



Prevalence of Legionella in retirement homes and group homes water distribution systems



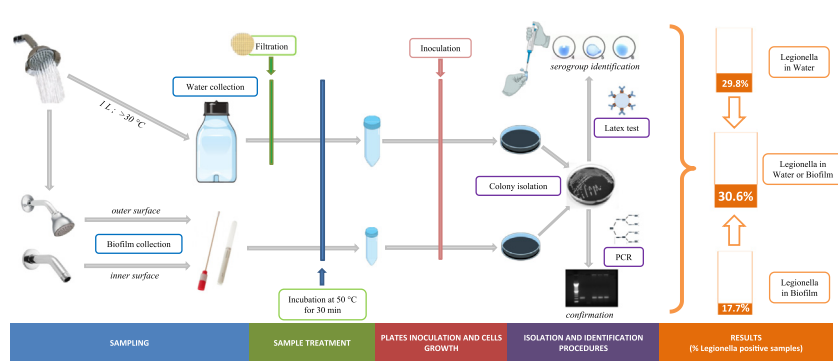
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HIGHLIGHTS

- Legionella in retirement and group homes is a problem still poorly investigated.
- Water quality and Legionella presence evaluated for estimating exposed population.
- Legionella found in 30.7% of retirement homes' and 11.1% group homes' water systems.
- Non-linear trend found correlating Legionella and Heterotrophic bacteria at 22 °C.
- Only in 1 sampling point Legionella detected in biofilm but not in water sample.

GRAPHICAL ABSTRACT



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ABSTRACT

Background: Although historically the focus has been placed above all on hospital infections and travel-associated outbreaks, most of the cases of Legionella infection are sporadic and occur in community-dwellers.

Objectives: To evaluate the presence and load of Legionella in hot water systems of non-healthcare facilities that host closed communities. Furthermore, we tried to verify the association between Heterotrophic Plate Counts (HPCs) and presence of Legionella.

Methods: We collected hot water and biofilm samples from the showerheads of retirement homes and group homes. Samples were tested by culture method for the presence of Legionella. Confirmation and identification were carried out through Latex test and PCR. We determined the HPCs at 22 and 37 °C by the pour plate method. Statistics performed through STATA.

Results: We collected 140 hot water and biofilm samples, 95 from 26 retirement homes and 35 from 9 group homes. Legionella was found in 36.8% samples collected from retirement homes and only in 10.3% group homes' samples ($p = 0.01$). Legionella was identified more frequently in water than in biofilm (29.8% vs 16.9%); just in one case the pathogen was found in the biofilm only. *L. pneumophila* sg 1 was the pathogen more frequently isolated (65.8%), with an average load of 2720 CFU/L (SD = 8393 CFU/L). We have often noticed a high microbial contamination (67% of HPCs >200 CFU/mL) and identified a higher prevalence of Legionella for intermediate values of HPC 22 °C ($p = 0.011$). 32% of people hosted in retirement homes were exposed to Legionella.

Conclusions: Colonization of water-systems of retirement homes and group homes is anything but occasional, and in our survey it mainly affects the former, moreover often due to *L. pneumophila* sg 1. The search for the pathogen in the biofilm has proved to be of little use. The relationship between HPC and Legionella deserves further studies.

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1. Introduction

Legionella pneumophila is a Gram-negative waterborne bacterium responsible for Legionnaires' disease (LD), a severe and sometimes fatal form of pneumonia (Fields et al., 2002). The case fatality rate during outbreaks is lower than the one of sporadic cases, but it can be higher for hospital-acquired infections. In fact, the main risk factors for acquiring the infection are: chronic lung disease, diabetes, various conditions associated with immunodeficiency, but also increasing age, male sex, and smoking (Phin et al., 2014).

Legionella is found in freshwater environments worldwide, but major reservoirs are man-made aquatic environments such as warm water system (Cunha et al., 2016). In these environments, the *Legionella* growth both in water and biofilm is influenced by many factors, such as aged plumbing, low flow rate or stagnation of the water, dead legs, storage tanks or surface materials, roughness, physicochemical constituents of the water, and poor management (Liu et al., 2006; Exner et al., 2005; Napoli et al., 2010; Ferranti et al., 2014; D'Alessandro et al., 2015; World Health Organization, 2017; Collins et al., 2017). Even the complexity of the water system can be a risk factor: for example, an unbalanced looped hot-water system has consequences on the temperature of circulating water causing an increased risk of legionellosis. The presence of biofilm or protozoa increases the survival and the development of *Legionella*, by protecting the bacterium from the effects of disinfection (Borella et al., 2005; Bonadonna et al., 2009; Cunliffe et al., 2011; Simoes et al., 2010; Ashbolt, 2015; Lau and Ashbolt, 2009).

The disease is mainly transmitted by inhalation of contaminated aerosols from shower heads, certain medical equipments (e.g. respiratory equipment), cooling towers, hydrotherapy equipment, decorative fountains, etc. Less commonly, the disease occurs from aspiration of *Legionella*-containing water observed in some hospital-acquired cases or even from instillation into the lung during respiratory tract manipulation (Fields et al., 2002; Cunha et al., 2016; Falkinham et al., 2015). Since *Legionella* has been isolated, it has never been proven inter-human transmission; however, a case has been recently described, even if the scientific community expects further evidence to confirm it (Correia et al., 2016; World Health Organization, 2017).

The number of LD cases in the United States reported to CDC has been on the rise since 2000. This could be due to a true increase in the frequency of disease because of a number of reasons (e.g., older population, more at-risk individuals, aging plumbing infrastructure), or otherwise to increased testing for LD. Although 6000 cases of this disease in 2015 were reported, however the true incidence may be underestimated (Centre for Disease Control and Prevention, 2018).

ECDC in 2015 reported 7034 cases of LD in Europe, 6573 (93.4%) of which were classified as confirmed. As in the previous year, the number of notifications per 100,000 inhabitants was 1.4, which remains the highest number recorded; in this context, Italy was the country with the largest number of cases in 2015 (1535), and an annual incidence of 2.6 cases per 100,000 inhabitants. Most cases (69%) were community-acquired, whereas 22% were travel-associated, and 8% linked to healthcare facilities (nosocomial and other healthcare). Healthcare-associated cases represented a substantial proportion of cases in older age groups (>60 yr) and the case-fatality rate was almost four times higher in these setting than in community-acquired cases (28% vs. 7%). This is not surprising since healthcare-associated cases are probably more likely to suffer from underlying conditions. *L. pneumophila* serogroup 1 was the most commonly identified pathogen. Of all environmental sites testing positive, 411 were water systems, 22 cooling towers and 13 pools (European Centre for Disease Prevention and Control, 2017).

The annual report regarding cases of LD diagnosed in Italy in the year 2016 shows 1710 cases notified to the National Surveillance System, of which 1680 confirmed. The number of notifications per million inhabitants was 28.2, higher than the one of the previous year (25.8). Out of these cases, 79.6% were community-acquired, whereas 10.4% were

travel-associated, 5.0% nosocomial acquired and 2.1% involved persons living in nursing homes, rehabilitation facilities or retirement homes. Case-fatality rate was 10.8% for community-acquired cases whereas 45.9% for hospital-acquired cases. In 100% of cases, the agent responsible for the disease was *L. pneumophila* (Rota et al., 2017).

Besides LD, *Legionella* is the etiologic agent of Pontiac Fever (PF). This disease, being characterized by flu-like symptoms, is often misdiagnosed, and its incidence is almost certainly underestimated, due to its benignity and lack of specificity, together with the lack of appropriate surveys and reporting definition. In fact, among 828 elderly subjects residing in nursing homes, and followed up during 4 months to ascertain incidence of symptoms associated with PF in a non-epidemic setting, 29 subjects complied with the study definition of PF, with an overall incidence density of PF of 0.11 case/person-year (95% CI 0.07 to 0.15) (Bauer et al., 2008).

Outbreaks of waterborne disease may affect large numbers of persons, and available evidence also suggests that drinking-water can contribute to background rates of disease in non-outbreak situations (World Health Organization, 2017). In fact, notwithstanding the attention generated by clusters of legionellosis, outbreaks are responsible for a small portion of total cases, approximately 4% in the United States; thus, sporadic cases are much more common (Prussin et al., 2017). In Italy too, where LD cases are on the rise, closed communities such as retirement homes are confirming over the years as a stable source of cases (Rota et al., 2017). For these reasons, control of drinking-water quality should also address waterborne disease in the community (World Health Organization, 2017).

Although LD outbreaks among hospitalized patients and residents of long-term care facilities have been reported in several reviews (Napoli et al., 2010; Ditommaso et al., 2014), few published reports have described LD outbreaks in retirement homes and other aged-care facilities (Bauer et al., 2008).

This segment of population has a high likelihood of becoming ill; in fact, according to the ISS 2017 report (2016 data), annual incidence rates of LD increase with age to >68 cases per million among persons ≥70 years old (it was 57/1000,000 in 2013), and LD-related mortality is higher among older adults compared with younger persons. In addition, certain medical conditions known to increase the risk of acquiring LD are more frequent among the elderly (Sopena et al., 2007; Cunliffe et al., 2011; Silk et al., 2013; Rota et al., 2017). Due to these facilities' characteristics, water systems can be extensive and supply water to wards and rooms that are not always occupied. Hot-water distribution systems may be maintained at lower temperatures or have thermostatic mixing valves installed to reduce the risk of scalding (Cunliffe et al., 2011). This preventive measure, together with the above mentioned water stagnation and the complexity of the pipeline network, could further facilitate the proliferation of *Legionella*.

According to CDC, 82.5% of people living in Residential Care Communities in 2013–2014 were 75 or older (Harris-Kojetin et al., 2016). In Italy an estimate of 2.3% individuals over 65 years of age lived in about 6700 retirement homes in 2010 (Montemurro, 2017).

The population aging, together with the decreasing family size and increasing employment rates among women may reduce the traditional pool of family caregivers, further stimulating demand for paid long-term care services (Harris-Kojetin et al., 2016).

The Study objective was precisely to assess the exposure to the risk of contracting legionellosis among the elderly living in retirement homes through controls in the water system of these structures. Therefore, the primary outcome of our study was to assess the prevalence of *Legionella* in the water systems of retirement homes, through collection of most of our water and biofilm samples from the distal points of the pipeline network of these structures. We have chosen to collect all the samples from the showers, given their ability to carry the infection through the dispersion of infected aerosols.

The secondary objective of this study was to assess the prevalence and the level of *Legionella* contamination in premises that host a

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