Outbreak of Legionnaires' disease in immunosuppressed patients at a cancer centre: usefulness of universal urine antigen testing and early levofloxacin therapy

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ABSTRACT

This report describes an outbreak of Legionnaires' disease in severely immunosuppressed patients hospitalised at a cancer centre. Universal urine antigen testing and early levofloxacin therapy appeared to lower case fatality rates in comparison with previous reports concerning this high-risk population. This diagnostic and therapeutic strategy should be considered when facing a nosocomial outbreak of Legionnaires' disease in immunosuppressed hosts.

Keywords Immunosuppressed patients, Legionnaires' disease, levofloxacin therapy, nosocomial outbreak, pneumonia, urine antigen testing

Original Submission: 23 December 2006; **Revised Submission:** 22 May 2007; **Accepted:** 14 June 2007

Clin Microbiol Infect 2007; **13**: 1125–1128
10.1111/j.1469-0691.2007.01805.x

A reduction in case fatality rates among non-immunosuppressed patients with Legionnaires' disease (LD) has recently been observed, suggesting that new management strategies may result in improved outcome [1–6]. These new approaches include the use of urinary antigen testing for rapid diagnosis in combination with highly active antimicrobial agents against *Legionella*, e.g., azithromycin and fluoroquinolones [7,8]. However, little is known about the potential impact of these new strategies on immunosuppressed patients, who have the highest mortality rates. Accordingly, this report describes the impact of these new diagnosis and treatment strategies on an outbreak of LD caused by *Legionella pneumophila* serogroup 1 among severely immunosuppressed patients hospitalised at a cancer centre.

The outbreak of LD occurred over a 4-week period among patients in a 300-bed tertiary referral cancer centre for adults in Barcelona, Spain. The hospital opened in 1991 and no previous cases of nosocomial LD had been observed. The first case of *Legionella* pneumonia was detected on 24 February 2005. When two further cases were diagnosed within 48 h, *Legionella* urinary antigen testing was instituted for all patients hospitalised in the same wards as the first three cases, and for any patient presenting with a new onset of fever or respiratory symptoms ($n = 155$ patients). *L. pneumophila* serogroup 1 antigen in urine was detected using the NOW *Legionella* Urinary Antigen test (Binax, Scarborough, MA, USA). The test was performed on a 24-h basis, 7 days a week. To exclude false-positive results, all urine samples were boiled and concentrated. All patients with a positive urine antigen test underwent a chest radiograph and were given early intravenous levofloxacin therapy, whether or not they presented with pneumonia.

Samples for microbiological investigations were obtained from patients with pneumonia whenever possible, and also from the water system supply. All samples were cultured on selective buffered charcoal yeast extract-α medium, and pulsed-field gel electrophoresis typing was performed for all isolates [9].

The attributable case fatality rate was defined as death during symptomatic infection or as a consequence of its complications. The overall case fatality rate was defined as death from any cause within 28 days of the diagnosis. Table 1 shows the characteristics of the 12 patients involved in the outbreak. Three patients had profound neutropenia ($<0.1 \times 10^9$ cells/L), and one also had invasive pulmonary aspergillosis; this patient's condition was complicated by diffuse alveolar haemorrhage, requiring mechanical ventilation.

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Table 1. Baseline characteristics, clinical features and outcome of 12 immunosuppressed patients involved in a nosocomial outbreak of Legionnaires' disease in a cancer centre

Patient no.	Age (years)	Gender	Underlying disease	Immunosuppressive therapy	Clinical symptoms	Radiological findings	Treatment	Outcome/ time to death
1	41	Male	Disseminated, cancer, central nervous system metastases	Corticosteroids	Dyspnoea, diarrhoea	Left lower lobe infiltrate	Clarithromycin (500 mg twice daily)	Died/<24 h
2	81	Male	Lung cancer	Corticosteroids	Fever, respiratory symptoms	Right upper upper lobe infiltrate	Levofloxacin (500 mg daily)	Died/18 days
3	41	Male	Aplastic anaemia	Corticosteroids, cytotoxic chemotherapy, cyclosporine	Fever, respiratory symptoms	Bilateral infiltrates, cavitation	Levofloxacin (500 mg twice daily), cyclosporine stopped	Survived
4	61	Male	Lung cancer	Corticosteroids, radiotherapy	Dyspnoea	Right lower lobe infiltrate	Levofloxacin (500 mg daily) ↓ corticosteroids	Survived
5	43	Female	Acute myeloid leukaemia, neutropenia	Cytotoxic chemotherapy	Fever, respiratory symptoms	Multilobar infiltrates on the left lung	Levofloxacin (500 mg twice daily)	Survived
6	74	Female	Lymphoma, neutropenia	Corticosteroids, cytotoxic chemotherapy	Malaise, diarrhoea	Left lower lobe infiltrate	Levofloxacin (500 mg daily), ↓ corticosteroids	Survived
7	68	Male	Chronic lymphocytic leukaemia, haemolytic anaemia	Corticosteroids	Asymptomatic	Left lower lobe infiltrate	Levofloxacin (500 mg daily), ↓ corticosteroids	Survived
8	57	Female	Acute lymphocytic leukaemia, neutropenia, invasive pulmonary aspergillosis	Corticosteroids, cytotoxic chemotherapy	Fever, respiratory symptoms, diffuse alveolar haemorrhage	Bilateral infiltrates	Levofloxacin (500 mg twice daily)	Intensive care unit admission, mechanical ventilation, survived
9	68	Female	Lymphoma	Corticosteroids, cytotoxic chemotherapy	Fever	Right lower lobe infiltrate	Levofloxacin (500 mg daily)	Survived
10	54	Female	Lymphoma	Corticosteroids	Asymptomatic	No abnormal findings	Levofloxacin (500 mg daily)	Survived
11	71	Male	Pancreatic cancer	Corticosteroids	Asymptomatic	No abnormal findings	Levofloxacin (500 mg daily)	Died/21 days
12	57	Female	Lung cancer	Corticosteroids, cytotoxic chemotherapy	Fever, respiratory symptoms	Left lower lobe infiltrate	Levofloxacin (500 mg daily)	Survived

Ten of the 12 patients involved in the outbreak presented with pneumonia. The most frequent clinical symptoms in these ten patients were fever and respiratory symptoms, although two patients presented with atypical symptoms and one was asymptomatic, despite a chest radiograph demonstrating a new pulmonary infiltrate. Two additional patients were completely asymptomatic at the time when urine antigen testing was performed. Patient no. 3 presented with a large lung cavitation, persistent fever and persistent positive sputum cultures, but his condition steadily improved, with eventual full recovery.

The index patient (no. 1) was diagnosed initially with atypical pneumonia and was treated with clarithromycin. Despite early initiation of macrolide therapy, the patient's clinical condition rapidly worsened, with death occurring within 24 h of the diagnosis of pneumonia. Once an outbreak of LD was suspected and universal urine antigen testing was performed, all patients were treated with levofloxacin, including the two patients with asymptomatic infection. Two more patients died within 28 days, but their deaths were considered to be unrelated to the infection. One patient (no. 2) had a rapid progression of underlying lung cancer with liver metastases, with death occurring on the 18th day of levofloxacin treatment, at which time all signs and symptoms of pneumonia had disappeared. The second patient with a positive urine antigen test without pneumonia (no. 11) was treated with levofloxacin as an outpatient, but died 21 days later during a subsequent period of hospitalisa-

tion because of local cancer progression and massive gastrointestinal bleeding. The attributable case fatality rate was 8.3% (one of 12 patients), and the overall case fatality rate (≤28 days) was 25% (three of 12 patients).

L. pneumophila serogroup 1 was isolated from five sputum samples obtained from five patients with pneumonia, and also from several samples obtained from the water system supply. Pulsed-field gel electrophoresis typing indicated that all the isolates from water systems were identical and were clonally related to the isolates from sputum samples. The outbreak, which lasted 4 weeks, was successfully controlled by measures that included the immediate restriction of water use, and repeated superheating and flushing of the water system. After the outbreak, a copper–silver ionisation unit was installed to provide permanent reservoir disinfection in conjunction with ongoing active surveillance of the water system.

The baseline characteristics of the patients described here are similar to those found in earlier studies of LD in immunosuppressed hosts that reported overall case fatality rates of 50–80% [10,11]. A previous outbreak occurred during 1985 in a nearby hospital among immunosuppressed patients with cancer [10]. At that time, *Legionella* urine antigen testing was not available, and all patients were treated with erythromycin (plus rifampicin in some cases). The case fatality rates in the present study were substantially lower than those in the previous outbreak (attributable case fatality rates of 8% vs. 43%, and overall case fatality rates of 25% vs. 50%, respectively).

The present results are in line with recent reports involving non-immunosuppressed patients with community-acquired LD, indicating that early diagnosis, by means of urine antigen testing and fluoroquinolone therapy, may improve patient outcome [3–6]. In the outbreak reported here, universal urine antigen testing allowed asymptomatic patients to be identified at an early stage of *Legionella* infection. The prompt treatment of these patients may have modified the course of their infection and avoided the development of pneumonia and a more severe disease.

Fluoroquinolones have been shown to be more effective than older macrolides in inhibiting the intracellular growth of *L. pneumophila*, both *in vitro* and in animal models [12–14]. Improved outcome (with respect to time to defervescence and length of hospital stay) for patients treated with levofloxacin has been documented in three observational studies [4–6]. The results of the present study provide additional information regarding the usefulness of early monotherapy with levofloxacin for treatment of LD in severely immunosuppressed patients. However, while the low patient mortality was probably related to both universal urine antigen testing and prompt levofloxacin therapy, the small sample size and uncontrolled nature of the study mean that other testing and therapeutic strategies could have produced similar results. Nevertheless, the results suggest that the combined use of universal urine antigen testing and early treatment with levofloxacin might improve the outcome for severely immunosuppressed patients with LD. This diagnostic and therapeutic strategy merits consideration when facing a nosocomial outbreak of LD involving similar patient populations.

ACKNOWLEDGEMENTS

The study was supported by Ministerio de Sanidad y Consumo, Instituto de Salud Carlos III, Spanish Network for Research in Infectious Diseases (REIPI RD06/0008).

REFERENCES

1. Benin AL, Benson RF, Besser RE. Trends in Legionnaires' disease, 1980–1998: declining mortality and new patterns of diagnosis. *Clin Infect Dis* 2002; **35**: 1039–1046.
2. Plouffe JF, Breiman RF, Fields BS *et al*. Azithromycin in the treatment of *Legionella* pneumonia requiring hospitalisation. *Clin Infect Dis* 2003; **37**: 1475–1480.
3. Yu VL, Greenberg RN, Zadeikis N *et al*. Levofloxacin efficacy in the treatment of community-acquired legionellosis. *Chest* 2004; **125**: 2135–2139.
4. Mykietiuk A, Carratalà J, Fernández-Sabé N *et al*. Clinical outcomes for hospitalized patients with *Legionella* pneumonia in the antigenuria era: the influence of levofloxacin therapy. *Clin Infect Dis* 2005; **40**: 794–799.
5. Blázquez Garrido MR, Espinosa Parra FJ, Alemany Francés L *et al*. Antimicrobial chemotherapy for Legionnaires' disease: levofloxacin versus macrolides. *Clin Infect Dis* 2005; **40**: 800–806.
6. Sabrià M, Pedro-Botet ML, Gómez J *et al*. Fluoroquinolones vs macrolides in the treatment of Legionnaires disease. *Chest* 2005; **128**: 1401–1405.
7. Pedro-Botet L, Yu VL. Legionella: macrolides or quinolones? *Clin Microbiol Infect* 2006; **12** (suppl 3): 25–30.
8. García-Vidal C, Carratalà J. Current clinical management of Legionnaires' disease. *Expert Rev Anti Infect Ther* 2006; **4**: 995–1004.

9. Schoonmaker D, Heimberger T, Birkhead G. Comparison of ribotyping and restriction enzyme analysis using pulsed-field gel electrophoresis for distinguishing *Legionella pneumophila* isolates obtained during a nosocomial outbreak. *J Clin Microbiol* 1992; **30**: 1491–1498.
10. Falguera M, Rufí G, Dorca J, Verdaguer R, Gudiol F, Manresa F. Neumonía por *Legionella pneumophila* en el paciente inmunodeprimido. *Enferm Infecc Microbiol Clin* 1988; **6**: 130–135.
11. Kirby BD, Snyder KM, Meyer RD, Finegold SM. Legionnaires' disease: report of sixty-five nosocomially acquired cases and review of the literature. *Medicine* 1980; **59**: 188–205.
12. Smith RP, Baltch AL, Franke M, Hioe W, Ritz W, Michelsen P. Effect of levofloxacin, erythromycin or rifampicin pretreatment on growth of *Legionella pneumophila* in human monocytes. *J Antimicrob Chemother* 1997; **40**: 673–678.
13. Edelstein PH. Antimicrobial chemotherapy for legionnaires' disease: time for a change. *Ann Intern Med* 1998; **129**: 328–330.
14. Roig J, Rello J. Legionnaires' disease: a rational approach to therapy. *J Antimicrob Chemother* 2003; **51**: 1119–1129.

RESEARCH NOTE

Detection of diverse SCC*mec* variants in methicillin-resistant *Staphylococcus aureus* and comparison of SCC*mec* typing methods

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ABSTRACT

Non-duplicate methicillin-resistant *Staphylococcus aureus* (MRSA) isolates (n = 436), collected from four hospitals located in three Korean cities between 2001 and 2005, were investigated by SCC*mec* typing and multilocus sequence typing (MLST). Variations within SCC*mec*, especially type II, were detected in 165 (37.8%) isolates, and these variants were characterised using four different SCC*mec* typing methods. The predominant SCC*mec* type was a type II variant that differed from type II by the absence of a pUB110 insertion. MLST analysis showed that most of the isolates carrying SCC*mec* variants belonged to ST5.

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Keywords Methicillin-resistant *Staphylococcus aureus*, multilocus sequence typing, SCC*mec* variants, typing

Original Submission: 9 March 2007; **Revised Submission:** 8 May 2007; **Accepted:** 13 June 2007

Clin Microbiol Infect 2007; **13**: 1128–1130
10.1111/j.1469-0691.2007.01806.x

SCC*mec* is a mobile element, comprising the *mec* gene complex, the *ccr* gene complex and the junkyard (J) regions, which can integrate into the *Staphylococcus aureus* chromosome [1,2]. In 2001, Ito *et al*. [1] described three types of SCC*mec* elements (designated I–III), based on the structure of the *mecA* complex and the *ccrAB* allele. Subsequently, SCC*mec* type IV was identified, initially in two community-acquired strains of methicillin-resistant *S. aureus* (MRSA) [2,3]. In 2002, Oliveira and de Lencastre [4] developed a single-step multiplex PCR (M-PCR) method designed to provide maximum resolution of the various structural variants of the SCC*mec* element, based on genes located within the J-regions of SCC*mec* elements. Although this method did not include identification of the *ccrAB* allele, it enabled discrimination of the four major SCC*mec* types and certain variants, such as IA and IIIA. This method is simple and easy to perform, and has therefore been used increasingly, but is limited by its inability to detect the newly identified SCC*mec* type V and subtypes IVa–IVd [5,6]. Subsequently Zhang *et al*. [7] developed a new M-PCR strategy that has the advantage of identifying the J1 region of eight SCC*mec* elements (I, II, III, IVa–IVd and V) simultaneously.

In the present study, the SCC*mec* structural type and sequence type (ST) of 436 clinical isolates of MRSA were determined. The isolates were collected from four hospitals located in three Korean cities between 2001 and 2005. All isolates were tested for phenotypic resistance to oxacillin by the salt agar dilution method according to CLSI (formerly NCCLS) guidelines [8], and for the presence of the *mecA* gene by PCR.

To determine the SCC*mec* structural type, the M-PCR developed by Oliveira and de Lancastre