

Isolation of *Legionella pneumophila* from the blood of a patient with Legionnaires' disease

Ronald S. Martin,*† PhD, DipBact
 Thomas J. Marrie,*† MD,
 FRCP[C]
 Linda Best,* BSc
 Robert K. Sumarah,* MSc
 Ruth Peppard,* ART

Legionella pneumophila is rarely isolated from blood cultures. Presently most cases of Legionnaires' disease are diagnosed retrospectively from the results of indirect fluorescent antibody tests, which possess inherent disadvantages. An 81-year-old woman with a history of diabetes mellitus presented symptoms of Legionnaires' disease. Five hours before her death 1.5 mL of blood was withdrawn from a scalp vein and seeded to a culture medium. Following incubation for 3 days *L. pneumophila* serotype 1 was isolated.

L'isolement de *Legionella pneumophila* à partir du sang est rare. Actuellement, le diagnostic de maladie du légionnaire se fait surtout en rétrospective par la technique indirecte des anticorps fluorescents; cette méthode a ses inconvénients. Observation d'une femme diabétique âgée de 81 ans dont les symptômes évoquent cette maladie. Une hémoculture à partir d'un échantillon de 1,5 mL prélevé dans une veine épicroânienne 5 heures avant le décès donne au bout de 3 jours d'incubation une *L. pneumophila* du sérotype 1.

Legionnaires' disease is an acute respiratory tract infection caused by *Legionella pneumophila*, a small, very fastidious, gram-negative organism.^{1,2} Thus far, the most fruitful specimens for isolating the organism

have been derived from open-lung biopsy tissue, pleural exudate, bronchoscopy tissue and, occasionally, sputum.^{3,4} Although the profound lobar pneumonia and other pathological features of the disease all suggest an early bacteremic phase, it has been difficult to isolate the organism from blood, and very few have reported doing so.⁵⁻⁷ This is unfortunate because isolating the organism directly from the patient's blood makes the diagnosis more definitive and, since the organism is isolated in pure culture, makes subsequent technical steps less cumbersome, thereby adding to the speed of diagnosis. (Legionnaires' disease is now usually diagnosed retrospectively from the results of indirect fluorescent antibody tests, which not only take time to develop but also depend

on the patient's ability to mount an immunologic response.)

We report the isolation of *L. pneumophila* serogroup 1 from a blood sample taken 5 hours before our patient's death and cultured; when the specimen was taken, however, the patient was receiving therapeutic doses of erythromycin, to which the recovered organism proved to be sensitive. The organism was also isolated from endoscopy specimens.

Case report

Clinical course

An 81-year-old woman was brought to the emergency department of our hospital, having been found lying unresponsive on the

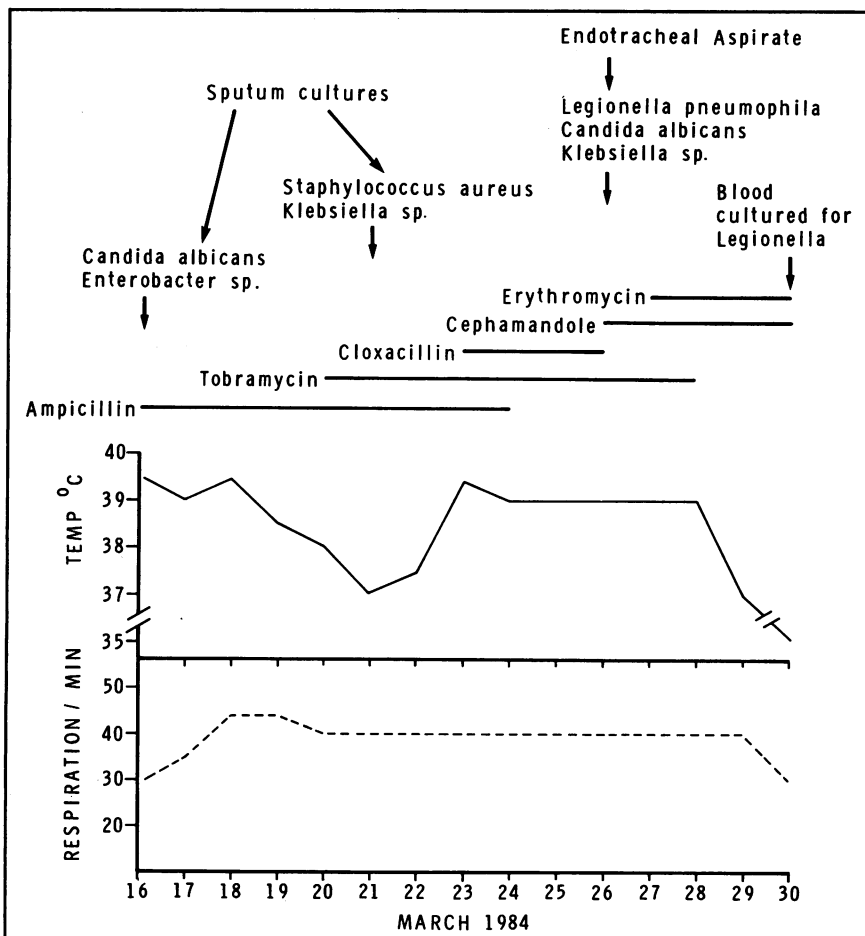


Fig. 1—Clinical course of patient with Legionnaires' disease during hospital stay until time of death.

From *the Victoria General Hospital, Halifax and †the Faculty of Medicine, Dalhousie University, Halifax

Reprint requests to: Dr. Ronald S. Martin, D.J. Mackenzie Building, Victoria General Hospital, 5788 University Ave., Halifax, NS B3H 1V8

floor of her home. Her daughter, with whom she had been living for the previous 3 months, reported that her mother had been becoming progressively confused and had fallen repeatedly during the 3 days before admission. Five years earlier diabetes mellitus had been diagnosed in the patient, who was currently being treated with chlorpropamide.

When she was admitted she had an oral temperature of 38.5°C, rales at the lung bases, a fracture of her left radius and a plasma glucose level of 2.0 mmol/L. She became responsive within minutes of being given glucose intravenously, was admitted to the orthopedic surgical service and later that day underwent open reduction and internal fixation of her radius. When her temperature later rose to 39.5°C antibiotic therapy was instituted (Fig. 1).

The next day the patient was short of breath; the results of a physical examination were sugges-

tive of congestive heart failure, and she was treated with diuretics, given intravenously. Over the next week she remained short of breath, and when examined by one of us (T.J.M.) 10 days after admission she was found to be severely ill, with a respiratory rate of 44/min and an infiltrate involving most of the right lung and the base of the left lung. Rales were present in the lung bases. There was no evidence of congestive heart failure; indeed, she was dehydrated. She was drowsy but responded appropriately to verbal stimuli.

The patient was unable to expectorate, so her trachea was suctioned under direct vision, and a moderate amount of mucoid material was obtained. Gram-staining of this material showed 10 squamous epithelial cells per low power field, occasional gram-negative rods and a moderate number of yeast cells. At this point therapy with cloxacillin and tobramycin was discontinued and

therapy with cefamandole begun. Culture of the tracheal aspirate grew *Klebsiella* sp. and *Candida albicans*. Legionnaires' disease was suspected on clinical grounds, and therapy with erythromycin, 500 mg given intravenously every 6 hours, was begun. Three days later *L. pneumophila* was grown from the endotracheal aspirate.

The patient's condition continued to deteriorate, and 3 days later the dose of erythromycin was increased to 1 g every 6 hours, given intravenously. The next morning 1.5 mL of blood was taken from a scalp vein and cultured for *L. pneumophila*. The patient died 5 hours later. Her clinical course is illustrated in Fig. 1.

Towards the end of her hospital stay the woman had also suffered from gastrointestinal hemorrhage and diarrhea. Her blood creatinine level had risen from 101 $\mu\text{mol/L}$ at the time of admission to 143 $\mu\text{mol/L}$

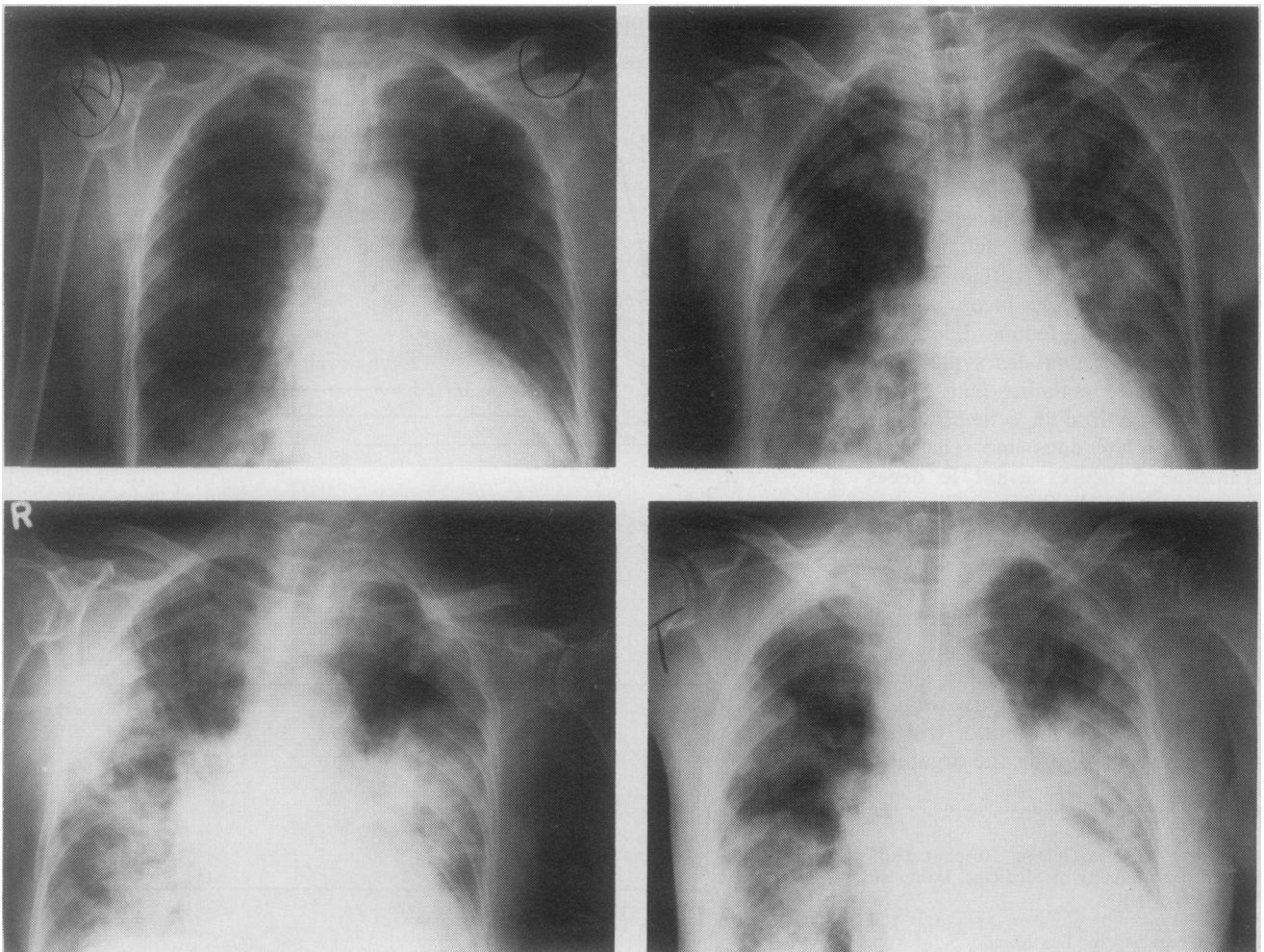


Fig. 2—Chest roentgenograms made Mar. 17 (top left), 23 (top right), 28 (bottom left) and 30 (bottom right), showing rapidly progressive pneumonia.

2 days before she died, by which time her urine output had decreased to 500 mL/24 h. Other important laboratory findings had included a leukocyte count at the time of admission of $3.0 \times 10^9/L$, with 75% polymorphonuclear leukocytes, 9% monocytes, 10% lymphocytes and 6% band forms; just before she died her leukocyte count had been $7.0 \times 10^9/L$, with 79% polymorphonuclear leukocytes, 1% monocytes, 1% lymphocytes and 19% band forms.

At the time of admission a chest roentgenogram had shown a small subsegmental infiltrate in the lower lobe of the right lung. Fig. 2 shows four chest roentgenograms made during the patient's hospital stay, from which it is evident that she had rapidly progressive pneumonia despite antibiotic therapy.

Isolation of L. pneumophila from blood

The blood was introduced into 10 mL of a broth that contained per litre of water 15.0 g of proteose-peptone (Oxoid, K.C. Biologicals, Inc., Lenexa, Kansas), 5.0 g of yeast extract, 2.5 g of liver extract, 0.125 g of $Fe_4(P_2O_7)_3$ (soluble), 1.0 g of α -ketoglutarate (potassium salt), 10.0 g of ACES buffer ([N-(2-acetamidol)]-2-amino ethane sulfonic acid), 0.5 g of sodium polyanethyl sulfonate and 0.4 g of L-cysteine-hydrochloride. After it had incubated at 37°C for 3 days the

broth was subcultured to buffered charcoal yeast extract agar and was incubated at 37°C for 3 days in an atmosphere of 4% CO_2 . After incubation the plates were found to contain three colonies, approximately 2 mm in diameter, of organisms whose morphologic features resembled those of *L. pneumophila*. Direct fluorescence microscopy confirmed that they were *L. pneumophila* serogroup 1.

Discussion

This is an interim report describing our first isolation of *L. pneumophila* serogroup 1 from a sample of blood cultured by the technique described, in a case in which the organism was also recovered from cultures of endotracheal secretions. To date our studies have shown that the broth used is satisfactory for blood culture but does not give good results when nonblood specimens (e.g., pleural fluid) are used. Moreover, although the broth adequately supports the growth of laboratory-stock strains of *L. pneumophila* (the only organism we have tested), we must add 10% blood to get uniform isolation of the organism from patients' specimens. Finally, we are not certain why we recovered the organism from this blood sample, since a sensitivity test showed that the organism was markedly sensitive to erythromycin (which cleared a zone 45 mm in diameter at a con-

centration of 15 $\mu g/disc$). It may be that the organisms were inside the body of a monocyte.

This work was funded by a grant from the Nova Scotia Lung Foundation.

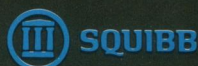
References

1. Kirby BD, Snyder KM, Meyer RD et al: Legionnaires' disease: clinical features of 24 cases. *Ann Intern Med* 1978; 89: 297-309
2. Jenkins P, Miller AC, Osman J et al: Legionnaires' disease: a clinical description of thirteen cases. *Br J Dis Chest* 1979; 73: 31-38
3. Feeley JC, Gorman GW, Weaver RE et al: Primary isolation media for Legionnaires' disease bacterium. *J Clin Microbiol* 1978; 8: 320-325
4. Greaves PG, Sharp G, Macrae AD: Isolation of *Legionella pneumophila* [C]. *Lancet* 1979; 1: 551-552
5. Edelstein PH, Meyer RD, Finegold SM: Isolation of *Legionella pneumophila* from blood. *Ibid*: 750-751
6. Macrae AD, Greaves PG, Platts P: Isolation of *Legionella pneumophila* from blood culture. *Br Med J* 1979; 2: 1189-1190
7. Dorn GL, Barnes WR: Rapid isolation of *Legionella pneumophila* from seeded donor blood. *J Clin Microbiol* 1979; 10: 114-115

Vafia

In congestive heart failure,
CAPOTEN
(captopril)

*The freedom
to live again*



Those non-urgent 'emergencies'

Emergency calls often turn out to be false alarms. If you consider that too many such non-emergencies are disrupting your schedule, tell your nurse to question the patient about his symptoms and the history of the illness. Often the patient will end up deciding it is not as serious as he thought. However, if he still feels he needs your immediate attention, the nurse can either switch the call to you or consult with your herself.