolate was catalase and β-lactamase positive, with weak and slow (4-week) gelatinase production. It was oxidase negative and failed to utilize dextrose, reduce nitrate, or hydrolyze urea and sodium hippurate.

The organism was nonreactive by indirect immunofluorescent-antibody testing (MonoFluo *Legionella pneumophila* test kit; Bio-Rad, Montreal, Quebec, Canada) and direct immunofluorescent-antibody (DFA) testing with monovalent fluorescein isothiocyanate-conjugated anti-*Legionella* (serogroup 1 to 14) rabbit sera (Pro-Lab Diagnostics, Richmond Hill, Ontario,

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Canada) for *L. pneumophila*. Commercially available, polyclonal anti-*Legionella* antibodies for DFA testing for individual, non-*pneumophila* species were used as described by the manufacturer (m-TECH, Alpharetta, GA). The results of these assays showed a strong reaction of this strain to antisera to *L. jordanis* and weak to strong cross-reactions with antisera to *L. longbeachae* serogroup 2, *L. hackeliae* serogroups 1 and 2, *L. erythra*, *L. rubrilucens*, and *L. parisiensis*. No reactivity was observed using antisera targeting *L. anisa*, *L bozemanae* serogroups 1 and 2, *L. cherrii*, *L. dumoffii*, *L. feeleii* serogroups 1 and 2, *L. gormanii*, *L. jamestowniensis*, *L. longbeachae* serogroup 1, *L. micdadei*, *L. maceachernii*, *L. oakridgensis*, *L. sainthelensi*, *L. santicrucis*, *L. spiritensis*, *L. steigerwaltii*, and *L. wadsworthii*.

Cellular fatty acid composition analysis was done after 48 h of growth on BCYE as described previously (3) except that version 4.5 of the software for the Sherlock system (MIDI, Newark, DE), MIDI's CLIN library, and the MIDI library generation system was used. The profile obtained had a similarity index value of 0.45 for the entry for *L. jordanis* in CLIN version 4.5 and was highly consistent with that of the *L. jordanis* type strain BL-540 (ATCC 33623T) obtained after in-house library generation system analysis (data not shown).

Genetics-based sequencing targeting the 16S rRNA (2) and macrophage infectivity potentiator (*mip*) genes (9) was undertaken to accurately assign this isolate to a species. The 16S rRNA gene from this isolate was 1,483 bp. The sequence was tested using BLAST software (www.ncbi.nlm.nih.gov/BLAST) and demonstrated 99.7% identity to that of the 16S rRNA gene from *L. jordanis* (type strain BL-540; GenBank accession number Z32667). Similarly, the sequence of the *mip* gene from this isolate was 639 bp and demonstrated 100% identity to the corresponding sequence from *L. jordanis* (ATCC 33623T; GenBank accession number U92209). The isolate was designated *L. jordanis* NML 060502.

Discussion. This case is the second reported culture-proven case of a lower respiratory tract infection in which *L. jordanis* was isolated and expands our knowledge of the possible clinical manifestations of infection with this uncommon pathogen. The previously reported case occurred in a 79-year-old man who also presented with a subacute course of progressive respiratory and constitutional symptoms and from whom *L. jordanis* was isolated from open-lung biopsy samples; that patient subsequently developed *Staphylococcus aureus* bacteremia and later died (13). Although cases of pneumonia and other cases of disease due to *L. jordanis* in which clinical presentations were not described have been reported previously (1, 5, 8, 12), those diagnoses of *L. jordanis* infection were made using serology only, without the isolation of the pathogen. However, diagnostic serology for *Legionella* spp. suffers from low sensitivity and specificity; cross-reactivity between serogroups, between the various species of the *Legionella* genus, and even with other genera has been consistently reported (10). Under the circumstances, the "gold standard" for diagnosing any form of infection with *Legionella* spp. remains isolation in culture (6), as was done in this case, with subsequent species identification by a polyphasic approach. Assignment to a species by DFA testing alone using specific fluorescein-labeled antisera for non-*pneumophila* taxa would have been difficult in this instance due to the high degree of cross-reactivity of this isolate with antisera from different species. Definitive characterization of the strain as *L. jordanis* had to be corroborated using sequence analyses of 16S rRNA and *mip* genes.

In the absence of a well-recognized syndrome or an experimental model to fulfill Koch's postulates, the clinical significance of an uncommon isolate, such as *L. jordanis*, is controversial. Possibilities other than infection include a nonpathogenic colonization state or exogenous contamination. However, *Legionella* spp., including *L. jordanis*, are not known to be commensals of the human respiratory tract. To date, the only natural reservoirs for *L. jordanis* to be identified are river water, tap water, and sewage (4, 7). This feature invites the possibility that acquisition by the patient may have been occupational, although this idea remains entirely speculative. Specimen contamination seems profoundly unlikely given the absence of any evidence of synchronous or metachronous isolates in the microbiology laboratory, despite the routine culture of all bronchoscopy specimens for *Legionella* spp. Furthermore, a review of infection control practices demonstrated no breach in procedures during the bronchoscopic examination, although water sampling was not performed. In support of the isolate's pathogenicity are the following observations: chronic lung disease is an established risk factor for legionellosis (11), and this patient had radiological evidence of structural airway disease; also, there was significant improvement in respiratory and constitutional symptoms with therapy possessing a spectrum of activity that includes *Legionella* spp. However, in contrast to the acute manifestations of Legionnaires' disease due to *L. pneumophila*, it appears that *L. jordanis* may tend to produce a more subacute-to-chronic respiratory infection. Although *S. pneumoniae* may have acutely contributed to his symptoms, it likely does not explain the protracted course of his respiratory illness or his chronic constitutional symptomatology.

In conclusion, we describe an indolent respiratory infection with associated constitutional symptoms due to *L. jordanis* in a patient with underlying lung disease, further demonstrating the pathogenicity of this organism.

Nucleotide sequence accession numbers. The 16S rRNA and *mip* gene sequences from this clinical isolate, *L. jordanis* NML 060502, have been deposited in GenBank under accession numbers EF036512 and EF036513, respectively.

REFERENCES

1. **Baty, V., B. Hoen, H. Schuhmacher, C. Amiel, M. Reyrolle, H. Garin, and P. Canton.** 1997. Legionella jordanis pneumonia unresponsive to fluoroquinolones in a non-immunocompromised host. Scand. J. Infect. Dis. **29:**319–320.
2. **Bernard, K., L. Shuttleworth, C. Munro, J. C. Forbes-Faulkner, D. Pitt, J. H. Norton, and A. D. Thomas.** 2002. *Propionibacterium australiense* sp. nov. derived from granulomatous bovine lesions. Anaerobe **8:**41–47.
3. **Bernard, K. A., M. Bellefeuille, and E. P. Ewan.** 1991. Cellular fatty acid composition as an adjunct to the identification of asporogenous, aerobic gram-positive rods. J. Clin. Microbiol. **29:**83–89.
4. **Cherry, W. B., G. W. Gorman, L. H. Orrison, C. W. Moss, A. G. Steigerwalt, H. W. Wilkinson, S. E. Johnson, R. M. McKinney, and D. J. Brenner.** 1982. *Legionella jordanis*: a new species of *Legionella* isolated from water and sewage. J. Clin. Microbiol. **15:**290–297.
5. **Chunhaswasdikul, B., A. Sukonthaman, K. Lind, and T. Chinachoti.** 1986. Legionella jordanis pneumonia: a case report. J. Med. Assoc. Thai. **69:**283–287.
6. **Den Boer, J. W., and E. P. F. Yzerman.** 2004. Diagnosis of Legionella infection in Legionnaires' disease. Eur. J. Clin. Microbiol. Infect. Dis. **23:** 871–878.

7. **Hsu, S. C., R. Martin, and B. B. Wentworth.** 1984. Isolation of *Legionella* species from drinking water. Appl. Environ. Microbiol. **48:**830–832.
8. **Huerta, M., H. Castel, I. Grotto, O. Shpilberg, M. Alkan, and I. Harman-Boehm.** 2003. Clinical and epidemiologic investigation of two Legionella-Rickettsia co-infections. Isr. Med. Assoc. J. **5:**560–563.
9. **Ratcliff, R. M., J. A. Lanser, P. A. Manning, and M. W. Heuzenroeder.** 1998. Sequence-based classification scheme for the genus *Legionella* targeting the *mip* gene J. Clin. Microbiol. **36:**1560–1567.
10. **Roig, J., and J. Casal.** 2002. Is serological diagnosis of legionnaires' disease a reliable method? Diagn. Microbiol. Infect. Dis. **43:**171–172.
11. **Stout, J. E., and V. L. Yu.** 1997. Legionellosis. N. Engl. J. Med. **337:**682–687.
12. **Tang, P., and C. Krishman.** 1993. Legionellosis in Ontario, Canada: laboratory aspects. *In* J. M. Barbaree, R. F. Breiman, and A. P. Dufour (ed.), *Legionella*: current status and emerging perspectives. ASM Press, Washington, DC.
13. **Thacker, W. L., H. W. Wilkinson, R. F. Benson, S. C. Edberg, and D. J. Brenner.** 1988. *Legionella jordanis* isolated from a patient with fatal pneumonia. J. Clin. Microbiol. **26:**1400–1401.
14. **Wilkinson, H. W.** 1988. Hospital-laboratory diagnosis of *Legionella* infections. Centers for Disease Control and Prevention, Atlanta, GA.