

## **A model for autumn outbreaks of Legionnaires' disease associated with cooling towers, linked to system operation and size**

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### SUMMARY

Cooling towers have been demonstrated to be amplifiers and disseminators of legionella, the causative organism of Legionnaires' disease. Community outbreaks associated with cooling towers have been reported with several common factors. Small towers (< 300 kW) have predominantly been implicated in outbreaks. Cooling tower-associated outbreaks are most frequent in autumn, and frequently implicated systems have been operated after a period of shutdown.

This paper reports field study data relating system operation to legionella colonization of systems. Operating systems have been shown to be more frequently colonized by legionella than shutdown systems. In some cases operation of systems after periods of shutdown raised legionella concentrations from below detection limits to between 50 and 950 c.f.u./ml within 10 min.

These data and previously reported data relating to biofilm and sediment colonization of the systems, and community outbreaks of Legionnaires' disease, have been used to develop a model explaining the seasonal nature of outbreaks associated with irregularly operated, small cooling tower systems.

### INTRODUCTION

Outbreaks of Legionnaires' disease associated with cooling towers have frequently occurred during autumn. Two outbreaks in Australia [1, 2] occurred in late April (mid-autumn). Bhopal and Fallon [3] have reported an autumn peak in incidence of cooling tower-associated outbreaks in Scotland. Reasons for this peak are unclear, and they speculated cleaning of towers or higher legionella concentrations at the end of summer might be contributing factors. Other outbreaks in the UK and US have also generally occurred in the late summer–autumn period. Fliermans [4] has suggested that the seasonal incidence of outbreaks may be due to an autumn bloom of legionella related to enhanced nutrient availability released from seasonal decline of aquatic flora.

The use of cooling towers after periods of inoperation is another factor common to many outbreaks of legionellosis. Dondero and colleagues [5] in the US and Timbury and colleagues [6] in Scotland both reported outbreaks associated with cooling towers which had been inoperative for some time. Onset of symptoms of disease pointed to outbreaks coinciding with these systems being turned on.

Table 1. *Summary of Legionnaires' disease outbreaks from small towers, recently switched on, in autumn*

| Author             | Year | Tower size | Recent operation* | Season                    | Country   |
|--------------------|------|------------|-------------------|---------------------------|-----------|
| Addis et al.       | 1989 | Small      | Yes               | August (early autumn)     | USA       |
| Bhopal et al.      | 1991 | N/A        | N/A               | Autumn                    | Scotland  |
| Breiman et al.     | 1990 | Small      | ?                 | July (summer)             | USA       |
| Christley et al.   | 1989 | Small      | ?                 | April (autumn)            | Australia |
| Christopher et al. | 1987 | Small      | Yes               | April (autumn)            | Australia |
| Dondero et al.     | 1980 | Small      | Yes               | Aug./Sept. (autumn)       | USA       |
| Garbe et al.       | 1985 | Large      | Yes               | June/Aug. (summer)        | USA       |
| Glick et al.       | 1978 | Small      | Sporadic          | July/Aug. (late summer)   | USA       |
| Hunt et al.        | 1991 | ?          | Yes               | Aug.-Oct. (autumn)        | UK        |
| Johnson            | 1986 | Small      | Yes               | January (summer)          | Australia |
| Levy et al.        | 1992 | Small      | ?                 | April (autumn)            | Australia |
| Mitchell et al.    | 1990 | ?          | Yes               | Sept./Oct. (autumn)       | UK        |
| Morton et al.      | 1986 | Small      | ?                 | Sept./Oct. (autumn)       | UK        |
| Nordstrom et al.   | 1983 | Yes        | ?                 | Aug./Sept. (early autumn) | Sweden    |
| O'Mahony           | 1989 | Small      | ?                 | October (autumn)          | UK        |
| Timbury et al.     | 1986 | Small      | Yes               | Oct./Nov. (late autumn)   | Scotland  |

\* Recent operation indicates whether the tower was put into service after a period of shutdown before the outbreak.

† ? indicates details not recorded in the report.

Similar circumstances were reported for other outbreaks [1, 7]. Glick and colleagues [8] reported that the highest attack rates for an outbreak of Pontiac fever, associated with a cooling tower, occurred on Monday mornings. Investigations revealed that the system was switched off on Saturdays and restarted on Monday mornings. This observation was attributed to 'simultaneous exposure on a Monday morning of a large group of susceptible persons'. Stagnation and legionella multiplication in stagnant systems has also been suggested as a causative factor.

Outbreaks of Legionnaires' disease from large systems have not been reported [9]. The majority of outbreaks have been associated with relatively small systems (i.e. < 100 tons or 300 kW). The relevance of cooling tower size in the incidence of outbreaks remains unexplained. Table 1 summarizes common factors relating to outbreaks of legionellosis associated with cooling towers in the summer/autumn period. Other major outbreaks have been reported from small cooling towers during periods of infrequent operation outside the autumn period. Some of these have been at the beginning of the cooling season during periods of intermittent operation, for example outbreaks at Stafford, the B.B.C. building in Portland Place and Piccadilly Circus [18, 19].

Investigations of the colonization of cooling towers by legionella have been carried out in a field study in Adelaide, South Australia. This study has reported that pond water temperature and system operation are major determinants of legionella colonization of systems [20]. As in other studies, these investigations indicated 20 °C as a minimum temperature for legionella multiplication. The role of operation in colonization was attributed to its influence on pond water temperature. Bentham and colleagues [21], in another report from this study,

stated that the planktonic population of legionella in cooling towers is seeded directly from sediments and biofilms, located in the warmer areas of the system pipework. These sites, it was proposed, were the major areas of legionella multiplication within the system.

This paper reports further investigation of the influence of system operation and size on planktonic legionella concentrations and tower colonization. These and historical data have been used to produce a model that may explain the relationships between cooling tower operation and size, and the seasonality of outbreaks of legionellosis.

## MATERIALS AND METHODS

### *Hours of operation*

Samples of cooling-tower pond water (70 ml) were taken twice weekly from a group of 30 cooling towers over a 2-year period. Samples were taken 15 cm below the pond surface and at least 50 cm from the sides of the pond, and away from make-up water inlets. Details of the hours of system operation for each system were recorded using 'hours run' meters attached to the pump on each tower.

### *Surface area: volume ratios of systems*

Towers included in this study varied in size from 30 to 290 kW cooling capacity. For 20 of the 30 towers the surface area:volume ratio of the system was calculated. These ratios were compared with mean summertime pond water legionella concentrations of each system.

For the purpose of these calculations only the surfaces that were permanently wet were included, as it was assumed that intermittently wet surfaces such as the pack, and dry surfaces would be unlikely sites for legionella colonization. Mean summertime (beginning of November to end of April) legionella concentrations were calculated, because intermittently operated systems would otherwise give consistently lower mean results for the whole year than continuously operated systems. This is because system water temperature in seasonally operated towers fall below 20 °C during cooler months, whereas the water temperatures of continuously operated systems do not.

### *Operation of shutdown systems*

In April (autumn) a group of 7 of the above 30 cooling towers were selected. These towers had shown consistent colonization by legionella throughout the summer, and all but one tower (A) had been shut down for a minimum of 7 days (maximum 44 days, Tower F).

Water samples (70 ml) were taken from the basins of these towers as previously described and the systems turned on. Water samples were taken again after 10 min, and in one case after 70 min operation.

### *Sample treatment*

Samples were not concentrated, but were pretreated by heat at 50 °C for 30 min [22] and 0.025 ml inoculated on Oxoid-buffered charcoal yeast extract agar supplemented with 80 mg/l cycloheximide (BCYE) and 0.25 ml inoculated on

Oxoid Modified Wadowsky and Yee selective agar supplemented with 80 mg/l of cycloheximide (MWY). This gave a limit of detection of 4 c.f.u./ml. Plates were incubated at 37 °C in 5% CO<sub>2</sub> for 7 days, and examined using a plate microscope, and under long-wave u.v. light to detect autofluorescent species.

Presumptive identification of suspect legionella colonies was by subculture on to BCYE and blood agar plates and testing by latex agglutination tests (Disposable Products Pty Ltd, Technology Park, Adelaide, South Australia 5000) for *L. pneumophila* SG 1-14.

### *Data analysis*

Possible correlations between the hours of operation and legionella colonization and concentration were investigated by statistical analyses. Non-parametric statistical analysis was used for the data, as the legionella colonization values were not distributed normally.

## RESULTS

### *Hours of operation and legionella concentration in pond water*

Grouping the tower operation data into 10-h ranges (see Table 2) showed that the percentage of pond water samples containing legionella was least when towers were not operating. The numbers of legionella in water samples from non-operating towers were noticeably less than those of operating towers. Increasing the hours of operation of the towers did not appear to cause a corresponding increase in the number of towers with legionella in the pond water.

### *Hours of operation and legionella concentration*

The data showed low legionella concentrations in samples from non-operating systems (Table 2). In operation systems pond water legionella concentrations were significantly greater. There was no significant difference between legionella concentrations in any of the operating ranges.

### *Surface area:volume ratios*

Two of the towers were excluded from analysis because there was limited data for them. Statistical analysis showed a significant correlation,  $r = 0.48$ , ( $P < 0.05$ ), between legionella concentrations and permanently wet surface area:volume ratio of the systems (Fig. 1). The data showed a trend of increasing mean legionella concentration with increasing surface area:volume ratio of the systems (Fig. 1).

### *Operation of shutdown systems*

As shown in Table 3 legionella was detected before operation in 2 of the 7 systems tested (A and E). After 10 min operation, 5 of the systems showed detectable legionella concentrations in the tower water (A, B, C, E and G). In two of these systems legionella counts rose from undetectable to exceed 200 c.f.u./ml (B and C). In the two towers with legionella in the pond water prior to operation, counts were raised after operation, but not significantly (A and E). Tower G (Table

Table 2. *The number of positive legionella samples recorded with varying hours of cooling tower operation*

| Hours of operation (per 4 days) | No. of samples | No. of samples colonized | Percentage of samples colonized | Range of legionella concentrations | Mean legionella concentration |
|---------------------------------|----------------|--------------------------|---------------------------------|------------------------------------|-------------------------------|
| 0                               | 454            | 74                       | 16.3                            | 20.0-31.6                          | 25.8                          |
| 1-10                            | 143            | 59                       | 41.3                            | 74.2-109.6                         | 91.9                          |
| 11-20                           | 68             | 37                       | 54.4                            | 83.6-134.6                         | 109.1                         |
| 21-30                           | 73             | 53                       | 72.6                            | 161.6-230.0                        | 195.8                         |
| 31-40                           | 38             | 29                       | 76.3                            | 69.4-121.6                         | 95.5                          |
| 41-50                           | 54             | 36                       | 66.7                            | 61.8-120.0                         | 90.9                          |
| 51-60                           | 45             | 33                       | 73.3                            | 75.0-154.6                         | 114.8                         |
| 61-70                           | 48             | 39                       | 81.3                            | 163.1-251.1                        | 207.1                         |
| 71-80                           | 55             | 36                       | 65.5                            | 93.1-169.1                         | 131.1                         |
| 81-90                           | 13             | 9                        | 69.2                            | 20.9-45.9                          | 33.4                          |
| 91-100                          | 36             | 19                       | 52.8                            | 52.9-129.1                         | 91.0                          |

'Hours of operation' refers to the 4-day period prior to sampling. Legionella concentrations are expressed as c.f.u./ml.

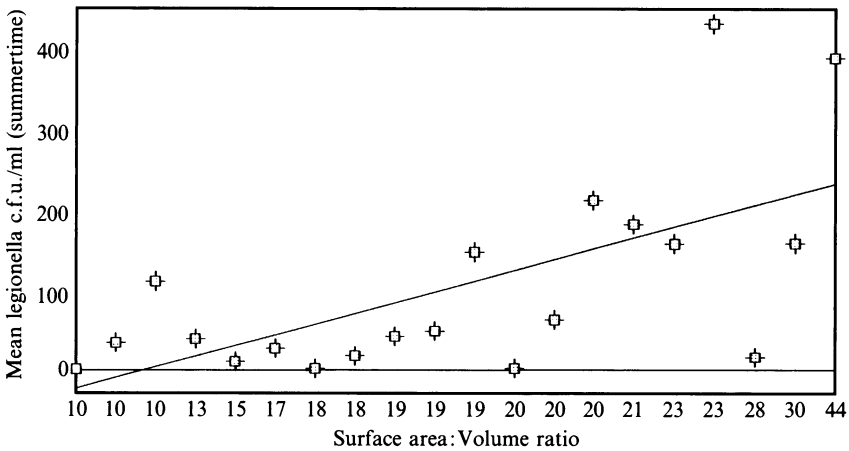


Fig. 1. Variation of mean summertime legionella counts in basin water with increasing surface area: volume ratio of the system. (Surface areas calculated from permanently wet surfaces only.)

Table 3. *Legionella concentrations cultured from cooling towers before and after system operation*

| Tower | Legionella concentrations* at |        |        |
|-------|-------------------------------|--------|--------|
|       | 0 min                         | 10 min | 70 min |
| A     | 20                            | 60     | —      |
| B     | 0                             | 232    | —      |
| C     | 0                             | 920    | —      |
| D     | 0                             | 0      | —      |
| E     | 4                             | 40     | —      |
| F     | 0                             | 0      | —      |
| G     | 0                             | 8      | 40     |

\* Legionella concentrations are expressed as c.f.u./ml.

3), when tested after 70 min operation, showed increased legionella concentration from that observed after 10 min. Other towers were not sampled after this time period.

#### DISCUSSION

The results of this investigation have indicated that the incidence of legionella and concentration of legionella in the pond water is influenced by tower operation. The duration of operation does not appear to influence the extent of colonization of legionella concentrations within the systems. This suggests that the effect of operation is not related to introduction of material into the system or by other changes in the environment which may increase with increasing operation, such as water temperature.

The tendency for elevated legionella concentrations to be found with increasing system 'wet' surface area:volume ratio may be attributable to the biofilm complement of the legionella population of the system. Higher pond water legionella concentrations could be expected in systems with a greater surface area for biofilm development and sediment deposition.

The increase in legionella concentrations in systems shortly after being put into service is too rapid to be explained by their multiplication in such a short period. Results from this study reported previously that the planktonic population in pond water is seeded directly from biofilms [21]. The increase is most probably explained by seeding and sloughing of biofilms and disturbance of sediments by water flow through the system.

In periods when towers are shut down, but ambient temperatures remain above 20 °C, legionella populations in sediments and biofilms will continue to multiply [20]. Those areas of the system that are contained within buildings may also be expected to remain at temperatures suitable for legionella multiplication. This multiplication is less likely to be inhibited by residual biocide concentrations than in operating systems. Active biocide concentrations are likely to be negligible or short lived in these circumstances, especially if non-penetrant or oxidizing biocides are being employed.

Cessation of water flow over the biofilms will reduce oxygen and nutrient availability within biofilms. This may result in areas of starvation, causing loosening of attachment of biofilms [19]. Once the system is brought back into operation, the water flow would then distribute the loosened biofilms through the circulating water. This phenomenon may be used to explain the observed sudden rise in legionella concentrations once towers were operated (see Table 3).

In autumn, legionella populations in cooling towers are likely to be well established after the summer period. If systems are put back into service after a short period of inactivity, i.e. in response to a period of warm weather ('Indian summer'), a sudden and very rapid increase in planktonic legionella concentrations could be expected. This occurrence has been demonstrated in this study. This increase in legionella concentrations in tower water may coincide with reduced sunlight intensity and increased relative humidity, factors that would prolong the viability of cells entrained in aerosols [7].

During an investigation of an outbreak of Pontiac fever in an automobile plant [24], onset of the outbreak coincided with operation of an aerosol-producing

industrial coolant system which had been out of use for the previous week. It was speculated that, during the period of disuse, bacterial overgrowth had led to favourable conditions for legionella growth. Upon resumption of use the system had then disseminated aerosols to the factory workers. This speculative model presumes a rapid multiplication of legionella, after 'overgrowth of bacteria' within a 7-day period. It may be hypothesized that the growth, and particularly, the loosening of an already present biofilm population of legionella during system disuse may have occurred. The disturbance and dissemination of this population when the system was restarted would explain the concentrations of legionella in the aerosols without a requirement for a rapid multiplication over a short period.

Glick and colleagues [8] observed that the highest attack rate for an outbreak of Pontiac fever in a health department occurred on Monday mornings. The tower implicated in this outbreak was shut down on Saturdays and restarted on Monday mornings. This operating routine may have resulted in a temporary increase in the legionella concentrations in the system water due to biofilm loosening and then disturbance, resulting in elevated dose concentrations being inhaled in aerosols.

Large refrigeration-cooling water systems tend to operate continuously until ambient temperatures fall below 20 °C. In these towers legionella concentrations are not likely to increase after shutdown. Further, continuously operating systems allow dispersion of biocide preparations throughout the system, which affords some control of biofilm populations.

Many cooling water systems are thermostatically controlled, and operation of the tower is dependent on heat load. In large towers, when heat loads are low the system pumps may circulate water without the fans running. This avoids cooling the water to temperatures that would excessively lower the refrigerant in the condenser below its boiling point and cause the system to shut down on low evaporating pressure. If towers are operated without fans running there will be a dramatic reduction in the amount of aerosol generated.

Like larger towers, many small systems operate by thermostatic control; however, once heat loads are sufficient to trigger operation it is not uncommon for the system to circulate water with the fans on. This means that under these conditions, small towers are likely to generate more aerosol than large towers.

Smaller systems (< 100 tons/300 kW) are generally used to service irregular (beer coolers) or seasonal (air-conditioning) heat loads. These systems are commonly shut down when ambient temperatures drop to around 20 °C, temperatures suitable for legionella multiplication. In autumn, these smaller systems are more likely to be subject to operation after short periods of inactivity. It may take several days of ambient temperatures of 20 °C or greater to warm buildings sufficiently for these towers to be operated. If this occurs, once towers are switched on the generation of high concentrations of legionella in tower water is possible in a short period, through biofilm and sediment disturbance.

There is a general tendency for the surface area: volume ratio of cooling water systems to decrease as system size increases. The data presented in this study suggest that the legionella concentrations in the system water may therefore decrease as system size increases.

In summary, this paper has proposed a model explaining the frequency of autumn outbreaks associated with small cooling towers recently returned to

service. This model may be extended to explain summer outbreaks as well, but does not seek to explain all outbreaks associated with cooling towers, as other factors may be involved.

It is important to note that measures can be taken to avoid the circumstances leading to this type of occurrence. Draining of the water from the entire systems of small towers at autumn shutdown may prevent legionella multiplication. If further operation of the towers is required after draining, the cost of refilling would be minimal. It has been recommended that air-cooled refrigerative units be used for heat loads of less than 300 kW [25, 26]. It is noticeable that all the outbreaks listed in Table 1 were attributed to cooling towers of less than 300 kW. Smith and Stewart [27] reported that replacement of cooling towers in the UK by air-cooled units is economically viable for system capacities of up to 1200 kW. The majority of cooling tower-associated Legionnaires' disease outbreaks, worldwide, have been attributed to small units [9]. Installation of air-cooled systems instead of these systems would have avoided these outbreaks and might have been mechanically and economically superior.

This model may be applicable to other sources of Legionnaires' disease outbreaks, such as whirlpools, showers, misting machines and fountains. Periods of disuse of contaminated systems at temperatures suitable for legionella multiplication are likely to result in transient high concentrations of these and other bacteria in the water on resumption of operation.

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