

## OCCURRENCE OF *LEGIONELLA* SPP. IN SELECTED PUBLIC FACILITIES IN POLAND IN 2009-2013 AND 2014-2016: A COMPARATIVE STUDY

### WYSTĘPOWANIE BAKTERII *LEGIONELLA* SPP. W WYBRANYCH OBIEKTACH UŻYTECZNOŚCI PUBLICZNEJ W POLSCE W LATACH 2009-2013 ORAZ 2014-2016: ANALIZA PORÓWNAWCZA

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A. Study design/planning  
zaplanowanie badań  
B. Data collection/entry  
zebranie danych  
C. Data analysis/statistics  
dane – analiza i statystyki  
D. Data interpretation  
interpretacja danych  
E. Preparation of manuscript  
przygotowanie artykułu  
F. Literature analysis/search  
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#### Summary

**Background.** This paper evaluated *Legionella* spp. water system contamination in the public facilities.

**Material and methods.** Samples of warm water from the inner water supply system of 41 facilities in years 2009-2013 and 46 facilities in years 2014-2016 were collected for analyses. The facilities included school dormitories, boarding houses, school-education centers, halls of residence, nursery schools, kindergartens, and correctional institutions.

**Results.** The results of research carried out on tap water in public facilities in 2014-2016 showed that in comparison to 2009-2013 the percentage of facilities from group I increased. In group III facilities a decreasing tendency was observed in comparison to research carried out in 2009-2013.

**Conclusions.** The results indicate that it cannot be explicitly ascertained that the eradication methods of *L. pneumophila* from tap water systems of the analyzed facilities within the studied period were sufficient. Epidemiological analyses of this type should be successively and systematically continued and based of the largest possible number of facilities.

**Keywords:** *Legionella pneumophila*, Legionnaire disease, environmental surveillance

#### Streszczenie

**Wprowadzenie.** Celem pracy była ocena skażenia systemów wodnych w budynkach użyteczności publicznej przez bakterie *Legionella* spp.

**Materiał i metody.** Materiałem były próbki wody ciepłej pobrane z wewnętrznej instalacji wodociągowej 41 obiektów w latach 2009-2013 oraz 46 obiektów w latach 2014-2016. Były to: bursy szkolne, internaty, ośrodki szkolno-wychowawcze, domy studenta, żłobki, przedszkola, zakłady karne.

**Wyniki.** Wyniki badań sieci wody użytkowej obiektów użyteczności publicznej w latach 2014-2016 w porównaniu do lat 2009-2013, wykazały, że odsetek obiektów z grupy I wzrósł. W obiektach z grupy III stwierdzono tendencję spadkową w porównaniu do lat 2009-2013.

**Wnioski.** Wyniki badań świadczą, że w analizowanym okresie nie można jednoznacznie stwierdzić czy stosowane metody eradykacji *L. pneumophila* z systemów wody użytkowej analizowanych obiektów są wystarczające. Tego typu analizy epidemiologiczne należałoby sukcesywnie i systematycznie kontynuować w oparciu o jak największą liczbę obiektów.

**Słowa kluczowe:** *Legionella pneumophila*, choroba legionistów, nadzór środowiskowy

#### Introduction

The natural environment of *Legionella* type bacteria is water [1]. These bacteria appear in natural and artificial water systems, groundwater, littoral zones of seawater, thermal water, moist soil, and clay soil as well as near wastewater discharge. The optimal temperature for the development of the bacteria ranges between 20°C and 50°C [2]. In the natural environment, they constitute less than 1% of the general population of bacteria and they do not pose an epidemiological threat [3].

Over 50 species and 72 serological groups of *Legionella* type bacteria were determined. Out of them at least 21 species may cause diseases in humans [4]. The majority of infections are caused by the *Legionella pneumophila* species (around 80-90%). *L. pneumophila* causes 75-90% of diseases and belongs to the first serological group

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(sg 1) [5]. Other species causing Legionnaires' disease are *L. longbeachae*, *L. micdadei*, *L. bozemanii*, *L. feeleii*, *L. anisa*, and others [4]. The infection occurs most often through the inhalation of water-air aerosol with drop diameter ranging from 2.0 to 5.0  $\mu\text{m}$  containing invasive forms of *Legionella* bacteria [6]. To date, three clinical forms of Legionnaires' disease have been characterized:

- legionellosis pneumonia (Legionnaires' disease, pulmonary form),
- Pontiac fever (Lochgoilhead fever),
- extrapulmonary form [5].

The severity of legionellosis pneumonia depends on coexisting diseases [5]. After the period of incubation (2-10 days) the following symptoms occur: cough of varying intensity, malaise, myalgia, fever up to 40°C and shivering [4]. Neurological symptoms (ataxia, headaches, confusion) and digestive disturbances (diarrhea, abdominal pain, nausea, vomiting) may also appear. Mortality in the case of the pulmonary form amounts at 15-20% [7].

Pontiac fever is characterized by flu-like symptoms which include: fever, headaches, myalgia, and arthralgia. Diarrhea and vomiting are rare. Next, sore throat, cough, and rhinitis occur. The incubation period lasts from 30 to 120 hours, and most frequently is 36 hours long. The incidence rate amounts to around 80-95% [4, 7]. The disease starts rapidly and the majority of cases do not require hospitalization [8].

The extrapulmonary form of Legionnaires' disease is rarely reported. In the course of the disease, apart from pneumonia, the following symptoms indicating *Legionella* bacillus infection of other organs occur: acute circulatory failure, sepsis syndrome, coagulation disorders and nephritis [4, 8].

In Poland, infectious diseases including legionellosis are reported based on the Act of prevention and combating infections and infectious diseases in humans. The entire population is sensitive to *Legionella* bacterial infections [9]. Smokers, people with immunodeficiency, people with coexisting diseases (diabetes, cancer, chronic respiratory diseases) belong to groups of patients with increased risk. Male gender and those over the age of 50 are risk factors [10]. People who have contact with tap water especially in the form of water-air aerosol experience risk of occupational exposure to *Legionella* bacteria [11].

Pontiac fever and Legionnaires' disease should be registered in Poland in accordance with the International Statistical Classification of Diseases and Medical Procedures introduced by WHO. In 2009-2013, 83 cases of legionellosis and 2 cases of Pontiac fever were reported while in 2014-2016 there were 59 cases of legionellosis and 5 cases of Pontiac fever [12-17].

According to reports published by the European Centre for Disease Prevention and Control in the corresponding periods, 22,897 cases of legionellosis were reported in 2009-2013 and 21,079 cases in 2014-2016 [18, 19].

The aim of this paper was to evaluate contamination of water supply networks in public facilities like dormitories, boarding houses, school-education centers, halls of residence with *Legionella* spp. bacillus as well as the analysis of eradication dynamics of *Legionella* spp. bacillus. The comparative analyses included chosen public facilities in Poland in the years 2009-2013 and 2014-2016.

## Material and methods

Warm water samples for testing for *Legionella* spp. were collected by local staff members of 16 Voivodship Sanitary and Epidemiological Stations in Poland as a part of water quality monitoring. Water samples were collected in the period from January 2009 to December 2013 from water supply network of 41 facilities including nursery schools (7), kindergartens (6), penitentiary institutions (6), night shelters (5), halls of residence (6), orphanages (5) and boarding houses (6). In the period from January 2014 to December 2016 samples were collected from water supply networks of 46 public facilities such as nursery schools (9), kindergartens (9), penitentiary institutions (4), night shelters (10), halls of residence (4), orphanages (6) and boarding houses (4).

Warm water samples collected from the water supply network were tested in the laboratories of the Sanitary Inspections according to the norms and legal acts in force during the time of research:

1. PN-EN ISO 11731-2:2008 "Water quality – detection and quantitative marking of *Legionella* bacteria – Part 2: The methods of membrane filtration for water with a small number of *Legionella* bacteria",
2. PN-ISO11731, December 2002 "Water quality. Detection and marking of *Legionella* bacteria",
3. PN-EN ISO19458:2007 "Water quality. Sample collection for microbiological analyses",
4. Decree of the Minister for Health of March 29, 2007, concerning the quality of water intended for human consumption (Journal Of Law, no. 61, item 417 as amended),
5. Decree of the Minister for Health of November 13, 2015, concerning the quality of water intended for human consumption (Journal of Laws, 2015, item 1989).

Test results from two periods of time which were obtained from sanitary-epidemiological stations underwent statistical analysis. The statistical analysis was carried out in the STATISTICA 7.1 software. Pearson's Chi-square independence test was conducted with the relevance level  $p \leq 0.05$ .

#### The evaluation threat posed by *Legionella* spp. bacteria in the studied facilities

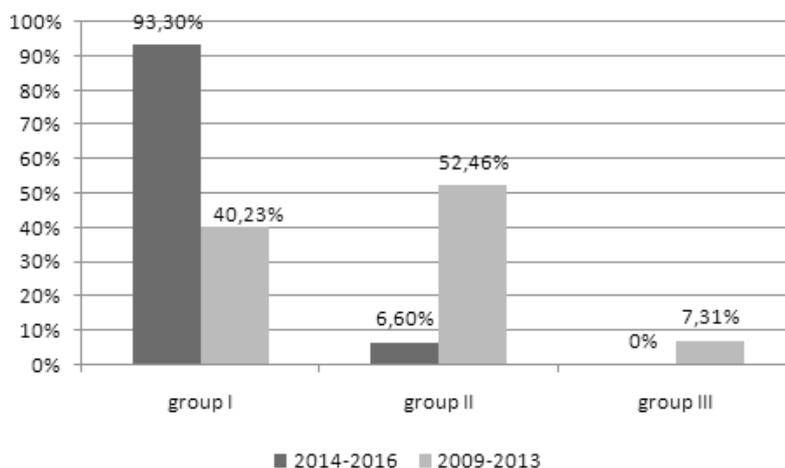
The following arbitrary division of results was assumed [20]:

- group I – negative samples – samples in which  $<100$  CFU/100 ml was found,
- group II – samples in which in the same test throughout the studied period, positive (value  $>100$  CFU/100 ml) and negative ( $<100$  CFU/100 ml) were obtained alternately,
- group III – positive samples – samples in which  $>100$  CFU/100 ml was reported (according to the Decree of the Minister for Health concerning the quality of water destined for human consumption valid for the period of studies).

The epidemiological analysis concerned solely the results of laboratory tests and did not include issues connected with *Legionella* spp. eradication.

## Results

Research results showing the division into groups of facilities studied between 2009-2013 and comparatively in 2014-2016 where positive, negative as well as positive and negative samples containing *Legionella* spp. bacillus was detected in warm water supply system are presented in Figure 1 and Table 1. Results of tap water supply systems in public facilities in years 2009-2013 in comparison to years 2014-2016 show that the proportion of facilities qualified to group I increased from 40.23% in years 2009-2013 to 93.13% in 2014-2016. The proportion of facilities qualified to group II in the analyzed years shows a decreasing tendency from 52.46% in 2009-2013 to 6.60% in 2014-2016. In turn, the facilities qualified to group III in the analyzed years show a decreasing tendency from 7.31% in 2009-2013 to 0% in 2014-2016 (Figure 1). These data indicate that it cannot be explicitly ascertained that eradication methods of *L. pneumophila* from tap water systems of the analyzed facilities within the studied period are sufficient.



**Figure 1.** Evaluation threat posed by *Legionella* spp. bacteria in studied public facilities in years 2009-2013 in comparison to years 2014-2016

The mean number of colonies of *Legionella* spp. in the water samples studied in 2014-2016 was distinctly higher in comparison to 2009-2013, however the difference was not statistically significant (Table 1).

**Table 1.** The number of *Legionella* spp. colonies in years 2009-2013 and 2014-2016

no.	Year of sample collection	Number of facilities	Mean number of colonies $\bar{x}$	Standard deviation $\pm$ SD	Standard error SE
1	2009-2013	41	223.33	589.74	60.51
2	2014-2016	46	485.00	1518.09	216.87

Notes: 1-2:  $Z=1.52$ ;  $p=0.1274$

Relations between the years 2009-2013 and 2014-2016.

## Discussion

The obligation of supervision over the quality of water destined for human consumption in public facilities in which smaller or larger groups of people reside is enforced by a Decree of the Minister for Health. On January 11, 2018, an amended Decree of the Minister for Health of December 7, 2017, concerning the quality of water intended for human consumption became effective (Journal of Laws, 2017, item 2294). The changes pertain among others the obligation to conduct tests of warm water for the presence of *Legionella* spp. bacteria. According to the current regulations, such tests should also be carried out in public facilities where water-air aerosol is produced during utilization [21].

The analyzed group of public facilities such as dormitories, boarding houses, school-education centers, halls of residence, nursery schools, kindergartens, penitentiary institutions has been underfunded for many years. Lack of renovations including water supply systems contributed to the colonization of *Legionella* bacillus. The analyses of research carried out in two periods show that the epidemiological situation is improving but not to a satisfactory extent.

Research conducted in 111 public facilities in the Małopolska region in 2016 shows the colonization of *Legionella* bacillus in 43.2% of analyzed facilities such as primary schools, secondary schools, post-secondary schools, dormitories, boarding houses, halls of residence and school-education centers [21]. Research carried out in the years 2009-2013 shows a very high level of contamination with *Legionella* bacteria in the water supply system of buildings. It is connected with the underfunding of such institutions [7]. The results obtained in the comparative period may be a positive trend. Similar research was conducted by Italian authors in orphanages and retirement homes demonstrating the colonization by *Legionella* bacillus in 36.8% of the collected samples [22].

In the available databases, there were not enough published papers concerning the evaluation of threat in dormitories, boarding houses, school-education centers, and halls of residence to compare obtained results with the results by other authors.

## Conclusions

1. The results of the research indicate that it cannot be explicitly ascertained that eradication methods of *L. pneumophila* from tap water systems of the analyzed facilities within the studied period are sufficient.
2. Epidemiological analyses of this type should be successively and systematically continued on the bases of the largest possible number of facilities.

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