Effectiveness of different methods to control legionella in the water supply: ten-year experience in an Italian university hospital

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Summary

We report our ten-year experience of hyperchlorination, thermal shock, chlorine dioxide, monochloramine, boilers and point-of-use filters for controlling legionella contamination in a hospital hot water distribution system. Shock disinfections were associated with a return to pre-treatment contamination levels within one or two months. We found that chlorine dioxide successfully maintained levels at <100 cfu/L, whilst preliminary experiments gave satisfactory results with monochloramine. No contamination was observed applying point-of-use filters and electric boilers at temperatures of >58 °C and no cases of nosocomial legionellosis were detected in the ten-year observation period. Our performance ranking in reducing legionella contamination was filter, boiler, chlorine dioxide, hyperchlorination and thermal shock. Chlorine dioxide was the least expensive procedure followed by thermal shock, hyperchlorination, boiler and filter. We suggest adopting chlorine dioxide and electric boilers in parallel.

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Introduction

Contamination of hot water distribution systems is the most important risk factor for nosocomial legionellosis.1,2 National and international guidelines aimed at controlling and preventing legionella infections advocate routine use of biocides for treatment of hot water.3,4 The optimum method has not been established since each of the various options has advantages and disadvantages. Methods used include superheating, ultraviolet light, copper–silver ionisation, ozone, hyperchlorination, chlorine dioxide, point-of-use water filters, and monochloramine.5–14 Their effectiveness has been described in recent reviews and original papers, but long term investigations in health facilities are lacking.9,10,15,16 Here we report our ten-year experience in an Italian hospital whose hot water distribution system is contaminated by Legionella pneumophila. The objective was, as far as possible, to compare the effectiveness of different methods. A contribution to the evaluation of colonisation risk in new water distribution systems and a cost-benefit analysis are also reported.

Methods

University Hospital, Modena, is a 765-bedded facility consisting of a nine-storey block (with three water networks A, B, C) and a separate building (D) constructed in the 1970s; plus three other buildings (E, F, G) built in the 1990s for specific activities (operating theatres, infectious disease and oncology). Incoming cold groundwater, disinfected with chlorine dioxide, is provided by the municipality. Hot water is produced on site using heat exchangers and is stored in stainless steel tanks with return loops.

In 2000, in response to the publication of national guidelines, a programme was implemented to assess legionella contamination in the hospital’s water distribution systems.5 These guidelines do not offer advice on the frequency or number of sites to be sampled in non-epidemic situations, hence the sampling strategy took account of hospital and patient characteristics in order to include high and medium risk wards. The protocol scheduled sampling at least one remote point every 50 beds, possibly reiterating the same sites and testing other points when high risk patients were hospitalised.17

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Italian guidelines advocate no intervention when levels of *Legionella* spp. are <100 cfu/L, clinical surveillance when levels exceed this threshold but are <10^4 cfu/L, and adoption of disinfection measures at levels >10^4 cfu/L. High levels detected in this hospital have led to the implementation of a range of control strategies over several years:

- Shock hyperchlorination (sodium hypochlorite, 20–50 ppm of free chlorine at distal points for 1–2 h) was performed twelve times over seven years (2001–2008) in all hospital blocks except buildings F and G; each intervention required considerable manpower (ten workers for one night).
- Superheating (two days >60 °C at distal points) was carried out eight times in four years (2005–2008) in the same blocks as hyperchlorination, requiring two workers to flush every outlet for at least 5 min.
- Electric boilers (total 53 units, 50 L capacity) were installed on the cold water line where transplant, oncology and other high risk patients were hospitalised in order to produce hot water at the point of use without exposure to the contaminated hot ring main water. Boilers have to be replaced every five years due to hardness of the groundwater supply.
- Point-of-use water filters were installed in rooms used for high risk patients. These were replaced every 30 days according to the manufacturer’s specifications.
- Two continuous chlorine dioxide systems were installed in 2005 in hot water plants A and B, assuring 0.3 ppm at distal outlets. The level has been occasionally increased to 1 ppm in response to increased bacterial load.
- Experimental equipment continuously injecting monochloramine at 3 ppm has been in place since March 2009 in plant D.
- In addition, monthly inspection, cleaning and maintenance of water distribution systems, decalcification and/or replacement of showers/taps are provided after each treatment.

**Sample collection and analysis**

Hot water samples (N = 432) were collected from storage tanks, return loops and distal outlets (showers or taps), without flaming and after flushing for 1 min, measuring water temperature and chlorine levels (free chlorine, chlorine dioxide and/or monochloramine; DPD method, Microquant, Merck, Darmstadt, Germany). Sample transport and laboratory processing are described elsewhere.\(^{18,19}\) Control samples (N = 67) were collected before treatment and at least one year after any treatment. Post-treatment samples were collected at regular intervals until six months after superheating (N = 47) and hyperchlorination (N = 75), and annually for boilers (N = 57) and chlorine dioxide (N = 80). Twenty samples were collected following monochloramine installation and 16 from endpoints with filters. Only viable planktonic bacteria were enumerated. Forty-seven cold water samples were collected, both from inlets and distal taps, but these were all negative and are excluded from the subsequent analysis.

**Active clinical surveillance**

Since 2000, active surveillance has been maintained with the aim of detecting both community-acquired and nosocomial cases of * legionella pneumonia*. The inclusion criterion was pneumonia diagnosed by clinical and/or radiological tests without aetiological identification. *Legionella* urinary antigen (Biotest EIA kit, Dreieich, Germany) was systematically performed, followed by culture of sputum on MWY agar (Oxoid Ltd, Basingstoke, UK) and testing for serum anti-*Legionella* antibodies (IFA, RIDA® Fluor Legionella IgG, R-Biopharm AG, Darmstadt, Germany), requiring a titre of 1:256 or seroconversion for confirmation.

**Data analysis**

Statistical calculations were performed using SPSS/pc (SPSS Inc, Chicago, IL). Logarithmic transformations were used to normalise the bacteriological data. Results are presented as geometric mean values. The results were analysed by χ²-test, one-way analysis of variance and correlation analysis.

**Results**

Buildings F and G were never contaminated (N = 70 samples) and are excluded from the analysis. The other buildings have been heavily contaminated by *L. pneumophila*; in 2000, before any treatment, 14/16 samples (87.5%) were positive with a median concentration of 1.5 × 10^4 cfu/L (range 1.2 × 10^7–9.5 × 10^3 cfu/L). Seven of these 14 exceeded 10^4 cfu/L. Since then, and following treatment, the proportion of positive samples has not substantially changed with time: 15/28 (53.6%) in 2000–2001, 69/83 (83.1%) in 2002–2004, 85/130 (65.4%) in 2005–2007 and 69/105 (65.7%) in 2008–2009. However, the proportion of heavily contaminated points (bacterial loads >10^4 cfu/L) has significantly reduced to 47.7%, 43.4%, 20.8% and 31.4%, respectively (P < 0.001).

In all (pre- and post-treatment), *L. pneumophila* was isolated from 252 out of 362 water samples (69.6%). The most representative serogroups were 9 and 6, isolated alone (N = 81 and 22 respectively), in combination (N = 26) and in association with other serogroups (N = 30). Serogroups 3–4–10 were sporadically identified (N = 20), and 54 isolates were defined as 2–14. Serogroup 1 was isolated alone in 19 samples and in combination with other serogroups in 32 samples. No differences were observed according to floor, sampling point (shower, tap, return loop or tank) or season. Our experience of control measures was as follows:

- Superheating was associated with a non-significant reduction in contamination within the first month (2500 vs 8100 cfu/L) after which values returned to baseline (7000 cfu/L).
- Hyperchlorination (Figure 1A) was initially effective but levels returned to (or exceeded) pre-treatment levels after two months.
- Chlorine dioxide (Figure 1B) maintained *Legionella* contamination at low levels during a three-year observation period. Chlorine dioxide was strongly and negatively related to *Legionella* concentration (r = −0.70, P < 0.01). The regression line parameters (y = 2.55–1.98x) suggest that 0.3 ppm chlorine dioxide is associated with <100 cfu/L, and 0.6 ppm with <25 cfu/L (detection limit). Only 43 out of 80 samples (53.7%) were positive following installation of chlorine dioxide decontamination compared to 65/67 (97.0%) of pre-treatment samples (P < 0.001), although *L. pneumophila* serogroup 1 was more frequently isolated: 37.2% (16/43) vs 7.7% (5/65) (P < 0.001).
- Boilers were rarely contaminated (5/57 samples), at levels between 40 and 5300 cfu/L when water temperature was 40–42 °C, with the exception of one point at 57.8 °C colonised by serogroup 1 (40 cfu/L).
- No contamination was observed at outlets where filters were installed.
- Preliminary results for monochloramine showed a good reduction in contamination during the first month (from 10^3 to 70 cfu/L) but a slight increase in the second month (1600 cfu/L); subsequently no contamination was observed until September 2009.
Horizontal lines represent the limits for intervention according to Italian guidelines.

Comparison of antimicrobial effectiveness and cost of methods used to control legionella contamination

Table I compares the effectiveness and related costs of each method. The decreasing order of effectiveness in reducing contaminated points and points exceeding $10^4$ cfu/L was filter, boiler, chlorine dioxide, hyperchlorination, and superheating. Cost-benefit analysis suggested that chlorine dioxide was the least expensive procedure, followed by thermal shock, hyperchlorination, boiler, and filter.

No cases of hospital-acquired pneumonia were recorded in the hospital during the observation period, so comparative benefit could not be calculated. Review of laboratory data from 2003 to 2008 revealed that 1941 patients with pneumonia were investigated, 13.8% of whom were suffering from suspected hospital-acquired pneumonia. In total, 56 out of 1871 urinary tests and 36 out of 321 serum samples were positive, while 13 of 421 sputum cultures yielded _L. pneumophila_ serogroup 1. All of these were classified as community-acquired pneumonia.

**Discussion**

In our hospital, 14 out of 16 pre-treatment hot water samples were contaminated with _Legionella_ spp. when testing began in 2000. Seven of these were heavily contaminated, $>10^4$ cfu/L, which according to Italian guidelines requires the adoption of control measures. The water distribution systems serving the older buildings were the most widely colonised, as has been reported previously.

Of the more recent buildings, two have never been contaminated whereas the third exhibited high colonisation since we started testing. The materials used for the hot water networks are identical (galvanised steel), as are the constructive methods and building dimensions. The only difference is that the two uncontaminated buildings were constructed rapidly and were occupied immediately after the water systems were tested. In the colonised building, occupation was progressive with some sections of the water system remaining unused for long periods after the system was tested. Thus for new structures we recommend that when testing water system efficiency, water is completely drained until use, or else that the system is immediately put into activity, thus avoiding stagnation.

Once _legionella_ has colonised a water system, our observations support the consensus that eradication is usually unachievable. Superheating has questionable effectiveness and is not suitable for large buildings where temperatures $>60$ °C at each outlet cannot be reliably maintained. Shock hyperchlorination can effectively deal with acute problems but must be performed overnight, which increases the cost. It can also lead to pipe corrosion. Point-of-care filters achieved 100% negative samples but the high costs make them impracticable for widespread application. For example, our hospital has more than 1000 outlets in patients’ bathrooms and sinks, which would cost about €1 million per year to serve with filters.

Chlorine dioxide is highly efficient and is the least expensive procedure, but does not eradicate _legionella_ from the system. In addition, strict control of chlorine injection is required in order to prevent malfunction. One or two days of inadequate levels are sufficient to permit levels to increase again, suggesting that this biocide kills bacteria in the stream but not those inside protozoa. To achieve _L. pneumophila_ concentration $<100$ cfu/L we suggest $>0.3$ ppm chlorine dioxide at outlets, which provides a good compromise between preservation of pipes and infection risk. For reduction below the detection limit, levels of 0.6 ppm are required. A tendency to select serogroup 1 was observed, although this was

**Table 1** Comparison of antimicrobial effectiveness and cost of methods used to control legionella contamination

<table>
<thead>
<tr>
<th>Application</th>
<th>Method</th>
<th>$\Delta$s (pre-treatment - post-treatment)/pre-treatment</th>
<th>$\Delta$s% positive points</th>
<th>$\Delta$s% points $&gt;10^4$ cfu/L</th>
<th>Plant €/year</th>
<th>Live cost €/year</th>
<th>Management €/year</th>
<th>Total €/year</th>
<th>Total €/year/100 water points</th>
</tr>
</thead>
<tbody>
<tr>
<td>Building with 120 bathrooms and 380 water points</td>
<td>Chlorine dioxide (in continuous)</td>
<td>$-46.2$</td>
<td>$-82.3$</td>
<td>2100 (average life 10 years)</td>
<td>900 for ClO$_2$</td>
<td>8,640</td>
<td>1000 for ClO$_2$</td>
<td>11640 per 380 points</td>
<td>3,063</td>
</tr>
<tr>
<td></td>
<td>Shock superheating (monthly)</td>
<td>$-30.5$</td>
<td>$-17.9$</td>
<td>$-400$ for energy</td>
<td>13,700</td>
<td>14100 per 380 points</td>
<td>14100 per 380 points</td>
<td>3,710</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Shock hyperchlorination (monthly)</td>
<td>$-3.8$</td>
<td>$-83.5$</td>
<td>Negligible</td>
<td>28,600</td>
<td>28600 per 380 points</td>
<td>28600 per 380 points</td>
<td>7,526</td>
<td></td>
</tr>
<tr>
<td>Room with 3 water points</td>
<td>Electric boiler</td>
<td>$-94.3$</td>
<td>$-100$</td>
<td>120 (average life 5 years)</td>
<td>120 for energy</td>
<td>Included</td>
<td>240 per three points</td>
<td>240 per three points</td>
<td>8,000</td>
</tr>
<tr>
<td>Single water point</td>
<td>Filter</td>
<td>$-100$</td>
<td>$-100$</td>
<td>936 each filter (average life 30 days)</td>
<td>Included</td>
<td>Included</td>
<td>936 per one point</td>
<td>936 per one point</td>
<td>93,600</td>
</tr>
</tbody>
</table>

$\Delta$s = (pre-treatment - post-treatment)/pre-treatment.
not associated with any clinical cases. For this reason we are also testing monochloramine; preliminary results appear promising but long term assessment is needed to establish its efficacy. Installation of small boilers serving one or two adjacent rooms guarantees absence of contamination provided that the temperature is maintained at >58 °C. Disadvantages include the need to avoid dust and dirt in patients' rooms (so for instance we install boilers above false ceilings), to replace the boiler frequently in case of high water hardness, the cost of energy consumption and the need for a thermostatic mixer valve to avoid the risk of scaling.

Comparing the effectiveness of the procedures in terms of reduction of positive points and of points >10^3 cfu/L, performance was highest for filters, followed by boilers and chlorine dioxide, whereas the effects of hyperchlorination and thermal shock are limited and temporary. In terms of costs, the decreasing order was filter, boiler, hyperchlorination, thermal shock and chlorine dioxide.

We could not evaluate the effectiveness of the applied procedures in reducing risk infection because we have not detected any nosocomial cases. Although routine EIA urinary test to detect Legionnaires' disease could have missed cases of non-serogroup 1 infection, high physician awareness and use of sputum culture and serology should have minimised this risk.36,72 Another is that our control measures have been effective; in our hospital particularly strict control measures (boiler or filter) are adopted for high risk patients and staff are instructed to avoid exposing these patients to tap water. This recommendation is useful in decreasing the incidence of all waterborne nosocomial infections.28

In conclusion, chlorine dioxide represents the best choice for the general protection of patients and healthcare workers whose exposure is documented but disease risk is limited, in association with electric boilers to guarantee high risk patients.29 Both methods require careful management since chlorine dioxide concentrations of <0.3 ppm or boiler temperatures of <58 °C are ineffective. The two solutions applied throughout the entire hospital cost about €70,000 per year, a sustainable sum if the aim of preventing Legionnaires' disease is achieved.

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Conflict of interest statement

None declared.

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