

Research article

## Community acquired pneumonia due to *Legionella pneumophila* in a tertiary care hospital

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*Sri Lankan Journal of Infectious Diseases* 2016 Vol.6 (2):101-105

DOI: <http://dx.doi.org/10.4038/sljid.v6i2.8112>

### Abstract

**Introduction:** *Legionella pneumophila* can cause severe community acquired pneumonia which may be life threatening. This organism is found in aquatic environments and infection is acquired through inhalation of aerosols. Few studies conducted in Sri Lanka have confirmed the presence of this organism in cooling tower water in Sri Lanka. Published data regarding human cases of legionellosis in Sri Lanka is not available.

**Objective:** To determine the prevalence of community acquired pneumonia due to *L. pneumophila* among patients who required hospital admission and assess the risk factors associated with this infection.

**Methods:** The study was carried out from July 2014 to June 2015 at the Teaching Hospital, Peradeniya. Expecterated sputum or endotracheal secretions and urine specimen were collected within 24 hours of admission after obtaining consent from all adult patients admitted during the study period with community acquired pneumonia. Respiratory specimens, if obtained, were inoculated onto Buffered Charcoal Yeast Extract (BCYE) agar and were inoculated at 35 °C – 37 °C for 7 days and observed for typical colonies. Urine specimens were stored at -20 °C and ELISA test was performed for the detection of *L. pneumophila* serogroup 1a antigen.

**Results:** Eighty urine specimens and 27 respiratory specimens were obtained from 80 patients. None of the respiratory specimens grew suspected colonies of *L. pneumophila* and all urine specimens were negative for *L. pneumophila* serogroup 1a antigen.

**Conclusion:** *L. pneumophila* serogroup 1a was not identified as the pathogen responsible for community acquired pneumonia in this study sample.

**Keywords ::** *Legionella pneumophila*, Community acquired pneumonia, culture, urinary antigen detection

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## Introduction

*L. pneumophila* causes two forms of infections, legionnaires disease and Pontiac fever. Legionnaires disease presents with pneumonia and may progress to severe disease. Pontiac fever is a flu like illness and is usually self-limiting. Legionnaires disease can occur as both sporadic and epidemic forms, though the majority of reported cases are not associated with a known epidemic. *L. pneumophila* serogroup 1 is responsible for about 90% of cases of community acquired legionnaires disease, though the relative proportions of serogroups may vary in different geographic areas.<sup>1</sup>

*L. pneumophila* is ubiquitously found in both natural and artificial aquatic environments as well as in moist soil.<sup>2,3</sup> Two studies conducted in Sri Lanka have detected *L. pneumophila* in cooling tower water of some hotels and selected hospitals.<sup>4,5</sup> It is very likely that this organism is present even in our natural aquatic water collections which provide ideal environmental conditions required for the growth of this organism.<sup>4</sup>

*L. pneumophila* causes acute consolidating pneumonia that cannot be accurately differentiated from other causes of pneumonia on clinical presentation. The mortality of legionnaires disease can be 5% to 30%, especially when treatment with proper antibiotics has been delayed.<sup>1</sup>

Routine microbiological cultures of respiratory samples are not useful in identifying *L. pneumophila* as it needs special requirements for growth. The definitive diagnosis depends on laboratory tests which include culture in special media containing cysteine, urinary antigen detection and molecular diagnostic methods.

About 50% of patients with legionnaires disease cannot produce sputum. Culture yield when sputum is available depends on the severity of illness, prior antibiotic use and the quality of the sputum sample. Sensitivity of culture ranges from  $\leq 10\%$  to 80% and specificity is 100% although detection by culture may take 3 to 7 days.<sup>5</sup> In contrast, urinary antigen detection can be performed within an hour, the sensitivity and specificity of which are 70- 90% and  $\geq 99\%$  respectively.<sup>6</sup> The limiting factor of using urinary antigen detection is that it only detects *L. pneumophila* sero-group 1. Testing of serum with PCR is not fully validated for diagnosis, but has sensitivity of 30- 50% with a specificity of  $\geq 99\%$  in research studies.<sup>6</sup>

It is important to identify the epidemiology of community acquired pneumonia to establish diagnostic and treatment protocols. *L. pneumophila* does not respond to the beta lactam antibiotics routinely used in the management of community acquired pneumonia and needs different antibiotics.<sup>7</sup> The identification of the aetiological agent is therefore very important in deciding the antibiotic as well as the duration of treatment in patients with community acquired pneumonia.

This study was planned to determine the role of *L. pneumophila* as the aetiological agent in patients with community acquired pneumonia requiring admission to Teaching Hospital, Peradeniya.

## Methods

This study was initiated in July 2014 after obtaining approval from the Ethical Review Committee, Faculty of Medicine, University of Peradeniya. Patients with a history, clinical findings and radiological findings suggestive of community acquired pneumonia, were selected as the study population. Respiratory samples (sputum or endotracheal secretion) and a urine specimen were collected into sterile, screw capped individual plastic containers. Specimens were collected within 24 hours of hospital admission and transferred with minimal delay to the microbiology laboratory. However, all the patients were found to have been started on an antibiotic at the time of specimen collection.

A loopful of respiratory specimens were inoculated onto Charcoal Yeast Extract agar (CYE) (OXOID-UK) supplemented with Legionella BCYE growth supplement. Each batch of agar plates were quality controlled with a known *L. pneumophila* strain prior to use. The inoculated plates were incubated at 35 °C-37 °C for 7 days and examined daily for presence of small grey to white, iridescent colonies.<sup>8</sup> Such colonies were picked up and inoculated onto CYE without L-Cystine. If the colony did not grow on CYE agar without L-Cysteine, the colony was considered as probable *Legionella spp.*

Any iridescent colony 3-4 mm in size with cut glass appearance were identified using Gram stain, catalase and oxidase tests. Thin, faintly staining, filamentous Gram negative bacilli which were catalase positive and oxidase negative were identified as *L. pneumophila* if positive on the hippurate hydrolysis test.

Urine specimens were stored at -20 °C in duplicate and tested batch wise with a commercial ELISA kit (GRG *Legionella pneumophila* Ag (urine) -EIA-4205,USA) for the presence of *L. pneumophila* serogroup 1a urinary antigen. The test was performed according to the test procedure recommended by the manufacturer and ELISA plate was read at 450/620-650 nm. The results were interpreted according to the cut off values and instructions provided by the manufacturer.

## Results

*L. pneumophila* was not isolated from any of the cultured respiratory specimens and none of the urine specimens tested were positive for *L. pneumophila* serogroup 1a urinary antigen.

## Discussion

*L. pneumophila* is responsible for approximately 1- 3% of community-acquired pneumonias globally. A study carried out in Germany by Baum *et. al.* (2008) using urinary antigen testing, found 3.7% of ambulatory and 3.8% of hospitalized patients with community acquired pneumonia were due to *L. pneumophila*.<sup>10</sup> Furthermore,

studies carried out in USA reported increasing incidence of legionellosis from 1990 to 2005.<sup>11</sup> A study conducted in India using serology and urinary antigen test showed that 31 patients out of 113 patients (27.43%) with community acquired pneumonia were serologically positive while 17.69% were positive for urinary antigen.<sup>12</sup>

In the current study, legionellosis was excluded as the aetiological diagnosis in 80 patients admitted with community acquired pneumonia using culture and urinary antigen detection as the means of diagnosis. However, only 27 (33.8%) of the study population were able to provide a respiratory sample for culture as they were unable to produce sputum. In addition, all the patients had received varying antibiotics prior to sample collection. Limitation of sample size was due to restricted funding for purchase of antigen detection kits. The inadequate number of samples could have been the limiting factor of this study and perhaps increasing the number of cases would have yielded a different result.

*L. pneumophila* serogroup 1 remains the main pathogen worldwide.<sup>1</sup> However, the epidemiology of legionellosis in Sri Lanka may be different and as the urinary antigen detects only serogroup 1, the presence of other serogroups cannot be excluded in the population studied. Accordingly, further studies need to be carried out to find out the epidemiology of legionella infection in Sri Lanka.

## Conclusion

*L. pneumophila* was not identified as an aetiological agent responsible for community acquired pneumonia in this study sample.

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