too may our thinking regarding management of patients colonized with CPE.[8]

Prospective studies using innovative laboratory techniques are needed on patients with CPE during outbreaks and in endemic settings over periods of time, while in the hospital and after discharge. These studies will help us further understand the natural history of CPE colonization and determine the host/patient, environmental, and microbial factors that may help us predict which patients remain transiently, intermittently, or permanently colonized. This work is required to scientifically inform infection prevention and control strategies on the use of contact precautions and to facilitate risk stratification of patients previously identified as CPE colonized. This research becomes increasingly important as the number of patients with CPE increases, especially in settings where single room/isolation facilities are limited.

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Hilary Humphreys, MD FRCPI;[1,2]
Anna-Rose Prior, MB FRCPath[1,2]

Affiliation: 1. Department of Clinical Microbiology, the Royal College of Surgeons in Ireland, Dublin, Ireland; 2. Department of Microbiology, Beaumont Hospital, Dublin, Ireland.

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A Fatal Case of Nosocomial Legionnaires' Disease: Implications From an Extensive Environmental Investigation and Isolation of the Bacterium From Blood Culture

To the Editor—Legionella pneumophila (*Lp*) serogroup 1 is a well-known cause of hospital-acquired pneumonia. Extrapulmonary manifestations as well as *Lp* bacteremic pneumonia are rare and occur mainly in immune-compromised subjects.[1–5]

Potable water has been implicated in nosocomial cases of *Lp* infection via inhalation of contaminated aerosols from hot-water systems.[6] Culture is the gold standard for the diagnosis of *Legionella* infection, even though culture-proven legionellosis cases are not frequently reported and documented cases of bacteria isolation from the blood are even less common.[7]

On January 5, 2014, a 58-year-old woman with a clinical history of alcoholic liver cirrhosis was admitted for worsening dyspnea to the Emergency Department of IRCCS AOU San Martino–IST Hospital, Genoa, Italy. On admission, the patient presented peripheral edema and signs of portal hypertension. Chest and abdominal radiograph did not detect pleuroparenchymal lung lesions.

The same day, the patient was transferred to the gastroenterologic unit where she stayed until February 6, 2014. She underwent 3 paracentesis procedures, a transjugular intrahepatic portosystemic shunt, and 3 angiography controls because of a progressive deterioration in liver function.

On February 6, 2014, the patient experienced a severe respiratory failure due to acute pulmonary edema. She was intubated and chest radiograph (Figure 1) detected pleuroparenchymal lung lesions compatible with pneumonia.

Blood cultures (Bactec Plus aerobic; BD) were performed and the patient was empirically treated with piperacillin-tazobactam. No specific diagnostic test for *Legionella* (culture, urinary antigen, serology) was performed. On February 7, 2014, the patient died because of respiratory and multiorgan failure.

After 6 days, blood culture became positive and subculture on blood agar (Kima; Megalab) and chocolate-enriched agar plates (Kima) was performed, showing the growth of colonies

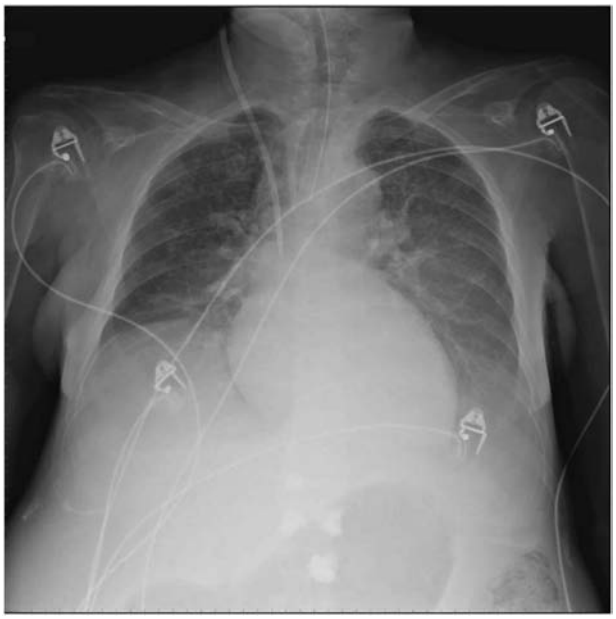

FIGURE 1. Chest radiograph of the patient performed during the hospital stay in the intensive care unit 1 day before death.

that, unexpectedly, turned out to be *Lp* by matrix-assisted laser/desorption-ionization time-of-flight mass spectrometry identification technique (bioMérieux).

Extensive environmental investigation was performed by the hospital's hygiene unit to identify the source of the infection.

No sources of infection could be detected in the operating room and no cooling towers or evaporative condensers were present in the neighborhood of the hospital. Invasive procedures, such as transjugular intrahepatic portosystemic shunt and paracentesis, were excluded as potential sources of infection because of use of disposable equipment and absence of water contact.

Hot- and cold-water samples were collected from the rooms (washbasins, showers, heaters) of all the wards where the patient was hospitalized, and analyzed at the *Legionella* Regional Reference Laboratory, according to national guidelines. All the samples had negative results ($Lp < 100$ colony-forming units/L) as expected, as the hospital's water system was continuously disinfected by a chlorine dioxide system and monitored for biocide level monthly.

Concomitantly, the clinical isolate and an aliquot of the collected water samples were sent to the *Legionella* National Reference Laboratory. A latex agglutination test (Oxoid) confirmed the identification of the clinical isolate as *Lp* serogroup 1. Typing assays performed by monoclonal antibodies and sequence-based typing typed the clinical strain as Olda and sequence type 154, respectively.[8] Furthermore, all water samples were analyzed by real-time polymerase chain reaction assay, using the iQ-Check quantification kit (BioRad), resulting in *Legionella* positive (7×10^2 to 2×10^3 genomic units/L).

Further sampling was carried out by the hygiene unit, increasing the volume of analyzed water to 2 liters and the amount spread on selective medium to 0.5 mL. The increased volume of the water tested allowed the isolation of *Lp* serogroup 1 at very low concentration (20 colony-forming units/L) from the hot-water samples collected from one of the taps routinely used by the patient. Monoclonal antibody typing and sequence-based typing performed at *Legionella* National Reference Laboratory showed that the environmental and clinical isolates were related and were both Olda monoclonal antibody subgroup and sequence type154, thus revealing the source of infection.

In the case here described, *Lp* infection was diagnosed postmortem by positive blood culture, whereas neither urine antigen assay nor sputum culture were performed.

The occurrence of a *Legionella* infection 31 days after the patient's hospital admission was consistent with a nosocomial case, thus justifying the extensive environmental investigation that eventually led to the isolation of *Lp* from the tap of the washbasin used by the patient. This route of transmission of Legionnaires' disease, rarely described in literature,[9] was probably favored by the severe immune deficiency of the patient, a consequence of the alcoholic liver cirrhosis at its final stage. The isolation of *Legionella* from blood was also remarkable.

Lp growth in blood culture is seldom investigated, although this occurrence is not so infrequent especially in aerobic condition.[10] The scanty use of blood culture probably comes from the bias that *Legionella* is not able to grow in broth media utilized for blood culture. Indeed, these media, as well as blood and chocolate-enriched agar plates used for *Legionella* subculture, are lacking in ferric pyrophosphate and L-cysteine essential for *Lp* growth. However, we can speculate that small amount of the essential nutrients might be supplied by erythrocytes and by the metabolism of other blood cells, allowing the growth of *Legionella* in blood culture.

In conclusion, this case highlights that Legionnaires' disease should be always suspected in severely immune-compromised patients when clinical and radiologic findings are suggestive for pulmonary infection, and the patient has been hospitalized for a long period, even in the absence or near-absence of *Legionella* water system contamination provided by a routine surveillance program. Another lesson learned by this case is that the adoption of blood culture, in addition to respiratory secretion culture, to isolate *Legionella* needs to be considered.

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Paolo Durando, PhD;[1]
Andrea Orsi, MD;[2]
Cristiano Alicino, PhD;[2]
Carola Tinteri, BiolD;[2]
Annamaria Di Bella, ICN;[2]
Maria Caterina Parodi, MD;[3]
Anna Marchese, BiolD;[4]
Paola Rosaria Gritti, MD;[4]
Stefano Fontana, PhD;[5]
Maria Cristina Rota, MD;[5]
Maria Scaturro, PhD;[5]
Maria Luisa Ricci, BiolD[5]

Affiliation: 1. Department of Health Sciences (DiSSal), Postgraduate School in Occupational Medicine, University of Genoa, Genoa, Italy; Occupational Medicine Unit and Hospital Infection Control Unit, IRCCS University Hospital San Martino–IST National Institute for Cancer Research, Genoa, Italy; 2. Department of Health Sciences (DiSSal), University of Genoa, Genoa, Italy; Hygiene Unit and Hospital Infection Control Unit, IRCCS University Hospital San Martino–IST National Institute for Cancer Research, Genoa, Italy; 3. Gastroenterology and Digestive Endoscopic Unit, IRCCS University Hospital San Martino–IST National Institute for Cancer Research, Genoa, Italy; 4. Department of Surgical Sciences (DiSC), University of Genoa, Genoa, Italy; Microbiology Unit, IRCCS University Hospital San Martino–IST National Institute for Cancer Research, Genoa, Italy; 5. Istituto Superiore di Sanità (ISS, National Institute of Health), Department of Infectious Parasitic and Immune-mediate diseases and National Centre for Epidemiology, Surveillance and Health Promotion, Rome, Italy

Address correspondence to Andrea Orsi, MD, Department of Health Sciences (DiSSal), University of Genoa, Via A. Pastore 1, 16132 Genoa, Italy (andrea.orsi@unige.it).

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Infectious Diseases of High Consequence and Personal Protective Equipment: A Didactic Method to Assess the Risk of Contamination

To the Editor—Infectious diseases of high consequence are serious threats to human health with no specific prophylaxis or treatment available. Patients may develop severe symptoms and require critical care. The protection of healthcare workers (HCWs) through personal protective equipment (PPE) and isolation of contagious patients are the 2 main principles to reduce the risk of spreading of infectious diseases of high consequence.[1] Since March 2014, more than 27,000 cases of Ebola virus disease and 11,000 deaths have been reported in West Africa.[2] Healthcare workers are between 21 and 32 times more likely to be infected with Ebola virus than people in the general population.[3] In light of this current outbreak and its high case-fatality rate, a broad range of challenges were reported, including conflicting PPE removal protocols and gaps in training and supplies.[4] This is of major concern because Ebola virus can persist on surfaces for up to 5 days[5] and the skin and clothing of HCWs can become contaminated. The fear of undetected contamination may result in increased stress levels for HCWs. In this context, the need for adherence to safe and validated protocols for removal of PPE is clear. Moreover, it has been shown, by evaluation of the errors, that repeated training achieved better proficiency.[6] However, to the best of our knowledge, the impact of training courses has not yet been assessed by an objective method.

During an educational training program, we undertook a systematic evaluation of the risk of contamination with a fluorescent powder (Hygikit; Voussert). We conducted a series of 47 care simulations. Nurses, nursing assistants, and physicians from 3 different wards (the infectious disease unit, emergency department, and intensive care unit) who use PPE and interact with highly contagious patients were voluntarily included. The study was conducted at a university hospital with a specialized treatment center for infectious diseases of high consequence. The standard components of PPE included