

## Acid-Fast Bacilli in Sputum: a Case of *Legionella micdadei* Pneumonia

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Received 27 May 1986/Accepted 11 September 1986

***Legionella micdadei* has been implicated as a cause of nosocomial pneumonia. There are no reports of *L. micdadei* pneumonia diagnosed by acid-fast stain of expectorated sputum. We report a case of *L. micdadei* pneumonia in which expectorated sputum harbored acid-fast bacteria that reacted specifically with fluorescent antiserum to *L. micdadei*, confirmed by culture. In a patient at risk for nosocomial infection, the differential diagnosis of a positive sputum stain for acid-fast bacilli should include *L. micdadei* in addition to mycobacteria. Therapy for *L. micdadei* infection should be considered pending confirmation of the diagnosis.**

*Legionella micdadei* (Tatlock, HEBA, and Pittsburgh pneumonia agent [5]) is a rare etiological agent of hospital-acquired pneumonia in immunocompromised patients. Its identification and role as a human pathogen were established in 1979 (6), leading to the retrospective identification of the rickettsialike organism obtained in 1943 from guinea pigs inoculated with blood from a patient with Fort Bragg fever (10). This bacterium has been reported to react with modified acid-fast stain when used with human lung imprints or tissues (3, 7-9). No Ziehl-Neelson- or Kinyoun-stained acid-fast bacteria subsequently identified as *L. micdadei* have been reported in sputum specimens of patients. We report the case of an immunocompromised patient whose sputum specimen contained numerous acid-fast bacteria which differed morphologically from mycobacteria and were identified by direct fluorescent antibodies (DFAs) and culture as *L. micdadei*.

A 75-year-old female was admitted for evaluation of a bleeding diathesis which proved to be secondary to a circulating factor 8 anticoagulant. Treatment included factor 9, prednisone, and cyclophosphamide. Her course was complicated by urosepsis with *Enterobacter cloacae*, for which she was treated with intravenous cefotaxime. On hospital day 22, the patient complained of right-side pleuritic pain. She had a nonproductive cough and a rectal temperature of 38.5°C. A chest roentgenogram showed right upper lobe and right lower lobe infiltrates. The leukocyte count was 2,100/mm<sup>3</sup>, with 61% neutrophils, 4% band forms, 7% lymphocytes, 1% eosinophils, and 21% atypical lymphocytes. A sputum sample was unobtainable at this time. The patient was treated with cefazolin and tobramycin but remained febrile. On hospital day 26, a Gram stain of her sputum showed numerous neutrophils, gram-positive cocci in clusters, pairs, and chains, and gram-negative bacilli. A Kinyoun stain of her sputum showed many acid-fast bacilli (AFB) (Fig. 1). Treatment was begun with isoniazid, rifampin, and ethambutol. The patient did not improve, and on hospital day 30, intravenous erythromycin (4 g/day) was begun for the possibility of *Legionella* infection. An expectorated sputum sample and bronchial washings obtained at bronchoscopy were positive for *L. micdadei* by DFA examination. Over the next 2 days, chest roentgenograms showed clearing of the right lower lobe while the right upper lobe

infiltrate persisted. The patient's course was complicated by progressive obtundation, rising liver function test results, and renal failure. On day 41, she had a percutaneous drainage of her gallbladder, and on day 45 she died. Post-mortem examination revealed acute necrotizing and hemorrhagic pneumonia of the right upper lobe, a thrombus in the right main stem bronchus, and acute necrotizing and ulcerative cholecystitis. *L. micdadei* was demonstrated in the lungs by the DFA technique. Cultures were negative for *L. micdadei* on buffered charcoal-yeast extract agar (BCYE; BBL Microbiology Systems, Cockeysville, Md.).

Expectorated sputum was stained directly by the Kinyoun method. The sputum was then examined by the DFA technique (1) using fluorescein-conjugated antisera to *Legionella* spp. provided by the Centers for Disease Control in Atlanta, Ga. Both bronchoscopic specimens and autopsy tissues were examined for AFB, yeasts, and the usual bacteria by prescribed methods (4). In addition, all specimens, including sputa, were cultured on BCYE in a 5% CO<sub>2</sub> atmosphere at 35°C at 80% relative humidity. *Legionella* identification was based on colonial morphology on BCYE, a weak oxidase reaction, a positive catalase reaction, and the absence of gelatinase, β-lactamase, and hippurate hydrolysis (2). Direct fluorescence occurred with *L. micdadei* antiserum exclusively. Cellular fatty acid composition and ubiquone profiles, kindly provided by C. Wayne Moss, Center for Infectious Diseases, Centers for Disease Control, Atlanta, Ga., confirmed the identification of the isolate as *L. micdadei*.

The patient had developed a rapidly progressive pneumonia while on immunosuppressive therapy. An acid-fast stain of the sputum showed numerous AFB, and the patient was initially thought to have tuberculosis, an impression reinforced by a history of tuberculosis in family contacts. The aberrant microscopic morphology of the AFB led to DFA testing of the sputum and bronchial washings with a battery of *Legionella* antisera; they reacted strongly with *L. micdadei* antibodies, whereas no reaction was elicited with available antisera against *L. pneumophila* serotypes 1 through 6, *L. longbeachae*, *L. bozemanii*, *L. dumoffii*, and *L. gormanii*. The diagnosis was further substantiated by the isolation of the bacterium on BCYE. However, growth on BCYE resulted in the loss of the acid-fast property, as reported previously (3). No evidence of tuberculosis was found on culture or by tissue examination.

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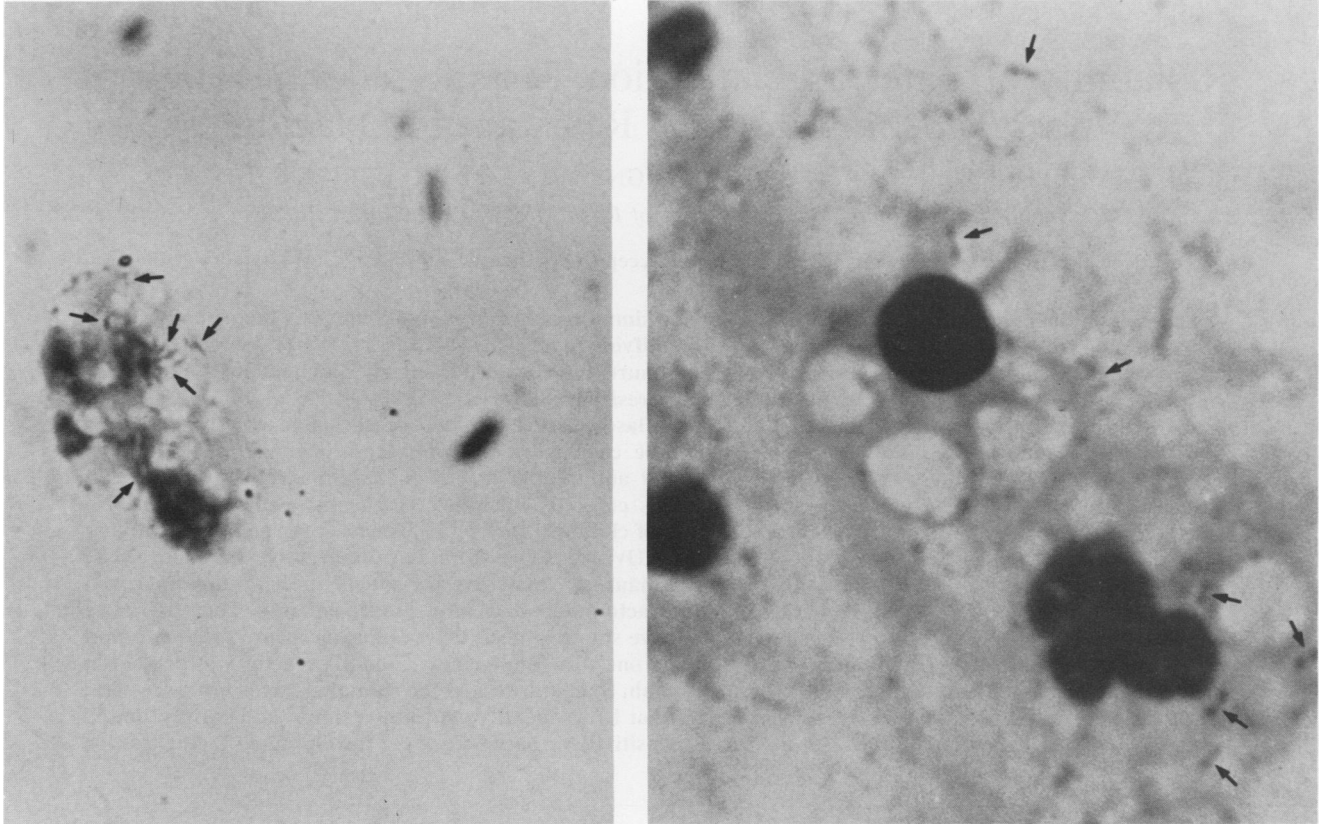


FIG. 1. Kinyoun stain of sputum. Note intracellular acid-fast bacteria (arrows). Left: Original magnification,  $\times 1,000$ ; right: original magnification,  $\times 2,500$ .

The diagnosis of infection with *L. micdadei* is often not obvious. In the 26 cases described in Pittsburgh, several patients had neither cough nor sputum production (6). Diagnosis can be made by DFA testing of sputum or bronchial washings of lung tissue and must be confirmed by culturing the organism on BCYE. In our case, examination of sputum for acid-fast organisms that were obviously not mycobacteria proved to be helpful in making the diagnosis.

To our knowledge, this is the first report of the diagnosis of *L. micdadei* pneumonia made from an expectorated sputum sample by acid-fast stain. Clinicians should be alerted to the possibility of *L. micdadei* when AFB of suspect morphology are seen on sputum Kinyoun or Ziehl-Neelson stains in a patient who is at risk for a nosocomial pneumonia. In addition to antituberculous medication, erythromycin should be considered pending DFA, culture, and serological confirmation of *L. micdadei* infection.

We gratefully acknowledge the assistance of William Pasculle and Wayne Moss for confirming the identification of the organism, K. Jagathambal and Leonard Rossoff for allowing us to report data on their patient, and Carol Bertsch for secretarial assistance.

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