Thermal disinfection of hotels, hospitals, and athletic venues hot water distribution systems contaminated by *Legionella* species

Varvara Mouchtouri, BSc,[a] Emmanuel Velonakis, MD, PhD,[b] and Christos Hadjichristodoulou, MD, PhD[a]
Larissa and Athens, Greece

Legionella spp. (≥500 cfu liter^{-1}) were detected in 92 of 497 water distribution systems (WDS) examined. Thermal disinfection was applied at 33 WDS. After the first and second application of the disinfection procedure, 15 (45.4%) and 3 (9%) positive for remedial actions WDS were found, respectively. *Legionella pneumophila* was more resistant to thermal disinfection than *Legionella* non-*pneumophila* spp. (relative risk [RR] = 5.4, 95% confidence intervals [CI] = 1–35). WDS of hotels with oil heater were more easily disinfected than those with electrical or solar heater (RR = 0.4 95% CI = 0.2–0.8). Thermal disinfection seems not to be efficient enough to eliminate legionellae, unless repeatedly applied and in combination with extended heat flushing, and faucets chlorine disinfection. (Am J Infect Control 2007;35:623-7.)

Hot water systems are the most frequently involved source of infection in many Legionnaires' disease outbreaks,[1] while cases of *Legionella* infection have been linked to exposure to water systems of hotels[2] and hospitals.[1] Many different control measures have been recommended to control the growth of the organism in water systems including thermal disinfection.[3]

Water temperatures more than 60°C are considered inhibitory for *Legionella* spp. Laboratory studies have shown that the time required to reduce a population of *Legionella* bacteria by one log at 45°C, 50°C, 60°C, and 70°C, was 2500, 380, less than 5, and less than 1 minute, respectively.[4] Increasing the temperature of hot water was the first method used to control *Legionella* in hospital hot water distribution systems (WDS).[5]

The aim of this study was to assess the effectiveness and to specify the advantages and disadvantages of the thermal disinfection at *Legionella* positive WDS of hotels, hospitals, and athletic venues. The water distribution system is defined as a system of conduits (laterals, distributaries, pipes, and their appurtenances), from the point where the water enters the building to the extraction points, by which water is distributed to consumers.

From the Department of Hygiene and Epidemiology,[a] Faculty of Medicine, University of Thessaly, Larissa; and the Department of Microbiology,[b] National School of Public Health, Athens, Greece.

Address correspondence to Christos Hadjichristodoulou, Department of Hygiene and Epidemiology, Faculty of Medicine, University of Thessaly, 22 Papakiriazi Str., Larissa, 41222, Greece. E-mail: xhatzi@med.uth.gr.

0196-6553/$32.00

doi:10.1016/j.ajic.2007.01.002

METHODS

The study was part of the Environmental Health Surveillance Program implemented for the Athens 2004 Olympic games. Methods used for sample collection, storage conditions, and microbiological analysis have been described elsewhere.[6] The methodology and criteria used to choose the premises sampled have been published and discussed elsewhere.[7,8]

The environmental health officers of the Prefecture Public Health Departments of seven Greek towns conducted the sampling. The National Legionella Reference Laboratory of Southern Greece in Athens, and the National Legionella Reference Laboratory of Northern Greece in Thessalonica, performed the microbiologic analysis.

The detection limit of legionellae was ≥ 500 cfu liter^{-1}. *Legionella* positive for remedial actions WDS were characterized according to the European Working Group for *Legionella* Infection guidelines:[9] when the concentration of *Legionella* bacteria was ≥1000 cfu liter^{-1} in more than two samples, or ≥10,000 cfu liter^{-1} in at least one sample.

Thermal disinfection was carried out by raising the temperature of the hot water storage tank content to 70–80°C and then circulating this water throughout the system for up to 3 days. The temperature at the hot water storage tank was raised high enough to ensure that the temperatures at the taps and appliances would not have fallen below 65°C. Each tap and appliance was run sequentially for at least five minutes at

Table 1. Summary of sampling sites and *Legionella*-positive results

Water distribution systems (WDS)/samples	Facilities and samples tested	No of WDS or No of samples/total (%)		
		Hotels	Hospitals	Athletic venues
No of WDS				
	Facilities tested*	385	18	94
	Positive for *Legionella*†	80/385 (20.8)	8/18 (44.4)	4/94 (4.2)
	Positive for remedial action‡	35/385 (9.0)	4/18 (22.2)	2/94 (2.1)
	Remaining positive§ after 1st hot water flush	14/27 (51.8)	1/18 (5.5)	0/94 (0.0)
	Remaining positive§ after 2nd hot water flush	3/27 (11.1)	0/18 (0.0)	0/94 (0.0)
No of samples				
	Collected from WDS and tested	1086	75	382
	Positive for *Legionella**	268/1086 (24.7)	31/75 (41.3)	15/382 (3.9)
	Remaining positive§ after 1st hot water flush	25/293 (8.5)	3/83 (3.6)	0/30 (0.0)
	Remaining positive§ after 2nd hot water flush	7/30 (23.3)	0/0	0/0

*6-20 samples tested per WDS.
†Concentration of *Legionella* bacteria $\geq$ 500 cfu liter^{-1} in at least one sample collected from the WDS.
‡Concentration of *Legionella* bacteria $\geq$ 1000 cfu liter^{-1} in more than two samples, or $\geq$ 10,000 cfu liter^{-1} in at least one sample.
§Concentration of *Legionella* bacteria $\geq$ 1000 cfu liter^{-1} in at least one sample collected from the WDS.

the full temperature, and this was measured.[9] Two to seven days after the disinfection, 6 to 20 samples were collected from each WDS that was resampled. Disinfection of the system was repeated until legionellae concentration remained less than 1000 cfu liter^{-1} at all samples.[9] Disinfection was considered successful when concentration at all samples was less than 1000 cfu liter^{-1}. To prevent scalding by the users when the temperatures of the hot water storage tanks were raised, tables were displayed to warn about the danger of hot water.

The standardized disinfection guidelines were printed and distributed to the persons responsible for the buildings by the Prefecture Departments of Public Health personnel. Hoteliers or the hospital staff applied the disinfection procedure, and when possible supervised by the Prefecture Departments of Public Health personnel.

Data were analyzed with Epi-Info 2000 (CDC, Atlanta, GA) and SPSS for Windows Release 11.0.1 software (SPSS Inc, Chicago, IL). The Mann-Whitney test was used for quantitative data and the chi-square test or Fisher's exact test for qualitative data. Results were considered statistically significant when the P value was $< .05$.

RESULTS

Out of the total 385 hotel WDS examined, 80 (20.8%) were colonized by *Legionella* spp. ($\geq$500 cfu liter^{-1}), 268 samples were positive of the 1086 samples examined, and 35 hotel WDS were needed remedial actions. In our study, of the total 35 hotels, only 27 were included. Furthermore, out of the total 18 hospital WDS examined, 8 (44.4%) were colonized by *Legionella* spp., 31 samples were found positive ($\geq$500 cfu liter^{-1}) of the total 75 examined, and four hospitals (22.2%) needed remedial actions. Of the 94 athletic venues hot WDS examined, four (4.2%) were positive ($\geq$500 cfu liter^{-1}), 15 positive samples were found of the total 382 examined, and two athletic venues hot WDS (2.1%) needed remedial actions (Table 1).

After the first application of the disinfection procedure, 293 samples were collected from the 27 hotels hot WDS needing remedial actions, while 83 samples were collected from the 4 hospitals, and 30 from the 2 athletic venues.

After the first thermal disinfection application, in 14 (51.8%) of the 27 hotel WDS resampled, at least one sample was found positive for *Legionella* spp. with concentrations of $\geq$ 1000 cfu liter^{-1}. Disinfection was considered successful when concentration at all samples was less than 1000 cfu liter^{-1}. After the second application of the disinfection procedure, in 3 of the 27 hotels (11.1%), at least 1 sample was found with *Legionella* spp. concentration of $\geq$ 1000 cfu liter^{-1}. In those 3 WDS, dismantlement, cleaning, chlorine disinfection, and scale removal of the shower heads and hoses was applied, and the following sampling in those hotels showed that *Legionella* spp. were $<$ 1000 cfu liter^{-1} in all the samples examined (Table 2).

On the contrary, after the first application of the disinfection procedure, *Legionella* spp. was eradicated below 1000 cfu liter^{-1} in all athletic venues positive for remedial actions WDS (Table 2).

After first disinfection, of the total four positive for remedial actions hospital WDS, one was needed second disinfection, while *Legionella* spp. were eradicated below 1000 cfu liter^{-1} in all samples examined, after second disinfection (Table 2).

No sample collected from hotels, hospitals, or athletic venues contained $\geq 10^4$ cfu liter^{-1} *Legionella* spp. after the second disinfection procedure.

Legionella pneumophila was more resistant to thermal disinfection than *Legionella* non-*pneumophila*

Table 2. *Legionella* spp. contamination of hotels, hospitals, and athletic venues hot water distribution systems after heat flushing disinfection

	No. of positive water distribution systems*					
Type of building	**10^3-10^4 cfu/l/total**	**$\geq 10^4$ cfu/l/total**	***Legionella* spp.**	***L. pneumophila* serogroup 1**	***L. pneumophila* serogroups 2-14**	***Legionella* non *pneumophila***
Hotels						
First sampling	26	17	27	15	10	7
Sampling after first disinfection	14	4	14	9	5	—
Sampling after second disinfection	3	—	3	2	1	—
Hospitals						
First sampling	3	2	4	3	3	—
Sampling after first disinfection	1	—	1	1	—	—
Sampling after second disinfection	—	—	—	—	—	—
Athletic venues						
First sampling	2	—	2	1	1	—
Sampling after first disinfection	—	—	—	—	—	—
Sampling after second disinfection	—	—	—	—	—	—

*Concentration of *Legionella* bacteria $\geq$ 1000 cfu liter^{-1} in more than two samples, or $\geq$ 10,000 cfu liter^{-1} in at least one sample.

spp. (RR = 5.4, 95% CI = 1–35). No significant difference was found to the resistance of *L. pneumophila* serogroup 1 and *L. pneumophila* serogroups 2 to 14.

To investigate whether other factors influenced the result of the disinfection procedure of hotels WDS, we examined the following confounders: WDS material, types of faucets, WDS age, hotel age, the presence of independent disinfection system, seasonal operation, presence of water tanker, type of building (bungalows or flats), and the heater type. Statistically significant association was found in hotels with oil heater (RR = 0.4, 95% CI = 0.2–0.8), which were more easily disinfected than those with solar or electrical heaters.

DISCUSSION

This study shows that thermal disinfection is not totally effective in eradicating *Legionella*, as suggested by previous studies as well.[10,11,12] Although hot water flush has been shown more effective than other strategies, its efficacy is neither complete nor sustained.[13] In our study, the majority of WDS needed two applications of the disinfection procedure, in order to achieve concentration of *Legionella* spp. below the limit of 1000 cfu liter^{-1}.[9] Recent data show *Legionella* thrives as body temperature and up to 42°C, but growth is inhibited beyond this.[14] This study confirms that hot water flush achieved temperatures at the faucet in excess of 65°C, a temperature more than adequate to destroy *Legionella*. However, *Legionella* was not totally eradicated according to our study results, suggesting protective mechanisms as operative in facilitating recolonization of the plumbing system. These protective mechanisms may have been provided by the repository of biofilm, which renders *Legionella* and other organisms refractory to systemic disinfection strategies employed. Biofilm cannot be totally destroyed and can rapidly repopulate serving as a repository for the sustained release of microbes.[15] Moreover, recently developed methods using peptide nucleic acids for *Legionella* detection, show 90% more viable *Legionella* in biofilm than is cultureable.[16] Our findings suggest that biofilm may limit the effectiveness of any intermittent systemic disinfection regimen and provide impetus for more creative solution to *Legionella* colonization of water supply lines.

In addition, another possible explanation of the disinfection procedure failure is that in large buildings such as hotels and hospitals, the temperature may not reach as high levels as needed at all points of the systems. The disinfection procedure was not possible to be supervised in all cases. This prevented us from monitoring the temperature raised and the duration of the flushing in all applications of the disinfection procedure, so variations cannot be excluded. It is possible that in practice, hot water reservoirs in large buildings might not permit a few minutes flushing without a drop in water temperature at peripheral sites. The only confounder that might influence the study results is the type of heater. Oil heaters could probably maintain higher temperatures than the electrical or solar heaters during the disinfection of hotels WDS.

Faucets and valves of a water system can be highly colonized by legionellae.[17] In taps and showers, the temperature of water falls because of the stagnation of water, thus permitting the survival and growth of *Legionella* spp. Moreover, distal points of the installation sediment may harbor *Legionella* spp. eventually protected from high temperatures, which can recolonize

the system from as little as a few weeks after treatment, particularly if it has not been accompanied by other remedial measures. The thermal treatment will not disinfect downstream of thermostatic mixer valves and so is of limited value where such valves are installed.[9] In our study, the recommended 5-minute flushing might has not been enough to eradicate *Legionella* spp. from faucets of hotels and hospitals.

Since *Legionella* spp. were eradicated below the detection limit after the application of dismantlement, cleaning, and scale removal of the shower heads and hoses, we recommend disinfection, cleaning, and frequent replacement of outlets in WDS. Moreover, when feasible, we believe that to improve the efficacy of the method, the duration of the heat flush disinfection should be extended from the recommended 5 minutes up to 10 minutes, or even more, as suggested by other researchers.[18]

Legionella non-*pneumophila* spp. was less resistant to thermal disinfection (RR = 5.4, CI = 1–35). No significant difference was found to the resistance of *L. pneumophila* serogroup 1 and *L. pneumophila* serogroups 2 to 14. In one of our previous reports, it was indicated that *L. pneumophila* serogroups 2 to 14 are more resistant to high temperatures (V. Mouchtouri, unpublished), whereas the findings of an Italian study indicate that *L. pneumophila* serogroup 1 is more resistant to higher temperatures.[19]

Hospitals were more colonized than hotels and athletic venues. These differences in *Legionella* spp. colonization prevalence for the three types of buildings could be attributed to the fact that hospitals were generally larger and older than hotels and athletic venues. In addition, most athletic venues were newly constructed. Furthermore, hoteliers were more aware in Legionnaire's disease prevention and applied *Legionella* control programs. On the contrary, most hospitals were not applying disinfection programs regularly.

Our data suggest that thermal disinfection seemed to be less effective in hotels than in hospitals and athletic venues. Practical difficulties in the application of the disinfection procedure in hotels, such as the inability to check the flush duration and the temperature in occupied hotels rooms, and the lack of available hotel personnel could have contributed to the inefficiency of the disinfection procedure in comparison with the disinfection results in hospitals WDS. In hospitals, the rooms could be accessed any time and there were more personnel available to contribute to the application of the disinfection procedure, in comparison with hotels.

The main disadvantage of the heat flush method was the inability to check flushing times and temperatures at hotels operating all year round and having continual occupancy due to the presence of clients. Moreover, numerous personnel were involved to monitor distal sites, water tank temperatures, and flushing times.

Our findings suggest that thermal disinfection seems not to be efficient enough to eliminate legionellae concentrations, unless repeatedly applied and in combination with extended heat flushing and faucets chlorine disinfection. Many studies have demonstrated that heat flushing was often inefficient, and rapid recolonization was obtained after the treatment.[20] This may be reasonable, taking into account that thermal disinfection might be followed by constant maintenance of the temperature of the hot water system between 55–60°C.[9] Unfortunately we could not document the possible recolonization of the WDS by further sampling of the same sites in each building.

Since eradication of *Legionella* spp. from colonized distribution systems is difficult to achieve and remedial action might be temporarily efficient, we believe that the application of regular measures to prevent water system colonization would be useful to be included in an effective *Legionella* control program. Such measures could include temperature control, chemical treatment, filtration, etc. Alternative methods to thermal disinfection could include the recent advent of point-of-use water filtration. Such filters have been tested for their efficacy in reducing or eliminating the passage of *Legionella* and other microbes that may cause disease.[21,22]

This work was supported by the Hellenic Ministry of Health. We extend thanks to all environmental health inspectors of Olympic cities and the staff of the National School of Public Health. We also thank Professor S. D. Alexiou, Professor A. Mavridou, Dr. Vasilios D. Daniilides, Mrs. L. Georgiadou, and Mr. George Panagako.

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