

Health risks from exposure to *Legionella* in reclaimed water aerosols: Toilet flushing, spray irrigation, and cooling towers

Kerry A. Hamilton ^{a,*}, Mark T. Hamilton ^b, William Johnson ^c, Patrick Jjemba ^c, Zia Bukhari ^c, Mark LeChevallier ^c, Charles N. Haas ^a

^a Drexel University, 3141 Chestnut Street, Philadelphia, PA 19104, USA

^b Yale University, New Haven, CT 06520, USA

^c American Water Research Laboratory, 213 Carriage Lane, Delran, New Jersey 08075, USA

ARTICLE INFO

Article history:

Received 2 August 2017

Received in revised form

30 November 2017

Accepted 12 December 2017

Available online 13 December 2017

Keywords:

Reclaimed water

Quantitative microbial risk assessment (QMRA)

Legionella pneumophila

Cooling towers

Toilet flushing

Spray irrigation

ABSTRACT

The use of reclaimed water brings new challenges for the water industry in terms of maintaining water quality while increasing sustainability. Increased attention has been devoted to opportunistic pathogens, especially *Legionella pneumophila*, due to its growing importance as a portion of the waterborne disease burden in the United States. Infection occurs when a person inhales a mist containing *Legionella* bacteria. The top three uses for reclaimed water (cooling towers, spray irrigation, and toilet flushing) that generate aerosols were evaluated for *Legionella* health risks in reclaimed water using quantitative microbial risk assessment (QMRA). Risks are compared using data from nineteen United States reclaimed water utilities measured with culture-based methods, quantitative PCR (qPCR), and ethidium-monoazide-qPCR. Median toilet flushing annual infection risks exceeded 10^{-4} considering multiple toilet types, while median clinical severity infection risks did not exceed this value. Sprinkler and cooling tower risks varied depending on meteorological conditions and operational characteristics such as drift eliminator performance. However, the greatest differences between risk scenarios were due to 1) the dose response model used (infection or clinical severity infection) 2) population at risk considered (residential or occupational) and 3) differences in laboratory analytical method. Theoretical setback distances necessary to achieve a median annual infection risk level of 10^{-4} are proposed for spray irrigation and cooling towers. In both cooling tower and sprinkler cases, *Legionella* infection risks were non-trivial at potentially large setback distances, and indicate other simultaneous management practices could be needed to manage risks. The sensitivity analysis indicated that the most influential factors for variability in risks were the concentration of *Legionella* and aerosol partitioning and/or efficiency across all models, highlighting the importance of strategies to manage *Legionella* occurrence in reclaimed water.

© 2018 Elsevier Ltd. All rights reserved.

1. Introduction

Growing global water scarcity has intensified the need to recover water resources from wastewater, especially as population growth, economic development, and urbanization increase pressures on existing water supplies (Levine and Asano, 2004). Reclaimed water can alleviate stress on municipal water systems and augment existing water portfolios. However, there is the potential for health risks from human contact with contaminants in

reclaimed water through dermal contact, inhalation, or ingestion during various water use activities.

Agricultural and industrial water reuse represent the sectors with the largest reclaimed water usage in the United States (Jiménez and Asano, 2008). Reclaimed water for cooling system purposes further represents the largest industrial water reuse application (Metcalf and Eddy, 2007). Cooling systems may consume 20–50% of a facility's water usage (Aoki et al., 2005). Common uses of reclaimed water such as spray irrigation or cooling towers can produce aerosols that are of concern because contaminants can travel beyond the immediate vicinity of application (Li et al., 2011). Additionally, toilet flushing has been identified as a top use of recycled water in a survey of ten United States recycled water systems (Jjemba et al., 2015; LeChevallier et al., 2017). Toilet

* Corresponding author. Drexel University, Department of Civil, Architectural, and Environmental Engineering, 3141 Chestnut Street, Philadelphia, PA, 19104, USA.
E-mail address: kh495@drexel.edu (K.A. Hamilton).

flushing can also generate aerosols that can potentially be associated with health risks (Fewtrell and Kay, 2007; Gerba et al., 1975; Hamilton et al., 2017). Together, spray irrigation (90% respondents), cooling towers (50% respondents), and toilet flushing (30% respondents) comprise the top three uses of recycled water identified in the survey (Jjemba et al., 2015; LeChevallier et al., 2017).

To date, no documented infectious disease outbreaks have been reported in association with reclaimed water, and epidemiological studies have focused primarily on potential fecal pathogen-associated health risks (Durand and Schwebach, 1989; Sheikh et al., 1990; Ward et al., 1989), without finding evidence of increased risk. One recent study of irrigation workers exposed to reclaimed water showed higher colonization with *Staphylococcus* and *Enterococcus* bacteria compared to office workers, although this difference was not significant (Goldstein et al., 2014). However, concerns exist regarding opportunistic pathogens such as *Legionella*, non-tuberculous *Mycobacteria* (NTM), *Pseudomonas aeruginosa*, *Stenotrophomonas maltophilia*, *Staphylococcus aureus*, and *Acanthamoeba* spp., especially as alternative non-potable routes of exposure are more relevant for reclaimed water use. The chemistry of reclaimed water is distinct from that of potable water, with elevated levels of dissolved organic matter and nutrients known to occur in reclaimed water systems associated with enhanced microbial growth (Garner et al., 2016). Intermittent usage of reclaimed water may also result in higher water age. Opportunistic pathogens can grow at low organic carbon concentrations, and are particularly prone to growth in water environments with high water age and biofilm growth, where free-living amoeba found in biofilms can enhance pathogen resistance to disinfectants, growth, persistence, and virulence (Cooper and Hanlon, 2010; Kirschner et al., 1992; Thomas et al., 2010). Although fully functioning water reuse treatment may be sufficiently robust to be protective for fecal pathogen risks (Chaudhry et al., 2017), it is not yet known whether the same is true for opportunistic pathogens.

Legionella is one of the most significant opportunistic waterborne pathogens as it is responsible for a substantial portion of the United States waterborne disease burden (Beer et al., 2015). Infection with *Legionella* can cause the pneumonia-like illness Legionnaires' Disease, or a milder form of infection known as Pontiac Fever (Fields et al., 2002). It is known to occur in engineered water systems including cooling towers (Ahmadrajabai et al., 2016), wastewater treatment facilities (Allestam et al., 2006; Fernandez-Cassi et al., 2016; Pascual et al., 2003; Sánchez-Monedero et al., 2008; Walser et al., 2017), ambient water environments (Fliermans et al., 1979), and soils (Amemura-Maekawa et al., 2012; Wallis and Robinson, 2005). *Legionella* spp. grow in biofilms in piping and can slough off and become aerosolized through water fixtures, at which point human exposure can occur. Aerosol production of *Legionella* is a particular concern as outbreaks have been associated with exposure to aerosols generated by cooling towers (Castilla et al., 2008; George et al., 2016; Nguyen et al., 2006; Walser et al., 2014; Weiss et al., 2017), wastewater treatment facilities (Gegersen et al., 1999; Kusnetsov et al., 2010; Olsen et al., 2010), decorative fountains (Haupt et al., 2012), and other common water uses (Hines et al., 2014). For this reason, it is warranted to investigate the potential for elevated risks from exposure to *Legionella* in reclaimed water.

Few studies have quantified *Legionella* in reclaimed water (Jjemba et al., 2010; Palmer et al., 1995), and a quantitative microbial risk assessment (QMRA) has not yet been performed for *Legionella* for exposures to reclaimed water. The QMRA framework integrates information regarding pathogen occurrence, infectivity, and exposure for determining the health implications of microbial hazards using a process of hazard identification, exposure

assessment, dose response assessment, and risk characterization (Haas et al., 2014). To inform appropriate usages of reclaimed water and identify factors which have the greatest implication for best management practices, a QMRA is presented for scenarios of toilet flushing, spray irrigation, and cooling tower-generated aerosols.

Additionally, due to the availability and practice of using different laboratory detection methods to enumerate *Legionella* spp., risks were also assessed using a comparison of three common detection methods: culture-based methods, ethidium-monoazide quantitative polymerase chain reaction (EMA-qPCR), and qPCR. Culture-based methods quantify viable, culturable cells (colony forming units [CFU] per L). EMA-qPCR quantifies gene copies from cells with an intact cell membrane and is intended to represent the concentration of viable cells as measured in gene copies per L (Delgado-Viscogliosi et al., 2005; Mansi et al., 2014; Qin et al., 2012). Typically, a QMRA will seek to use data that are most representative of viable, infectious microorganisms. However, these data are not always available for a particular situation, and it is not uncommon for QMRAs to rely upon qPCR data when viability and infectivity information is not available. Each detection method has its drawbacks (Whiley and Taylor, 2014), and there is currently no consensus regarding the implications of using datasets generated with varying methods on resulting health risks. Therefore, an additional objective of this work was to quantify the impacts of laboratory detection method on QMRA estimates using concentrations generated using all three methods (culture, EMA-qPCR, qPCR) from a large national study of *Legionella* occurrence.

2. Materials and methods

2.1. Exposure models

Three exposure scenarios of toilet flushing, spray irrigation, and cooling tower-generated aerosols were considered using a previously derived framework for *Legionella* risks (Hamilton and Haas, 2016). *Legionella* was considered to be present in reclaimed water at concentrations measured in a comprehensive national *Legionella* occurrence study by Johnson et al. (2017), with aerosols generated at rates specific to each process modeled. Not all aerosols released from a given activity will reach a receptor while possessing droplet diameters within the respirable range. Aerosol particles of median diameters between 1 and 10 μm were considered respirable for all three exposure scenarios, as *Legionella* is typically 1–2 μm long and 0.3–0.9 μm wide, and particles greater than 10 μm are not likely to reach the lower respiratory tract (Baron and Willeke, 1986; Metcalf and Eddy, 2007). Where available, information was used regarding how the bacteria partitions from bulk water to aerosol, and pathogen decay was considered during transport through air to a receptor. Most large aerosol droplets are trapped in the nasopharyngeal region, and smaller particles are able to travel to the alveoli, where *Legionella* infection is initiated and behaves according to an exponential dose response model. Human exposure patterns for each scenario were taken into account to annualize risks. Where necessary, data from published graphs necessary for aerosol calculations were extracted using aLcAsA Digitize It[®] v. 4.2.0.

2.1.1. Toilet flushing

Three QMRA methods were compared to assess infection risks from toilet flushing with multiple common toilet types (Johnson et al., 2013; O'Toole et al., 2009). The use of either a partitioning coefficient (C) (Armstrong and Haas, 2007b, 2008; Azuma et al., 2013; Medema et al., 2004; Sales-Ortells and Medema, 2014, 2015; Schoen and Ashbolt, 2011) or a calculated aerosol dose using the concentration of aerosols and volume of aerosols in relevant

size bins to estimate inhaled dose (Lim et al., 2015) are accepted QMRA practices for modeling aerosol inhalation exposures. Therefore, both the PC (Method 1) and aerosol dose (Method 2 and 3) methods are assessed and compared using two available datasets for toilet aerosol size distributions (Johnson et al., 2013; O'Toole et al., 2009) (Method 2 and 3).

Method 1 for toilet flushing is modified from the QMRA exposure model of Schoen and Ashbolt (2011) originally developed for showering and relying on the use of a PC as per Equation (1). Aerosols generated with the PC were only considered for sizes between 1 and 4 µm due to the upper bound PC used. The PC was derived from a study by Darlow and Bale (1959), and analyzed by Hines et al. (2014), indicating that 87% of produced aerosols were less than 4 µm in diameter, with no size information about other aerosol sizes produced. The study used for the lower bound estimate by Barker and Jones (2005) did not indicate the size of aerosols produced and the same aerosol size fraction (87% between 1 and 4 µm) was applied for all PC values (see section 3.2.1).

$$Dose_{Leg,pf} = C_{Leg} PC_{wa} It f_{1-4} F_{1-4} DE_{1-4} \quad (1)$$

where $Dose_{Leg,pf}$ = the dose of *Legionella* deposited in the lungs per toilet flush [Number of *Legionella*]; C_{Leg} = the concentration of *Legionella* in bulk water [# / L]; PC_{wa} = bacterial water to air partitioning coefficient [CFU m⁻³ / CFU L⁻¹]; I = the mean inhalation rate of air breathed after toilet flushing [m³ air/min]; t is the exposure duration or time spent in the room after toilet flushing [min]; f_{1-4} is the fraction of *Legionella* that partition to each of the 1 through 4 µm aerosol diameter fractions; F_{1-4} is the fraction of aerosols that are between diameter 1 and 4 µm (87%); and DE = the alveolar deposition efficiency of 1 through 4 µm diameter aerosols [fraction].

Method 2 is modified from the approach of Lim et al. (2015) using the concentration of 2.5 µm median diameter aerosols produced by a toilet flush measured 420 mm above a toilet by a cistern toilet suite (O'Toole et al., 2009) (Equation (2)).

$$Dose_{Leg,pf} = C_{Leg} C_{aero,2.5} V_{aero,2.5} It F_{2.5} DE_{2.5} \quad (2)$$

where $Dose_{Leg,pf}$ = the dose of *Legionella* deposited in the lungs per toilet flush; C_{Leg} = the concentration of *Legionella* in bulk water; $C_{aero,2.5}$ = the concentration of aerosols of diameter 2.5 µm measured 420 mm above the toilet by O'Toole et al. (2009) [# / cm³ converted to # / m³ by multiplying by a factor of 10⁶]; $V_{aero,2.5}$ = the volume of 2.5 µm aerosols [L/aerosol] calculated as $V = (4/3)\pi r^3$ where $d = 2r = 2.5 \times 10^{-6}$ m; I = the mean inhalation rate of air breathed after toilet flushing [m³ air/min]; t is the exposure duration or time spent in the room after toilet flushing [min]; and $F_{2.5}$ is the fraction of *Legionella* that partition to the 2.5 µm aerosol fraction.

Method 3 uses the approach Lim et al. (2015) as above, but uses recent data on the aerosol generation rate from modern flush toilets provided by Johnson et al. (2013) (Equation (3)).

$$Dose_{Leg,pf} = C_{Leg} It \sum_{i=1}^{10} C_{aer,i} V_{aer,i} F_i DE_i \quad (3)$$

where $Dose_{Leg,pf}$ = the dose of *Legionella* deposited in the lungs per toilet flush; C_{Leg} = the concentration of *Legionella* in bulk water; $C_{aer,i}$ = the average concentration of aerosols of each diameter of each MMAD i ; $V_{aer,i}$ = the volume of each MMAD i aerosols calculated as $V = (4/3)\pi r^3$ where diameters ranged from 1 to 10 µm; DE_i = the alveolar deposition efficiencies of aerosols of each size MMAD i ; I = the mean inhalation rate after toilet flushing (m³ air/

min); and t is the exposure duration or time spent in the room after toilet flushing (min).

2.1.2. Atmospheric dispersion model for cooling towers and spray irrigation

The primary types of atmospheric models for particle dispersion are simple box, Gaussian Plume (GP), Lagrangian, and Eulerian (Holmes and Morawska, 2006). Dungan (2010) and Van Leuken et al. (2015) reviewed fate and transport models for bioaerosols, which relied heavily upon modified GP models. QMRA models for wastewater, biosolids use, and spread of dusts containing pathogens between farms used GP models with various modifications (Brooks et al. 2005b, 2012; Dowd et al., 2000; Galada et al., 2012; Jahne et al. 2014, 2015; Ssematimba et al., 2012; Tanner et al., 2008; Teng et al., 2013; Viau et al., 2011), with several studies generating site specific, meteorological data-intensive estimates using US Environmental Protection Agency AERMOD software (Dungan, 2014; Jahne et al. 2014, 2015) or computational fluid dynamics (CFD) (Blatny et al. 2008, 2011; Fossum et al., 2012). The goal of this work was to develop a generalized model for long-range transport with reclaimed water containing *Legionella* under a range of meteorological conditions that does not rely upon intensive site-specific information.

The concentration of *Legionella* downwind from an aerosol-emitting source is dependent upon the concentration of *Legionella* in the originating reclaimed water, transport and dispersion, deposition (wet and dry), evaporation, and bacterial viability as a function of environmental conditions. These factors are incorporated into a Gaussian plume atmospheric transport model to calculate the dose of *Legionella* at a receptor downwind from a cooling tower or irrigation spray source using Equation (4), a combination of previously proposed models that account for organism decay within the plume (Lighthart and Mohr, 1987; Peterson and Lighthart, 1977; Teltsch et al., 1980; USEPA, 1982).

$$Dose(x,y,z) = \frac{Q_{Leg} It}{2I\mu\sigma_y\sigma_z} \exp\left[\left(\frac{-y}{2\sigma_y}\right)^2\right] \left\{ \exp\left[\frac{-(z-H_e)^2}{2\sigma_z^2}\right] + \exp\left[\frac{-(z+H_e)^2}{2\sigma_z^2}\right] \right\} \sum_{i=1}^n q_{i,s} F_i DE_i \exp\left[\frac{-(\lambda_s + \lambda_{uv})x}{\mu}\right] \quad (4)$$

Where $Dose(x,y,z)$ = Dose of *Legionella* at x , y , and z meters downwind from the source (Number of *Legionella* bacteria), x = distance downwind (m), y = horizontal distance perpendicular to wind (m) z = downwind receptor breathing zone height (1.5 m), Q_{Leg} = emission rate of *Legionella* bacteria [Number per s] (see Equation (7)); H_e = Effective height of plume source from ground level (m) calculated as the maximum stream height for sprinklers or the height of a cooling tower, μ = wind velocity [(m/s), determined by stability categories in Table 1], $q_{i,s}$ = the mass-weighted proportion of aerosols in each size i where $i = 1:10$ in the evaporated or aqueous aerosol state s (assumed to be uniform fractions); λ_s = microbial decay coefficient due to non-solar factors [s⁻¹]; λ_{uv} = solar microbial decay coefficient [s⁻¹]; σ_y = horizontal dispersion coefficient (m), σ_z = vertical dispersion coefficient (m), I = the mean inhalation (m³ air/min); and t is the exposure duration [min]. Downwind distances ranging from 50 to 10,000 m were simulated as this is the applicable range of the Gaussian plume model (Lighthart, 1994). Exposure was assumed to occur in line with the plume centerline ($y = 0$), which would be the maximum concentration distribution observed at a given distance (x). Dispersion coefficients were calculated as per Equations (5) and (6) where R_y , r_y , R_z and r_z are constants (Table 1).

Table 1
Pasquill Stability Classes for moderate solar radiation (Seinfeld, 1986).

Stability class (Moderate incoming solar radiation)	Wind speed (m/s)	R_y	r_y	R_z	r_z
A	1	0.469	0.903	0.017	1.380
B	3	0.306	0.885	0.072	1.021
C	5	0.230	0.855	0.076	0.879
D	7	0.219	0.764	0.140	0.727

$$\sigma_y = R_y x^{r_y} \quad (5)$$

$$\sigma_z = R_z x^{r_z} \quad (6)$$

Q_{Leg} is defined as per Equation (7):

$$Q_{Leg} = C_{Leg} F E \quad (7)$$

where C_{Leg} = concentration of *Legionella* in reclaimed water [organisms/L]; F = flow rate [L/s]; E = aerosolization efficiency = fraction of sprayed reclaimed water that leaves the immediate vicinity of the spray irrigation system as aerosols ($0 < E \leq 1$).

Assumptions inherent in this model are 1) the background concentration of aerosolized *Legionella* spp. in ambient air is negligible; 2) reclaimed water aerosols are generated during day-time only (only daytime solar insolation values and corresponding atmospheric stability values only are considered; 3) no overlapping cooling tower, irrigation sources, or other sources of *Legionella* in the system; 4) exposures occur at a constant distance directly downwind from the sprinkler or cooling tower; 5) protection of *Legionella* due to the presence of organic debris, algae, or free-living amoeba is not considered; 6) the impacts of aerosol dynamics including bubble burst, break up or agglomeration of aerosols, film collapse, and shear forces on *Legionella* are not considered; 7) effects of a moist aerosol plume thermodynamics are not considered; 8) no topographic effects; 9) no additional effects of biofilms and any biofilm with potential to slough off pipe surfaces was suspended in the bulk water at the time of sampling, therefore *Legionella* in bulk water represent 100% of *Legionella* available for aerosolization; 10) the fate of bacteria in individual aerosols is not tracked, however it is acknowledged here that larger aerosols in the starting distribution are likely to contain more bacteria and therefore result in higher concentration aerosols downstream than expected in some aerosols of smaller diameter (Blatny et al., 2011); 11) enrichment of the aerosolized water with bacteria compared to the bulk water is not considered; 12) reclaimed water is not blended with any other water source prior to use.

The plume model accounts for dispersion, but not the fraction of aerosols within the respirable size range, which is of crucial concern for *Legionella* inhalation (1–10 μm). To obtain this fraction, the approach of Hardy et al. (2006) was used, considering the mass-weighted fraction of aerosols likely to become fully evaporated as those that are $< 100 \mu\text{m}$ in diameter, and the fine mist fraction as those droplets with diameters 100–200 μm in diameter (represented in the model as $q_{i,s}$). Aerosols larger than 400 μm settled at distances $< 50 \text{ m}$ and were therefore assumed to settle at close range for both sprinklers and cooling towers and were not included in the model. It was assumed that all droplets with an initial diameter of $< 200 \mu\text{m}$ would reach a diameter of 10 μm or less by the time they reached the downwind receptor.

2.2. Legionella concentrations

For all models, concentrations of *Legionella* in reclaimed water C_{Leg} were computed from a national study (Johnson et al., 2017)

according to Equation (8):

$$C_{Leg} = \frac{1}{R} C_{RW} f_{LP} \quad (8)$$

where C_{Leg} = corrected concentration of *Legionella pneumophila* in reclaimed water or drinking water; R = recovery efficiency; C_{RW} = concentration of *Legionella* spp. measured in reclaimed water or drinking water (C_{DW}); and f_{LP} = fraction of *Legionella* spp. in the analytical method identified as *Legionella pneumophila*. Because some samples did not have any detectable *Legionella* (i.e., were below the limit of detection), interval-censored distributions were fit to concentration data using the package `fitdistrplus` (Delignette-Muller and Dutang, 2015) in R v.3.1.1. (www.rproject.org) whereby non-detect observations were censored between zero and the detection limit.

2.3. Dose response and risk characterization

The daily probability of each endpoint was calculated using the exponential dose response model for *L. pneumophila* (Equation (9)) (Armstrong and Haas, 2007a; Haas et al., 1999). Two dose response models were used for infection (corresponding to subclinical infection or potentially a Pontiac Fever endpoint) or clinical severity infection (corresponding to an infection requiring a clinical visit) (Table 2).

$$P_{inf} = 1 - e^{-rd} \quad (9)$$

where P_{inf} is the probability of infection or clinical illness per event, r is the probability of the bacteria bypassing the host defenses and initiating a given response, and d is the dose of *Legionella* at the target organ (alveoli). Annual risk was calculated as per Equation (10).

$$P_{inf,annual} = 1 - \prod_{i=1}^{nf} (1 - P_{inf}) \quad (10)$$

where f is the daily frequency of the activity (flushing a toilet, spray irrigation application, or being present outside near a cooling tower) and n is the yearly frequency.

A sensitivity analysis was conducted to identify variables contributing to uncertainty using 10,000 Monte Carlo iterations. All computations were performed in R and using the `mc2d` package (Pouillot and Delignette-Muller, 2010). The Spearman rank correlation coefficient was used to identify the most important predictive factors of annual infection or clinical severity infection risk, where 0 is no influence and -1 or $+1$ when the output is wholly dependent on that input. The model inputs were ranked based on their correlation coefficient with the output variable, annual risk.

3. Results

3.1. Legionella concentrations in reclaimed water

Concentrations of *Legionella pneumophila* in reclaimed water

Table 2
Monte Carlo model risk characterization input parameters.

Parameter	Symbol	Unit	Value	Distribution	Reference
All models					
Inhalation rate, light activity, breathing cycle period 8 s I and 1 L tidal volume		m ³ /min	Min = 0.013, Max = 0.017	Uniform	(USEPA, 2011)
Dose response parameter for <i>L. pneumophila</i> , infection endpoint	r_{inf}	Unitless	$\mu = -2.934$, $\sigma = 0.488$	Lognormal ^a	(Armstrong and Haas, 2007a; Muller et al., 1983)
Dose response parameter for <i>L. pneumophila</i> , clinical severity infection endpoint	r_{csi}	Unitless	$\mu = -9.688$, $\sigma = 0.296$	Lognormal	(Armstrong and Haas, 2007a; Fitzgeorge et al., 1983)
Toilet flushing					
Exposure frequency	f_{toilet}	Flushes/day	$\mu = 5.05$, $\sigma = 2.69$	Lognormal	(Mayer and DeOreo, 1999)
Exposure duration	t_{toilet}	Min/flush	1–5	Uniform	(Lim et al., 2015)
Spray irrigation					
Exposure frequency	f_{IR}	Days/year	Residential: Min = 81, Max = 99 Occupational: 255	Uniform Point	(Brooks et al., 2012; Brooks et al., 2005b; Chhipi-Shrestha et al., 2017; NRMCM, 2008)
Exposure duration	t_{IR}	Hours/day	Residential: 1 Occupational: 8	Point	(Brooks et al., 2005a; Brooks et al., 2012)
Cooling towers					
Exposure frequency	f_{CT}	Days/year	Residential: 365 Occupational: 255	Point	(Bhopal and Barr, 1990; Brooks et al., 2005a; Brooks et al., 2012)
Exposure duration	t_{CT}	Hours/day	Residential: 1 Occupational: 8	Point	(Brooks et al., 2005a; Brooks et al., 2012; OSHA, 2017)

^a Lognormal distribution parameters shown are mean, standard deviation.

were modeled using lognormal distributions based on data for *Legionella* spp. provided in Johnson et al. (2017) for nineteen United States reclaimed water systems (Table 7). Some data were generated during a “snapshot” screening using a culture-based assay after which six of the nineteen were sampled quarterly following the initial reconnaissance survey (using three methods, culture, EMA-qPCR, and qPCR). A summary of the six utilities chosen for follow-up sampling is provided in Supplemental Table S1. A total of 153 culture-based samples, 115 EMA-qPCR samples, and 115 qPCR samples were considered. Production at the six plants chosen for follow-up ranged from 14,000 to 75,000,000 gpd and the plants used two disinfectant types, free chlorine and chloramine. Forty-six percent of samples had disinfectant residuals below 1 mg/L and 79% of samples had residuals below 0.2 mg/L. *Legionella* spp. was quantified in the effluent, storage reservoir, and three locations in the distribution system (coded 1, 2, and 3) using three analytical methods: culture, EMA-qPCR, and qPCR. All locations (effluent, storage reservoir, and distribution system locations 1, 2, and 3) were pooled for generating the distributions. Ninety-six percent of *Legionella* spp. detected using the culture-based assay and 52% of *Legionella* spp. gene copies were determined to be *L. pneumophila*. This ratio was used to correct *Legionella* spp. concentrations obtained from each method. The recovery efficiency of the membrane filtration method reported and used in the current study was $70 \pm 18.6\%$ (Mean \pm SD).

3.2. Toilet flushing

3.2.1. Literature review to define toilet flushing parameters

Method 1. An upper bound PC derived by Hines et al. (2014) (1.3×10^{-6} CFU m⁻³/CFU L⁻¹) and lower bound PC calculated from a controlled toilet flushing experiment using Gram negative bacteria *Serratia marcescens* NCTC 10211 were used (Barker and Jones, 2005). Barker and Jones (2005) seeded toilets with 10^{10} bacteria on the toilet sidewalls and flushed 5 min after applying the inoculum. The bulk toilet water contained 10^8 CFU/mL *Serratia* prior to flushing. Bacterial air samples were collected 20 cm above and 30 cm in front of the toilet after flushing and a maximum concentration of 1370 ± 527 CFU/m³ in air was detected 1 min after the

flush resulting in a PC of 1.37×10^{-8} CFU m⁻³/CFU L⁻¹. The aerosol size information was not given and the authors suggest that based on the type of air sampling used, size ranges were likely to be above 20 μ m, indicating the assumption of Darlow and Bale (1959) may have been conservative in equation 1. The calculated PC is lower than previous PC values used in *Legionella* risk assessments and related work: 1) Showering PC based on *Brevundimonas diminuta* of 5.18×10^{-6} – 1.64×10^{-5} CFU m⁻³/CFU L⁻¹ (Schoen and Ashbolt, 2011); 2) Hot springs aerosol PC 2.3×10^{-5} CFU m⁻³/CFU L⁻¹ based on endotoxin data (Armstrong and Haas, 2008); 3) Bursting bubbles in distilled water at 22 °C seeded with *Serratia marcescens* with PC 1×10^{-6} CFU m⁻³/CFU L⁻¹ (Blanchard and Syzdek, 1982); and whirlpool spa PC ranging from $<3 \times 10^{-6}$ (no air injection)– 1.1×10^{-3} CFU m⁻³/CFU L⁻¹ (air injection) based on *Pseudomonas aeruginosa*, which was deemed with the experiment to be a more appropriate surrogate than MS-2 coliphage tested in the study due to its size and similarities with *Legionella* (Moore et al., 2015). A lower generation rate of bacteria-containing aerosol is expected for toilet flushing due to the less active generation process than for showering or aerated hot spring spas. It is assumed the decay in aerosol for toilet flushing is negligible over a 1–5 min exposure event directly after flushing. For toilet flushing exposure frequency, a mean of 5.05 flushes per day was used from a study of 22 municipalities, water utilities, water purveyors, water districts, and water providers (Mayer and DeOreo, 1999) (Table 2).

The deposition efficiency fraction for aerosols of diameter i μ m was derived from Heyder et al. (1986) for a breathing rate of 15 L air/min, an 8 s breathing cycle, and 1 L of tidal volume (Table 3). Ranges were specified using the nasal and oral deposition rates as the upper and lower bound, respectively. The fraction (F_i) of *Legionella* that partitions into various aerosol sizes after transitioning from bulk water to aerosol during a toilet flush was assumed based on an aerosol partitioning dataset from Allegra et al. (2016). Allegra et al. (2016) used a nebulizer to generate aerosols containing *L. pneumophila* and measured the percentage of total bacteria (measured using qPCR) in aerosols of various sizes (Table 3, values extracted from original reference Fig. 4). The distribution of the volume of water inhaled according to Method 1 was lognormally distributed with parameters (–20.9, 1.07), corresponding to a

Table 3
Exposure parameters for toilet flushing scenarios.

Parameter	Symbol	Unit	Value	Distribution	Source
Model 1^a					
Partitioning coefficient	PC	CFU m ⁻³ /CFU L ⁻¹	Min = 1.3×10^{-6} , Max = 1.37×10^{-8}	Uniform	(Barker and Jones, 2005; Hines et al., 2014)
	f_{1-4}	Fraction	0.87	Point	(Darlow and Bale 1959; Hines et al. 2014)
Fraction of total aerosolized <i>Legionella</i> in aerosols of MMAD ^d 1–4 µm	F_{1-4}	%	Min = 6.67, Max = 17.5	Uniform	(Allegra et al. 2016)
	DE_{1-4}	Fraction	Min = 0.23, Max = 0.62	Uniform (Nasal, Oral)	(Heyder et al. 1986)
Model 2					
Concentration of aerosol in air after toilet flush at 420 mm	$C_{aer,2.5}$	# aerosols/cm ³ air	$\mu = -1.246$, $\sigma = 1.885$	Lognormal ^b	(O'Toole et al., 2009)
Model 3					
Concentration of aerosol with MMAD ^c i , where $i = 1:10^d$:	$C_{aer,i}$	# aerosols/m ³ air		Lognormal	(Johnson et al., 2013)
1			$\mu = 10.53$, $\rho = 0.87$		
2			$\mu = 10.43$, $\rho = 0.87$		
3			$\mu = 10.33$, $\rho = 0.89$		
4			$\mu = 10.30$, $\rho = 0.90$		
5			$\mu = 10.31$, $\rho = 0.90$		
6			$\mu = 10.31$, $\rho = 0.89$		
7			$\mu = 10.30$, $\rho = 0.90$		
8			$\mu = 10.30$, $\rho = 0.91$		
9			$\mu = 10.29$, $\rho = 0.91$		
10			$\mu = 10.28$, $\rho = 0.91$		
Models 2 and 3					
Deposition efficiency for aerosols of MMAD ^c i	DE_i	Fraction			
1			Min = 0.23, Max = 0.25	Uniform (Nasal, Oral)	(Heyder et al., 1986)
2			Min = 0.40, Max = 0.53		
3			Min = 0.36, Max = 0.62		
4			Min = 0.29, Max = 0.61		
5			Min = 0.19, Max = 0.52		
6			Min = 0.10, Max = 0.4		
7			Min = 0.06, Max = 0.29		
8			Min = 0.03, Max = 0.19		
9			Min = 0.01, Max = 0.12		
10			Min = 0.01, Max = 0.06		
Fraction of total aerosolized <i>Legionella</i> in aerosols of MMAD ^d i µm	F_i	%		Point	(Allegra et al., 2016)
1			17.50		
2			16.39		
3			15.56		
4			6.67		
5			3.89		
6			2.50		
7			2.78		
8			5.00		
9			5.28		
10			3.89		

^a Toilet models defined in section 2.2.1. Model 1 uses a partitioning coefficient to define *Legionella* transfer to air; Model 2 considers a half-flush toilet; and Model 3 considers multiple toilet types.

^b Lognormal distribution parameters shown are mean, standard deviation.

^c MMAD = mass median aerodynamic diameter.

^d Concentrations of aerosol computed using average and standard deviation parameters across toilet types of Table 1 # Aerosols/m³ * Fraction of aerosols of MMAD i ; All concentrations and efficiencies listed by integer MMAD; bins considered for MMAD 1 through 10 were [0.5,1.5), [1.5,2.5), [2.5, 3.5), [3.5,4.5), [4.5, 5.5), [5.5, 6.5), [6.5, 7.5), [7.5, 8.5), [8.5, 9.5), [9.5, 10.5).

mean of 1.49×10^{-9} L reaching the alveoli.

Method 2. A toilet experiment by O'Toole et al. (2009) used a Caroma Uniset cistern model P/N 213012 and pan model P/N 601200W, operated at full capacity for either 9 L/4.5 L for full/half flush. Aerosols ranging from 0.06 to 20 µm were measured, however, aerosols were only observed in the 2–3 µm size bin and none of the other 3–10 µm bins for a single toilet flush. No aerosols were observable for a half-flush. The deposition efficiency fraction for aerosols of diameter 3 µm was used from Heyder et al. (1986). The distribution of the volume of water inhaled according to Method 2 was lognormally distributed with parameters (−26.3, 1.95), corresponding to a mean of 2.53×10^{-11} L reaching the alveoli.

Method 3. A toilet experiment by Johnson et al. (2013) used four types of toilets including a pre-FEPA gravity flow toilet (13.3 Lpf), a dual-flush high-efficiency toilet (HET) (3.8 or 4.9 Lpf), a dual-flush pressure-assisted gravity flow toilet (PAT) (4.2 or 4.9 Lpf), and a flushometer (FOM) toilet (5.3 Lpf). Data were available for 1–10 µm size mass median aerodynamic diameters (MMAD) and were digitally extracted and averaged across toilet types from Johnson et al. (2013) (Fig. 6 in the Johnson paper), and converted to a fraction of the total particles generated of each size MMAD using $10^9/100$ (Table 4). The fraction was applied to the total generation of particles (#/flush) and sampling volume (m³) averaged over the flush conditions (Table 3). Each size MMAD was corrected for its

corresponding alveolar deposition efficiency. The distribution of the volume of water inhaled according to Method 3 was lognormally distributed with parameters $(-24.4, 0.688)$, corresponding to a mean of 3.21×10^{-11} L reaching the alveoli.

3.2.2. Toilet flushing risk results

Three exposure model methodologies were used to compute the annual risk of infection and severe clinical infection from exposure to reclaimed water during toilet flushing. Annual risks for Method 1 were highest, followed by Method 3 and Method 2. Annual risks for infection (infection endpoint dose response model, “Inf”) and clinical severity infection (CSI) are shown in Fig. 1. Median annual risks of infection and CSI using culture-based *Legionella* concentration data ranged from 2.95×10^{-6} (Method 2) to 3.82×10^{-4} (Method 1) and 3.20×10^{-9} (Method 2) to 4.11×10^{-7} (Method 1), respectively (Fig. 1). Using EMA-qPCR *Legionella* concentration data, median annual risks for infection and clinical severity infection ranged from 2.07×10^{-5} (Method 2) to 2.75×10^{-3} (Method 1) and 2.56×10^{-8} (Method 2) to 3.00×10^{-6} (Method 1), respectively. For qPCR *Legionella* concentration data, median annual risks for infection and clinical severity infection ranged from 8.52×10^{-5} (Method 2) to 1.06×10^{-2} (Method 1) and 1.02×10^{-7} (Method 2) to 1.17×10^{-5} (Method 1), respectively. The 95th percentiles for annual infection risks ranged from 4.08×10^{-4} (Method 2) to 2.72×10^{-2} (Method 1) for culture, 5.69×10^{-3} (Method 2) to 2.59×10^{-1} (Method 1) for EMA-qPCR, and 2.23×10^{-2} (Method 2) to 7.41×10^{-1} (Method 1) for qPCR. The 95th percentiles for annual clinical severity infection risks ranged from 4.60×10^{-7} (Method 2) to 2.58×10^{-5} (Method 1) for culture, 6.40×10^{-6} (Method 2) to 3.19×10^{-4} (Method 1) for EMA-qPCR, and 2.70×10^{-5} (Method 2) to 1.38×10^{-3} (Method 1) for qPCR.

If compared to the USEPA annual infection benchmark of 10^{-4} infections per person per year for drinking water, median annual infection risks (infection dose response endpoint) exceeded this value for Method 1 (culture-based) and Method 3 (qPCR). Using a clinical severity infection dose response endpoint, no calculated models exceeded this value. The 95th percentile annual infection risks exceeded 10^{-4} for all methods. The 95th percentile clinical severity risks exceeded 10^{-4} for Method 1 EMA-qPCR and qPCR.

For all methods, the concentration of *Legionella* in reclaimed water was the most important predictive factor of the final estimate

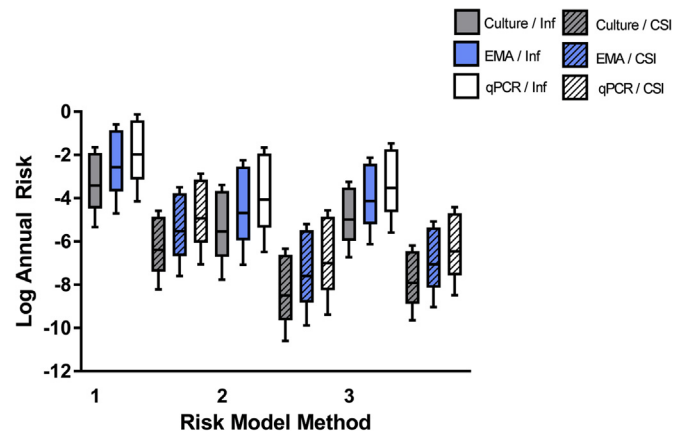


Fig. 1. Log₁₀ annual risks for toilet flushing scenarios for infection (Inf) or Clinical Severity Infection (CSI) animal dose response model endpoints using three different risk model methods (see Section 2.1.1.) and three analytical methods for *Legionella* quantification (culture-based, EMA-qPCR [EMA], and qPCR [qPCR]) in reclaimed water.

of annual risk of either infection or clinical severity infection (Fig. 2) (Spearman rank correlation coefficients ranging from 0.71 to 0.94). The large ranges of annual risks observed are therefore likely due to variability in this factor. For Method 1, the partitioning coefficient was the next most important factor and for Method 2 the concentration of aerosols ($C_{aer,2.5}$) was the next most important factor. For all models, exposure time (t), dose response variable (r), and exposure frequency (f) were also important factors.

3.3. Long range dispersion models

3.3.1. Literature review for spray irrigation parameters

Reclaimed water may be applied through a variety of mechanisms producing varying degrees of aerosols ranging from minimal (drip irrigation) to substantial (spray irrigation). It is assumed here that spray irrigation with reclaimed water would take place via a stationary sprinkler system and could therefore be considered a point-source. Sprinkler heights are all <1 m for commonly used sprinkler systems for reclaimed water (Table 5) (Jjemba et al., 2015), however the plume model was considered to commence from the

Table 4
Aerosol size distribution for modern flush toilets (Johnson et al., 2013).

	Toilet Type					
	PAT, high-volume flush	Pre-FEPA gravity flow	HET, low volume-flush	Flushometer	PAT, low-volume flush	HET, high volume-flush
Liters per flush	4.9	13.3	3.8	5.3	4.2	4.9
Air sampling volume (m3)	0.013	0.01	0.012	0.0108	0.012	0.012
Total droplets produced (SE)	40,521 (1955)	54,363 (6764)	8220 (616)	145,214 (8325)	25,762 (1855)	10,620 (1060)
Aerosols/m ³ = Total droplets/Air Vol.	3.12×10^6	5.44×10^6	6.85×10^5	1.34×10^7	1.98×10^6	8.85×10^5
Median droplet diameter (μm)	Fraction ^a					
1	1.49×10^{-2}	1.37×10^{-2}	1.27×10^{-2}	1.21×10^{-2}	1.31×10^{-2}	1.14×10^{-2}
2	1.29×10^{-2}	1.24×10^{-2}	1.18×10^{-2}	1.09×10^{-2}	1.24×10^{-2}	1.11×10^{-2}
3	1.11×10^{-2}	1.13×10^{-2}	1.07×10^{-2}	1.04×10^{-2}	1.04×10^{-2}	1.07×10^{-2}
4	1.07×10^{-2}	1.11×10^{-2}	9.90×10^{-3}	1.02×10^{-2}	9.89×10^{-3}	1.07×10^{-2}
5	1.06×10^{-2}	1.13×10^{-2}	1.03×10^{-2}	1.04×10^{-2}	1.00×10^{-2}	1.08×10^{-2}
6	1.06×10^{-2}	1.11×10^{-2}	1.08×10^{-2}	1.02×10^{-2}	1.01×10^{-2}	1.09×10^{-2}
7	1.06×10^{-2}	1.09×10^{-2}	1.07×10^{-2}	1.02×10^{-2}	1.02×10^{-2}	1.08×10^{-2}
8	1.05×10^{-2}	1.08×10^{-2}	1.05×10^{-2}	1.04×10^{-2}	1.01×10^{-2}	1.05×10^{-2}
9	1.04×10^{-2}	1.07×10^{-2}	1.04×10^{-2}	1.04×10^{-2}	1.02×10^{-2}	1.04×10^{-2}
10	1.02×10^{-2}	1.05×10^{-2}	1.04×10^{-2}	1.03×10^{-2}	1.01×10^{-2}	1.05×10^{-2}

^a Fraction of total generated aerosols measured in each size bin of MMAD $i = 1:10$. Data were available for 1–10 μm sizes and were digitally extracted and averaged from each toilet type from Johnson et al. (2013) original reference Fig. 6, and converted to a fraction of the total particles generated in each size bin with MMAD i using $10^i/100$.

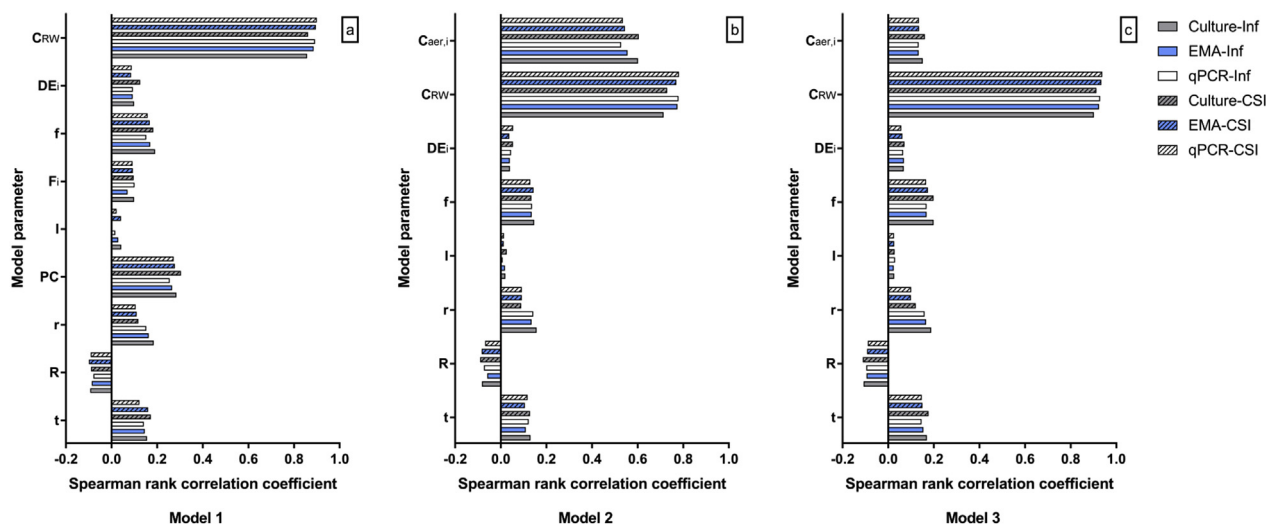


Fig. 2. Sensitivity analysis showing Spearman rank correlation coefficients for toilet risk model methods 1 (panel a), 2 (panel b), and 3 (panel c) for reclaimed water. Coefficients identify the most important predictive factors of annual infection or clinical severity infection risk, where 0 is no influence and -1 or $+1$ when the output is wholly dependent on that input. For visual clarity, only the highest ranking DE_i and $Ca_{er,i}$ parameters are shown.

maximum height of the spray stream. The maximum stream height value of 6 m reported across the commonly used sprinklers in Table 5 was used. The distance required to reach the apex of the sprinkler stream (25 m) was added to estimates for sprinkler setback distances reported in the text. The sprinkler efficiency is the portion of initially sprayed water that leaves the immediate vicinity of the spray irrigation system as aerosols, including aqueous aerosols and evaporated droplets (USEPA, 1982). The efficiency for low pressure smooth-plate sprinklers ranged from 0.5 to 1.4 percent (Kohl, 1974).

Inhalable aerosol size fractions (q_i) were reported by Hardy et al. (2006) for various sprinklers. The largest fractions for Rainbird sprinkler nozzles were chosen (Table 6). The fine mist fraction was evenly divided into ten bins with MMAD $i = 1:10$ to estimate the portion of downstream aerosols $<10 \mu\text{m}$. Decay rates specific to evaporation or aqueous transport were applied separately for these portions of the total downstream aerosol load.

A uniform distribution of residential exposures to spray irrigation activities was determined from estimates derived from Australian guidelines for water reuse ranging from 81 to 99 inhalation exposures per year (Chhipi-Shrestha et al., 2017; NRMCC, 2008). Occupational exposures were assumed to be similar to those estimated in a study of biosolids application, accounting for daily year-round exposures to biosolids for a total of 255 exposures

per year (Brooks et al., 2005a, 2012). This accounts for a 5-day work week with one week absent from exposure. Residential exposures were assumed to occur for 1 h during each exposure event (Brooks et al., 2005a, 2012), while occupational exposures were assumed to occur during a standard 8-h work shift, 255 days per year, as anything over an 8-h shift can be considered an extended or unusual shift (OSHA, 2017).

3.3.2. Literature review for cooling tower parameters

The principal categories for cooling water systems are once-through non-contact cooling, recirculation non-contact cooling, and direct contact cooling (Metcalf and Eddy, 2007). The majority of cooling water systems that use reclaimed water are recirculating non-contact systems (Metcalf and Eddy, 2007). In recirculating non-contact systems, warmed water, from a cooling operation or heat exchanger, is cooled by transferring its heat to air through evaporation in a cooling tower. Warm water from process cooling is sprayed on the top of the internal packing, used to break up the water through spray into droplets to enhance air/water contact. Cool, dry outside air is pulled up through the cooling tower by a large rotating fan to cool the warm water through evaporation. Water is removed by blowdown or purge, and small amounts of water capable of carrying microorganisms are also lost by drift. Typical water loss from drift is assumed to be 0.001–0.005% of the

Table 5
Common sprinkler systems used for reclaimed water irrigation (NA = information not available from manufacturer).

Irrigation sprayer	Type	Usage	Device height (m)	Recommended pressure range (kPa)	Flow rate (L/s)	Spray radius (m)	Max stream height (m)	Distance to max spray height (m)
Rainbird Eagle 900	Closed-case rotor	Golf course fairways	0.09	410–690	1.35 –3.60	19.2–29.6	6.1	18.3–24.4
Rainbird Eagle 700	Gear-driven rotor	Golf course roughs	0.07–0.31	410–690	1.03 –2.76	10.7–22.9	5.2	8.2–19.8
Toro 800 series	Rotor	Golf course	0.15–0.432	200–350	0.03 –0.63	9.7–15.2	NA	NA
Hunter Pro-Spray (spray head)	Rotor (spray or rotating)	Residential areas	0.05–0.3	100–700	0.01 –0.36	2.6–5.8	NA	2.2–4.5
Hunter PGP Rotors 4"	Rotor	Residential areas	0.10 (total device height 0.19)	206–482	0.032 –0.91	6.7–15.9	2.1–4.0 ^a	6.7–12.2 ^a

^a At optimum operating pressures of 50–60 psi; spray radius shown is across all operating pressures (https://www.hunterindustries.com/sites/default/files/PIG_PGP_dom.pdf).

Table 6
Monte Carlo model exposure input parameters.

Parameter	Symbol	Unit	Value	Distribution	Reference/Comments
Flow rate of circulating water	$F_{CT, 10m}$ $F_{CT, 100m}$	L/s	Min = 10^2 , Max = 10^3 Min = 10^3 , Max = 10^4	Uniform	(Adams et al., 1978, 1980; Chen and Hanna, 1978)
Aerosolization efficiency	F_{IR} E_{CT}	L/s %	0.329 Normal operating conditions: Min = 0.001, Max = 0.005 Less effective conditions: Min = 0.1, Max = 0.01	Point Uniform	(Kincaid et al., 1996) (ASHRAE, 2004) (Lucas et al., 2012)
Horizontal distance perpendicular to plume	E_{IR} y	% m	Min = 0.5, Max = 1.4 0; directly downstream along centerline of plume	Uniform Point	(Kohl, 1974) Assumption
Downwind receptor breathing zone height	z	m	1.5	Point	(Paez-Rubio et al., 2007)
Height of cooling tower	H_{CT}	m	Simulated for 10, 100	Point	Assumption
Height of irrigation sprinkler	H_{IR}	m	6	Point	Assumption
Relative humidity	RH	%	Simulated for 65, 80, and 90	Point	Assumption
Decay of <i>Legionella</i> in aqueous aerosol (non-solar)					
RH = 65%	$\lambda_{1,65}$	s^{-1}	Min = 8.40×10^{-5} , Max = 2.38×10^{-4}	Uniform	(Hambleton et al., 1983)
RH = 80%	$\lambda_{1,80}$	s^{-1}	Min = 1.82×10^{-4} , Max = 3.09×10^{-4}	Uniform	(Berendt, 1980; Hambleton et al., 1983)
RH = 90%	$\lambda_{1,90}$	s^{-1}	Min = 7.88×10^{-5} , Max = 4.09×10^{-4}	Uniform	(Dennis and Lee, 1988; Hambleton et al., 1983)
Decay of evaporated <i>Legionella</i> ($t_1 = \text{up to } 30s, t_2 = t - 30s \text{ if } t > 30s$)	$\lambda_{2,t1}$ $\lambda_{2,t2}$	s^{-1}	0.125 3.10×10^{-4}	Point Point	(Katz and Hammel, 1987)
Decay of <i>Legionella</i> in aqueous aerosol and evaporated state (solar)					
RH = 50–60% ^a	$\lambda_{uv,65}$	s^{-1}	$\mu = 1.00 \times 10^{-3}, \rho = 3.42 \times 10^{-3}$	Normal ^c	(Paez-Rubio and Peccia, 2005)
RH = 85–95% ^b	$\lambda_{uv,80}$ and $\lambda_{uv,90}$	s^{-1}	$\mu = 2.17 \times 10^{-3}, \rho = 8.70 \times 10^{-4}$	Normal	(Paez-Rubio and Peccia, 2005)
Deposition efficiency	DE_i	Fraction		Uniform	See Table 1, Models 2 and 3
Fraction of aerosols in respirable range	q (<100 μm , 100–200 μm fractions)	Fraction	0.0138, 0.0413 $0.0459, 6.03 \times 10^{-4}$	Point Point	(Hardy et al., 2006) (Peterson and Lighthart, 1977)
Cooling tower					
Fraction of total aerosolized <i>Legionella</i> in aerosols of MMAD $i \mu m$	F_i	%		Point	See Table 1, Models 2 and 3

^a Average UV-A and UV-B irradiance of 0.065 W/cm².^b Average UV-A and UV-B irradiance of 0.049 W/cm².^c Mean and lognormal parameters shown are mean, standard deviation.**Table 7**
Concentrations distributions for *Legionella* in reclaimed water.

Parameter	Symbol	Unit	Value	Distribution	Reference
Concentration of <i>Legionella</i> spp. in reclaimed water	C_{RW}	CFU/L	$\mu = 8.061, \sigma = 2.219$	Lognormal ^a	(Johnson et al., 2017)
Culture					
EMA-qPCR		Viable gc/L	$\mu = 10.666, \sigma = 2.609$		
qPCR		gc/L	$\mu = 12.051, \sigma = 2.699$		
Portion of <i>Legionella</i> spp. observed that is <i>L. pneumophila</i> for culture method	f_{LP}	%	96 ^b	Point	(Johnson et al., 2017)
Portion of <i>Legionella</i> spp. observed that is <i>L. pneumophila</i> for qPCR and EMA-qPCR methods		%	52	Point	(Johnson et al., 2017)
Recovery efficiency of membrane filtration method	R	%	$\mu = 70, \sigma = 18.6$, truncated between 0 and 100	Normal	(Johnson et al., 2017)

^a Lognormal distribution parameters shown are mean, standard deviation.^b BCYE agar is a selective media for *L. pneumophila*.

total recirculating water (ASHRAE, 2004). However, it is noted that this loss rate could be higher for older designs or certain choices of drift eliminator (up to 0.1%) (Lucas et al., 2012). Both typical (0.003–0.005%) and high drift (0.01–0.1%) conditions were simulated to examine their impact on annual health risks. For cooling towers, the mass-weighted proportions of aerosols <100 and 100–200 μm were calculated by simulating a lognormal distribution specified by Peterson and Lighthart (1977) with a geometric

mean of $230 \pm 1.59 \mu m$ (arithmetic mean \pm SD of $256 \pm 125 \mu m$).

The design and operating conditions of recirculating cooling water towers vary widely (Selby et al., 1996). The flow rate of total recirculating water is a parameter designed (using performance curves specific to a given set of equipment and process being served) to achieve a desired range of thermal capability of the cooling tower, given set of operating conditions (entering water temperature, leaving water temperature, and entering air wet-bulb

temperature) (ASHRAE, 2004). The entering air wet-bulb temperature, required system temperature level, cooling tower size, and number of cells will balance the heat rejected at a specific approach (difference between leaving water temperature and entering air wet-bulb temperature) (ASHRAE, 2004). The cooling tower size is a function of these factors as well as the quantity of water to be cooled, the air velocity through the cell, and the tower height (Zhang et al., 2012). Therefore, it is challenging to designate a set of typical operating conditions for use within a QMRA.

It is assumed for simplicity in this model that larger heat loads necessitate larger towers, which in turn require larger quantities of recirculated water. Several studies report flow rates (per cell) for large (up to 100 m stack height) cooling towers of approximately 10^3 – 10^4 L/s (Adams et al., 1978, 1980; Chen and Hanna, 1978). Cooling towers of heights 10 m and 100 m were simulated with flow rates of 10^2 – 10^3 L/s, and 10^3 – 10^4 L/s, respectively (Table 6).

It was assumed conservatively that operation occurred continuously and therefore could result in potential exposures 365 days per year for residential exposures (Bhopal and Barr, 1990). Residential exposures were assumed to occur for 1 h during each exposure event (Brooks et al., 2005a, 2012), while occupational exposures were assumed to occur during a standard 8-h work shift, 255 days per year (OSHA, 2017).

3.3.3. Decay rates

Several studies have examined *Legionella* decay in aerosol as a function of relative humidity and seeding matrix, as well as bacteria strain, type, and source (Berendt, 1980, 1981; Dennis and Lee, 1988; Hambleton et al., 1983). *Legionella* survival generally increases as the ambient relative humidity increases, and it survives particularly well at intermediate (65%) relative humidity. However, this relationship is not linear, and zones of instability are present. *Legionella* generally survived better in suspensions containing algal extracts compared to tryptose saline. Hambleton et al. (1983) held *L. pneumophila* 74/81 at various relative humidity for 15 min before aerosolizing them in a 3-jet collision nebulizer. The organisms survived best at 65% RH and worst at 55%. Survival was also high at 90% and 80% relative humidity. Therefore, 65%, 80%, and 90% relative humidities were chosen for modeling scenarios. Values were extracted from published graphs and decay constants were obtained by plotting log concentration versus time for two sets of experiments using water spray in Hambleton et al. (Table 6). Two other studies examined *Legionella* survival in aerosolized culture broth (Berendt, 1980; Dennis and Lee, 1988) and were used as lower bounds on the decay estimates at each humidity. Only one study examined the decay of dried *Legionella* for use with the evaporated mass fraction (Katz and Hammel, 1987). *Legionella pneumophila* Philadelphia 1 strain was dried for 90 min. A four-log drop in viability was observed during the first 30 s, followed by a more gradual decline. Biphasic decay values were derived by converting percent recovery to a concentration and plotting log concentration versus time. The higher decay rate was applied for up to the first 30 s after the average total evaporation time, while the lower decay rate was applied from $t=30$ s to downstream time t where applicable.

No study was available that simultaneously examined microbial decay impacts of RH and solar exposure on *Legionella*. A study by Paez-Rubio and Peccia (2005) allowed for parsing out of decay attributable to solar exposure versus other factors in aerosol at two RH ranges for *E. coli*, another gram-negative bacteria. The solar-induced inactivation rate was used for both moderate RH of 50–60% with mean \pm SE inactivation of $1.00 \times 10^{-3} \pm 3.42 \times 10^{-3} \text{ s}^{-1}$ ($n=5$), and high RH of 85–95% with mean \pm SE inactivation of $2.17 \times 10^{-3} \pm 8.70 \times 10^{-4} \text{ s}^{-1}$ ($n=5$). These decay rates were applied in addition to the non-solar decay rates at each RH, where

the “moderate” RH was applied with the 65% RH non-solar decay, and the “high” RH was applied with the 80% and 90% RH non-solar decay. Data were not available for the impact of solar inactivation on *Legionella* in droplets that evaporated; therefore, these decay fractions were applied to both the aqueous and evaporated fractions. Solar decay was assumed at every time t because exposure times were assumed to happen when people were present during daylight hours.

3.4. Cooling tower risk results

Annual health risks from exposure to aerosols from cooling towers were modeled for 4 Pasquill stability classes (A through D, corresponding to wind speeds ranging from 1 to 7 m/s), 3 humidity values with corresponding solar decay values (65, 80, and 90%), 2 stack heights (10 m and 100 m), 2 dose response endpoints (infection, clinical severity infection), 3 methods (culture, EMA-qPCR and qPCR), and 2 exposure durations/frequencies (residential, occupational) at various downwind distances from 50 to 10,000 m. A comparison of annual infection risks for residential populations with various combinations of wind speed and relative humidity parameters for culturable *Legionella* is shown in Supplemental Fig. S1; changing meteorological conditions did not have as important of an impact on risk as the other modeled factors. Generally, as wind speed increases, aerosols are carried farther, and annual risks peak farther away from the cooling tower. Although *Legionella* is more stable at 65% and 90% relative humidity than at 80%, this does not have as great an impact on annual risk of infection as changing the wind speed. *Legionella* is carried farthest at the highest wind speed (7 m/s) and a relative humidity of 65%. Using this (most conservative) set of meteorological parameters, a comparison of annual risks for infection and clinical severity infection at two stack heights and all three analytical methods are shown for residential and occupational populations in Figs. 3–6. Peak risks occur downwind from the source as some time must pass for the plume to reach human breathing height. Annual risks from qPCR are highest, followed by EMA-qPCR and culture-based assays. Annual clinical severity infection risks are up to 2.5 orders of magnitude lower than annual infection risks at a given downwind distance. Occupational risks were up to one log higher than residential risks. In addition, both types of annual infection risks peak further downstream for stack heights of 100 m compared to 10 m.

If the USEPA 10^{-4} annual infection risk target for drinking water is used for comparison to 95th percentiles from the cooling tower annual infection risk (infection dose response endpoint) distribution, the setback distance for both residential and occupational populations with a 10 m or 100 m tall cooling tower would be $\geq 5,000$ m with no other risk mitigation actions (Figs. 3–6). For a median comparison point, distances for a 10 m cooling tower would range from 500 m (culturable) to ~3,500 m (qPCR). Using a clinical-severity infection dose response endpoint and stack height of 10 m, setback distances for residential populations would cover a large range depending on the concentration detection method and percentile for comparison used. Using a 95th percentile for comparison, the setback distance would be less than 50 m for culturable, ~500 m for EMA-qPCR, or ~1000 m for qPCR (Fig. 3). For occupational populations, these corresponding distances would be <50 m, ~1000 m and >2500 m, respectively (Fig. 5). If median annual clinical severity risks for a stack height of 10 m are compared to the 10^{-4} benchmark, no residential or occupational models exceed this value.

The results of the sensitivity analysis for cooling towers are shown in Fig. 7. Regardless of the detection method used, the concentration of *Legionella* in reclaimed water was the most important predictive factor of the final estimate of annual infection

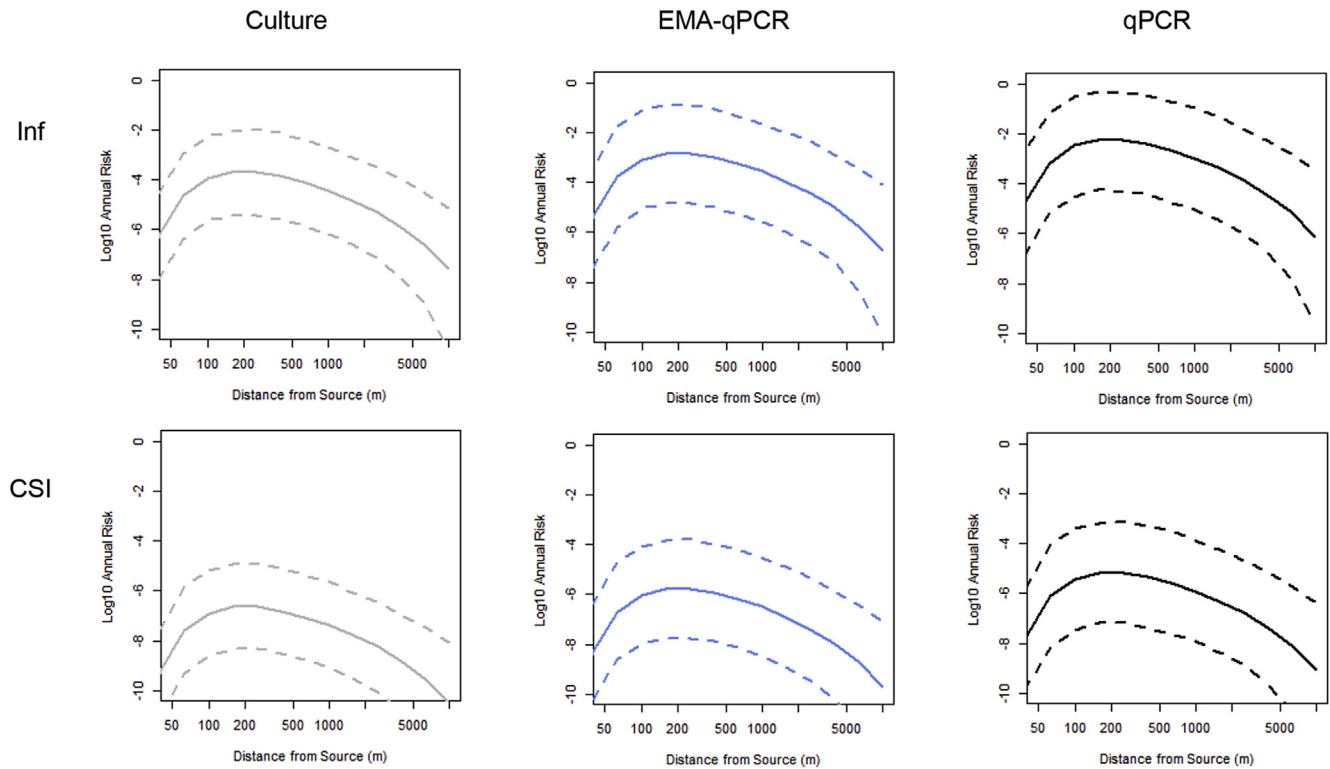


Fig. 3. Log₁₀ annual infection risks for *L. pneumophila* in residential populations due to cooling tower aerosol exposure (efficiency = 0.001–0.005%, stack height = 10 m) at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

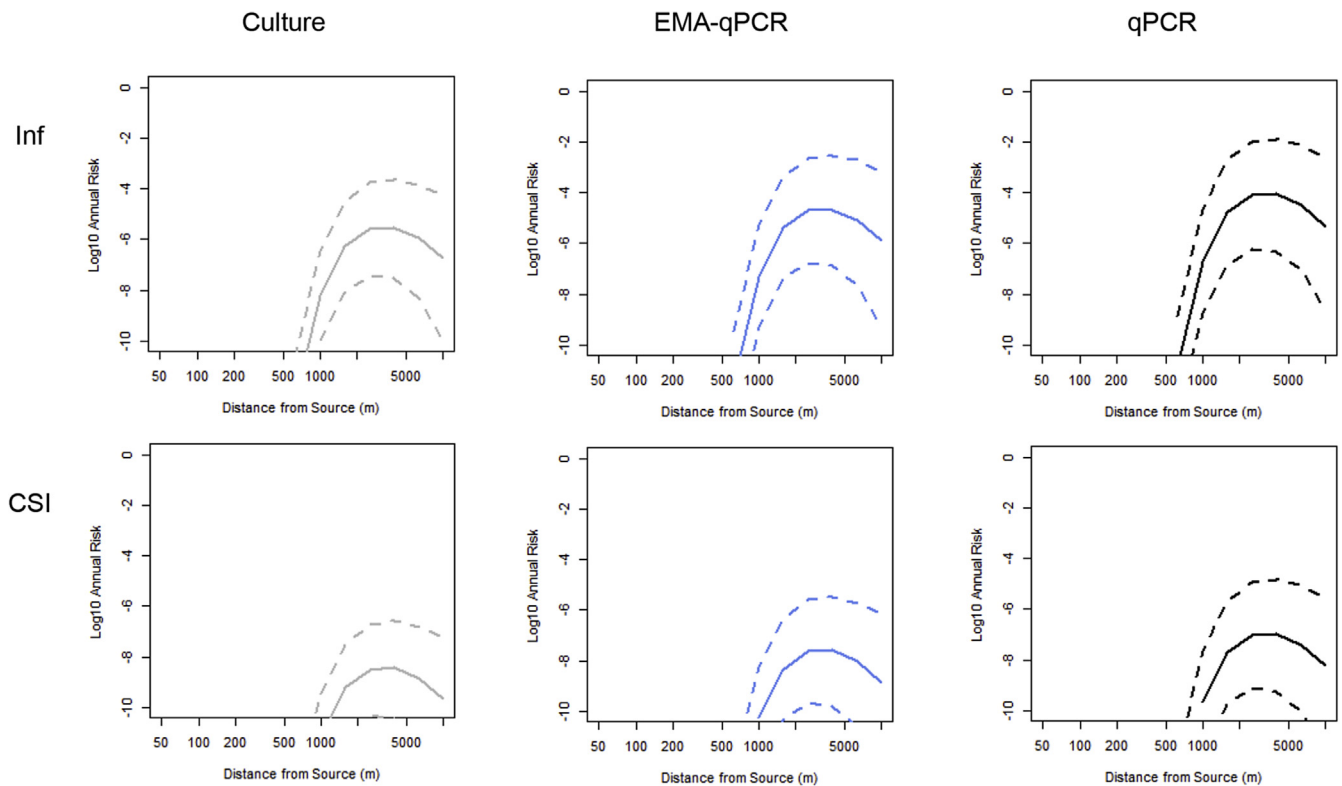


Fig. 4. Log₁₀ annual infection risks for *L. pneumophila* in residential populations due to cooling tower aerosol exposure (efficiency = 0.001–0.005%, stack height = 100 m) at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

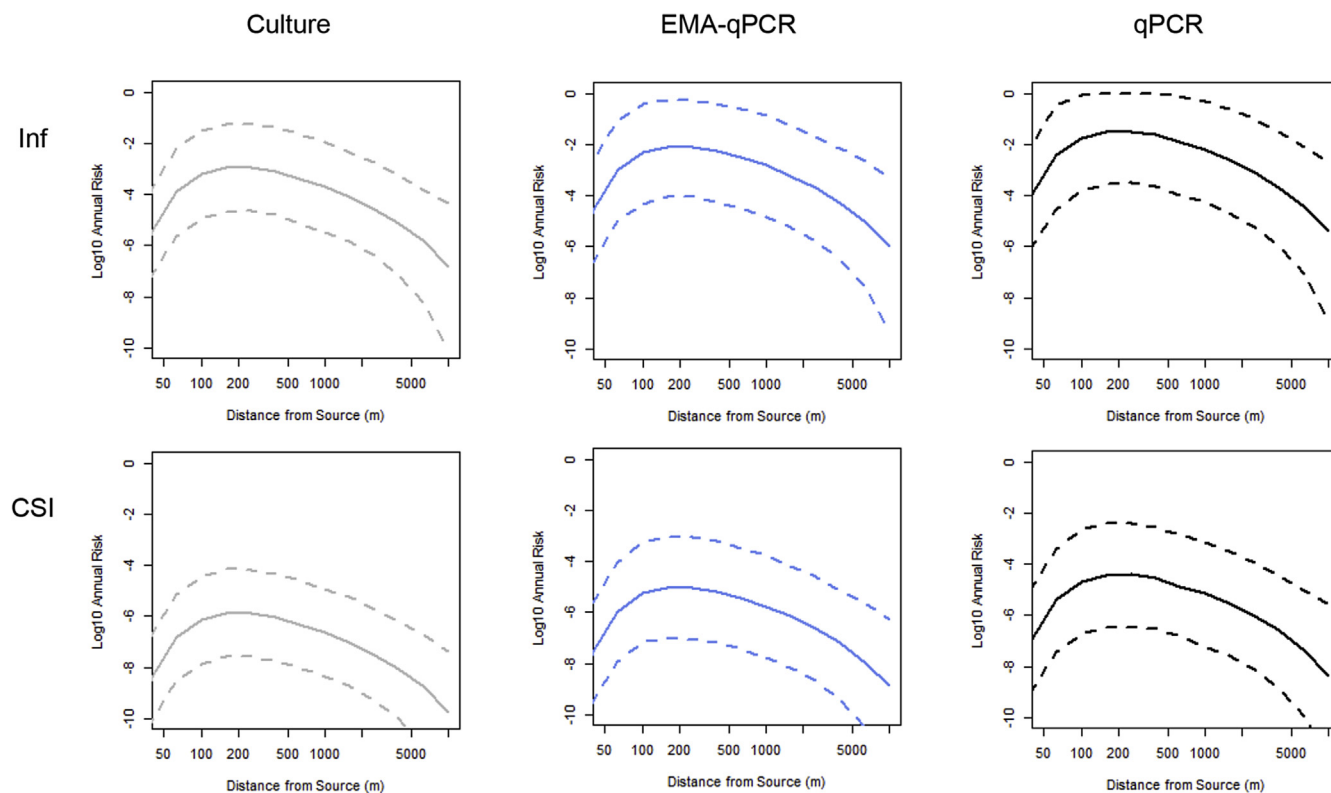


Fig. 5. Log₁₀ annual infection risks for *L. pneumophila* in occupational populations due to cooling tower aerosol exposure (efficiency = 0.001–0.005%, stack height = 10 m) at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

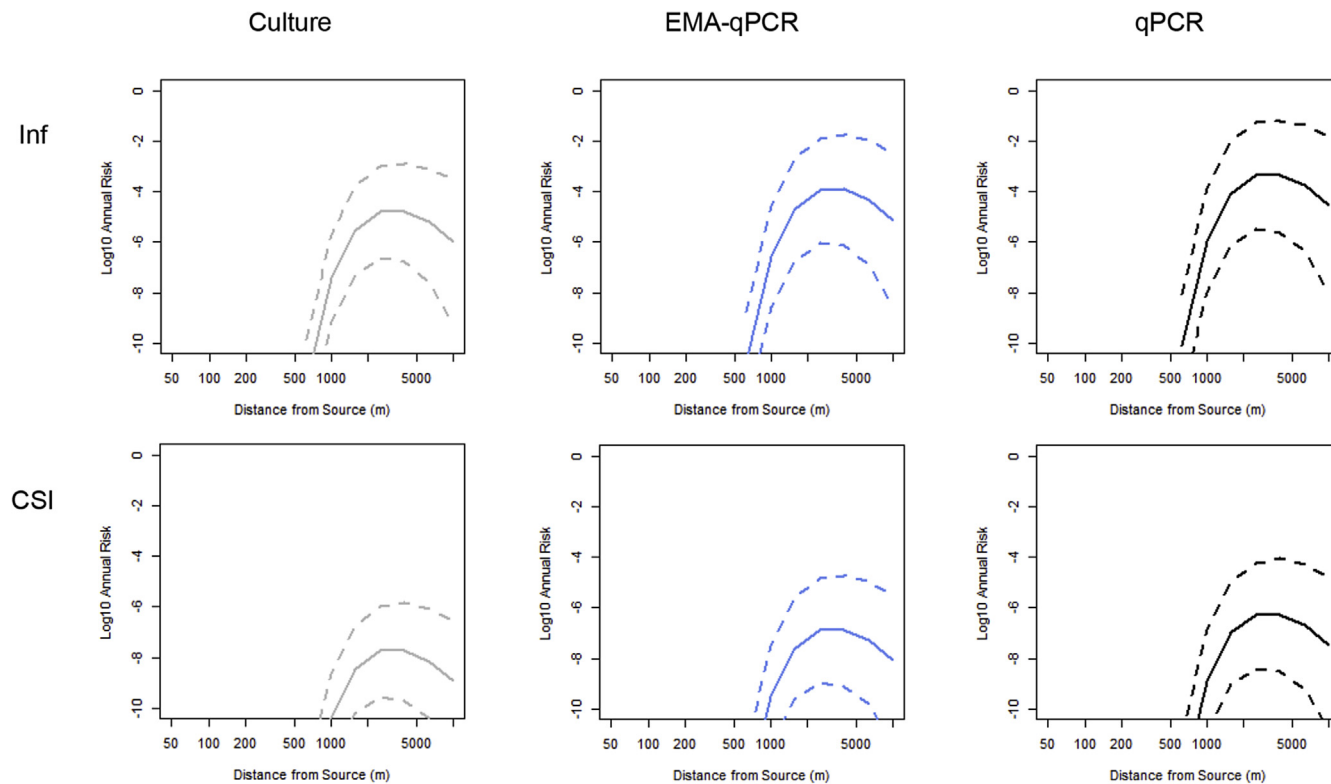


Fig. 6. Log₁₀ annual infection risks for *L. pneumophila* in occupational populations due to cooling tower aerosol exposure (efficiency = 0.001–0.005%, stack height = 100 m) at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid black line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

or clinical severity infection risk (Spearman rank correlation coefficient ranging from 0.81 to 0.89). The cooling tower circulating water flow rate, dose response parameter, and cooling tower drift efficiency were the next most influential factors. Owing to variation in the efficiency of various drift eliminators, a comparison of “typical” operating conditions of 0.001–0.005% and “less effective drift eliminator” conditions which might be typical of an older (pre-1970’s) cooling tower of 0.01–0.1% are presented in Supplemental Fig. S2. A higher efficiency drift eliminator can decrease risks by 1–1.5 logs, highlighting this factor as an important potential management strategy.

3.5. Spray irrigation risk results

A comparison of annual infection and clinical severity infection

risks for the most conservative set of meteorological conditions is shown in Figs. 8 and 9. For sprinklers, a stack height of 6 m was used, corresponding to the highest point of the sprinkler spray arc. For this reason, Figs. 8–9 should be interpreted as distance downwind from the horizontal distance at which the maximum height of the arc occurs. This distance has a maximum length of 24.4 m (Table 5) and is included in setback distance estimates below. Generally, sprinkler risks were lower than cooling tower risks.

For annual infection residential population risks, setback distances would range from ~1,025 m (culture-based assay) to > 10,000 m (qPCR-based assay) from the sprinkler under conservative meteorological conditions (conditions that promote *Legionella* dispersion of 7 m/s windspeed and 65% RH) and using a 95th percentile for comparison, if no other risk mitigation

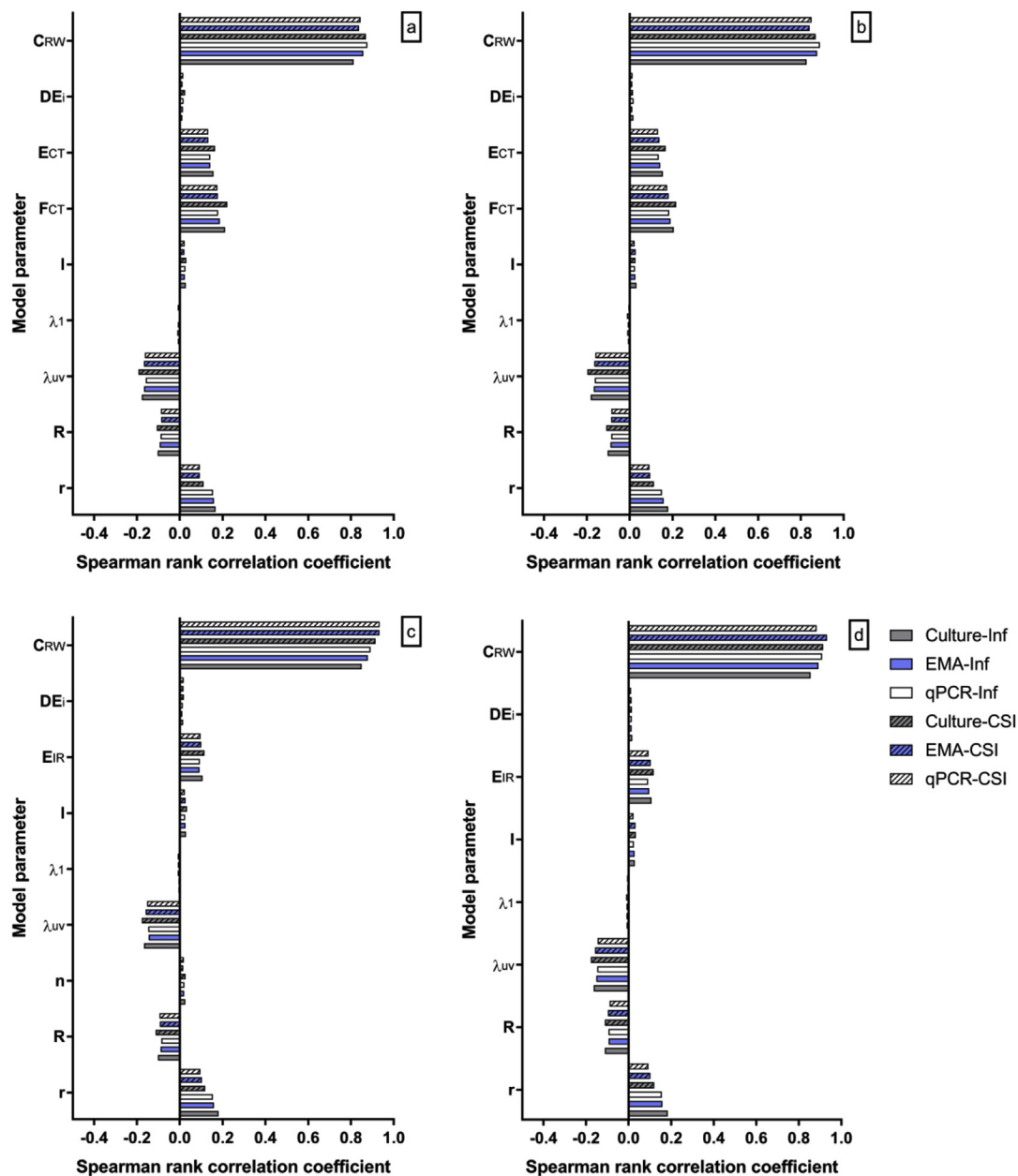


Fig. 7. Sensitivity analysis for long range dispersion models. Cooling towers are shown in panel (a) for 10 m stack height and “typical conditions” drift eliminator efficiency, wind speed = 7 m/s, RH = 65%, with residential or (b) occupational exposure; spray irrigation is shown in panel (c) for wind speed = 7 m/s, RH = 65% residential and (d) occupational exposure. For visual clarity, only the highest ranking DE_i parameter is shown for each model. Values for each parameter averaged for all x distances.

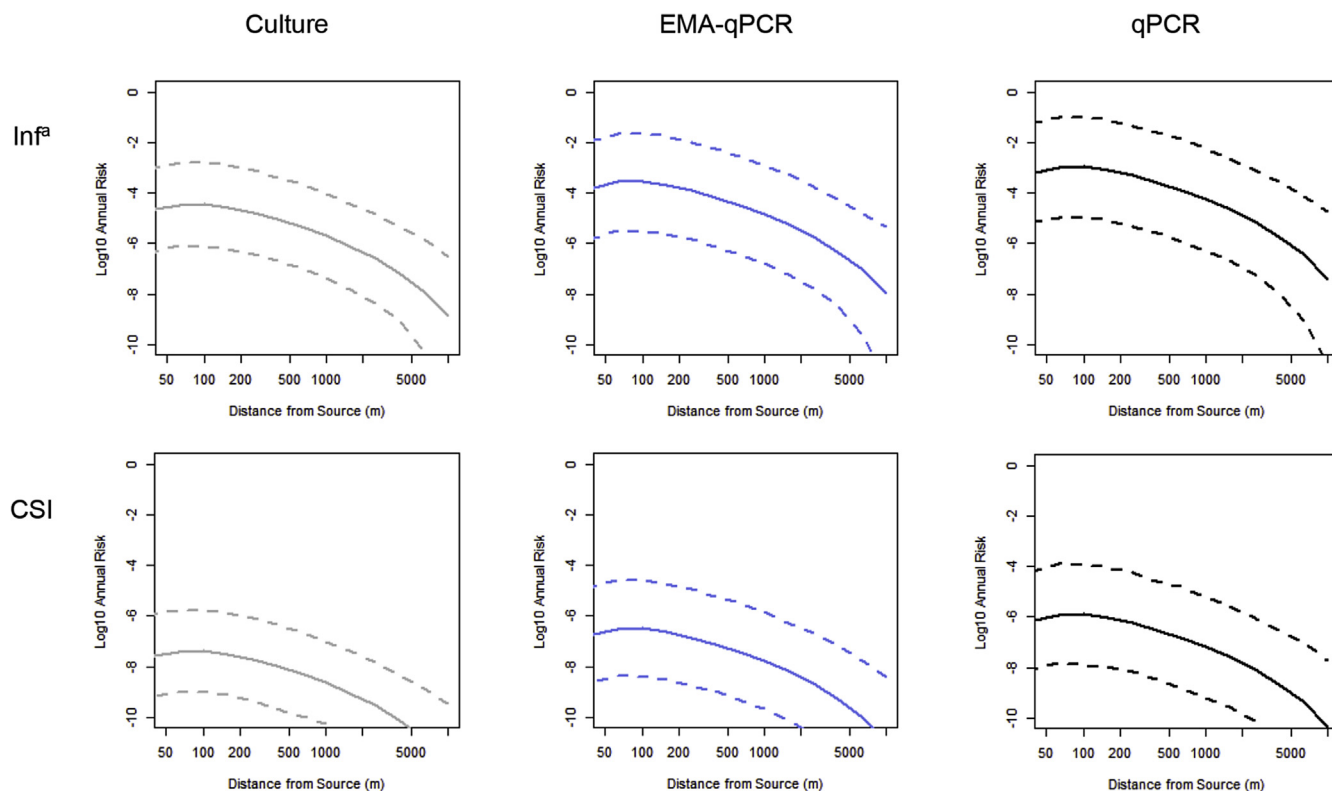


Fig. 8. Log₁₀ annual infection risks for *L. pneumophila* in residential populations due to sprinkler exposure at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

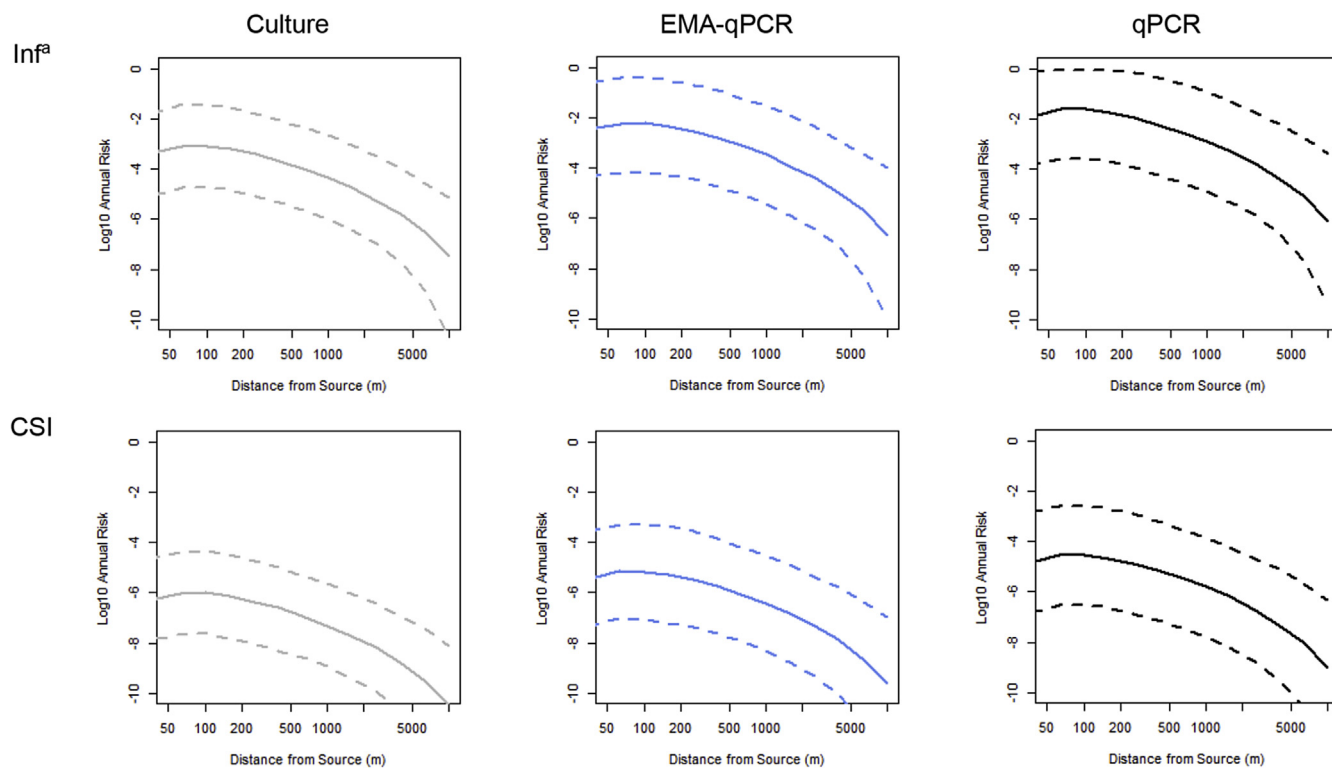


Fig. 9. Log₁₀ annual infection risks for *L. pneumophila* in occupational populations due to sprinkler exposure at varying downwind distances for wind speed = 7 m/s and relative humidity = 65%. The median (solid line) and 95% confidence interval (dotted lines) are shown. ^aInfection (Inf) or clinical severity infection (CSI) dose response model endpoints.

strategies are applied. These distances would be between <75 m (culturable) and ~625 m (qPCR-based) using median values for comparison. 95th percentile annual clinical severity residential population risks were <10⁻⁴ for culture-based and EMA-qPCR methods, but not for qPCR (corresponding to ~225 m setback distance). Occupational annual infection risks indicate a setback distance ~5,025 m (culture-based) or ≥ 10,000 m (EMA-qPCR or qPCR-based) and occupational clinical severity risks correspond to distances ranging from <75 m (culture-based) to ~1,225 m (qPCR-based) using a 95th percentile comparison and <75 m using a median comparison.

The sprinkler sensitivity analysis shown in Fig. 7 also highlights the concentration of *Legionella* in reclaimed water as the primary factor that influences annual infection estimates. The sprinkler flow rate and aerosol efficiency were the next most influential parameters and the dose response parameter was also an important factor.

4. Discussion

A QMRA is presented here for *Legionella* infection risks during toilet flushing, spray irrigation, and cooling tower mist inhalation with reclaimed water. Three toilet flushing exposure models were compared using different analytical methods and dose response endpoints. A modified Gaussian Plume dispersion model was used to compare risks for different analytical methods, meteorological conditions, exposure types, dose response endpoints, and downwind distances for sprinkler and cooling tower risks. Two operation conditions for informing cooling tower risk management were explored. These options were varying the stack height and aerosolization efficiency for cooling towers.

Moderate differences in annual infection and clinical severity risks were observed across the three methods (culturable, EMA-qPCR, qPCR) used to detect *Legionella* for each scenario. This resulted in 1–2 orders of magnitude differences in median annual infection or clinical severity infection risks across models. However, risk estimates for EMA-qPCR and qPCR had higher values than risk estimates using the culture-based data. This difference matters when choosing between percentiles to compare to the target or benchmark value in order to derive a setback distance, for example. Large differences (up to 3 orders of magnitude) across scenarios were observed when using an infection versus a clinical severity infection dose response model, with the infection model resulting in higher risks. Lesser differences were observed in the cooling tower and sprinkler models at any downwind distance due to residential versus occupational exposure (1–2 order difference) or raising the stack height (up to 2 orders of magnitude difference, with peak risks occurring further downwind). Changes in meteorological parameters varied between models based on downwind distances, with high (7 m/s) wind speed and 65% relative humidity resulting in the highest risk estimates. These findings indicate that differences between risk scenarios (versus the sensitivity analyses that identify important factors within a single scenario) are most largely influenced by the selection of a dose response model (infection or clinical severity infection), followed by the exposure model used (toilet flushing only), exposure duration (cooling tower and sprinkler only; residential or occupational), and method (culture-based, EMA-qPCR, or qPCR) used to quantify *Legionella* in the water sources.

Annual infection or clinical severity infection risks greater than a tentative 10⁻⁴ annual infection benchmark value for drinking water were observed for certain cases in all three exposure scenarios, depending on the conditions. For toilet flushing, Method 2 was the least conservative (producing lower risk estimates overall) and Method 1 was the most conservative (resulting in higher risk

estimates overall). This is likely due to the use of a PC in Method 1 which measured the ratio of bacterial concentrations in air and water; this indicates there is some discrepancy when comparing results computed using a PC versus computing bacterial concentrations in each size range based on an aerosol size distribution. Both approaches require assumptions to be made when assessing how *Legionella* is partitioned in aerosol; more experimental measurements are needed to validate these approaches to determine the relative strengths of each approach. The risks associated with toilet flushing may be somewhat mitigated by recommending reclaimed water users put the lid down prior to flushing, however, some studies noted that closing the toilet lid did not have a substantial effect on mitigating the spread of aerosols (Barker and Jones, 2005; Bound and Atkinson, 1966).

The cooling tower and sprinkler models indicate that *Legionella*-containing aerosols can be carried long distances in sufficient quantities to present health risks above 10⁻⁴ annual probability of infection or clinical severity infection. These findings are consistent with previous studies that have predicted long-range transport of *Legionella* and observed distances between outbreak cases and implicated cooling towers up to 12 km away (Borgen et al., 2008; Nygård et al., 2008; Rouil et al., 2004; Walser et al., 2014). However, these large spreads were likely attributed to hot weather and high humidity, or rare events such as thermal inversions (Chan and Iseman, 2013; Fisman et al., 2005). In many cases, outbreaks were also associated with inadequate maintenance of cooling tower systems such as lack of regular inspection, faulty dosing pumps, suboptimal disinfection, high pressure cleaning, intermittent operation modes, and restarting of cooling towers (Walser et al., 2014).

Depending on the model conditions selected, the setback distances associated with a 10⁻⁴ annual risk could be quite large. The setback distance was highly sensitive to the *Legionella* detection method used. As a result, additional risk mitigation strategies are likely to be warranted to decrease the setback distance needed. Interventions such as windbreaks using trees or walls around irrigation areas could also reduce risks. Information is not currently available regarding the degree to which microorganisms are removed due to these interventions.

Risks from cooling towers can be reduced by utilizing towers built with a lower stack height and efficient drift eliminators (Lucas et al., 2012). The simulations performed here demonstrate that a less-effective drift system can increase risks 1–1.5 logs. Although higher stack heights have slightly lower annual risks than lower stack heights throughout the zone of influence, higher stack heights result in farther transport of aerosols, and therefore could result in higher setback distances in some cases. A lower stack height reduces the distance aerosols can travel. An effective stack height for cooling towers would be higher than the actual stack height used for simulation here due to the effects of plume rise, which occurs because the plume is hotter than the surrounding air and rises buoyantly as it exits the stack with a vertical velocity (Thomson et al., 2013a). In the current simulations, plume rise was not considered. In order to calculate plume rise, it is necessary to obtain specific information regarding the stack height exit velocity, stack diameter, and temperature of exiting water vapor. Obtaining such specific information about the cooling tower was beyond the scope of the current simulations. Additional guidance documents suggest that stagnant water, nutrient growth including the presence of biofilms, poor overall microbiological water quality, cooling tower deficiencies, inadequate maintenance, poor design or a location of the system that results in large exposures and/or exposures to immune-compromised populations are risk factors for *Legionella* outbreaks (ASHRAE, 2015; CDC, 2016; Department of Health and Human Services, 2015; Sharville et al., 2017).

Measures such as treatment before initial startup following commissioning or any extended shutdown period; periodic inspection and monitoring; restriction of access to the cooling tower; use of drift eliminators; “bleed-off” to prevent solids accumulation; protection from sunlight; training cooling tower employees in health and safety practices; use of hazard analysis and critical control points (HACCP) methodology and/or documenting all strategies for planning, monitoring, controlling, and responding to issues that arise, conducting independent audits; and installing automatic biocide dosing devices can mitigate some of these risks (ASHRAE, 2015; CDC, 2016; Department of Health and Human Services, 2015; Sharvelle et al., 2017).

Generally, sprinkler risks were lower than cooling tower risks, most likely due to the smaller fraction of fine aerosols generated, lower dispersion height, and lower water flow rate. For the low pressure, low profile sprinklers modeled, annual clinical severity infection risks were $<10^{-4}$ for most scenarios with setback distances <75 m. If considering annual infection risks, setback distances could increase substantially beyond 75 m. The sprinkler simulation was initiated at the apex of the spray arc. As such, the velocity propelling droplets in the x direction was not considered, which may cause larger droplets to be propelled further distances. This factor could result in actual dispersion distances being greater than those used in the QMRA and therefore larger required setback distances.

In addition to using low-profile and low pressure sprinkler irrigation systems like the ones specified in this report, nozzles with larger orifices will reduce the formation of fine mist. Subsurface or drip irrigation would minimize drift formation, but can incur higher initial investment costs (Thomson et al., 2013b). Although increasing the droplet size distribution for sprinklers mitigates *Legionella* risk, reliance on larger droplets may increase erosion risk for fragile soils due to the greater kinetic energies associated with larger droplets (Montero et al., 2003).

Cooling towers generally must operate when process cooling is needed, but sprinkler application can be scheduled during periods of low wind velocity or directed away from areas such as hospitals where sensitive populations are located. In addition, operations can be scheduled during nighttime hours or off-hours when employees are away from the irrigation zone. Although not addressed in the models, conditions of high-humidity and lower temperature will reduce evaporative loss of droplets and prevent some of the size decrease that results in drift droplets reaching a respirable range before they settle. Solar decay of bacteria in aerosol is also greater at higher humidity (Paez-Rubio and Peccia, 2005). Although decay for evaporated *Legionella* is greater than for *Legionella* in aqueous aerosol, this factor had minimal influence in the model. Furthermore, it is not known how viability or infectivity changes during long-range aerosol transport.

While all three laboratory methods (culture, EMA-qPCR, qPCR) are currently in use for *Legionella* monitoring, there is no consensus on which method is most appropriate. Generally, qPCR will detect *Legionella* more frequently than culture-based methods (Whiley and Taylor, 2014). Culture-based methods are considered the “gold standard” and will detect viable *Legionella*, but will underestimate viable but non-culturable (VBNC) *Legionella*, while qPCR data will enumerate non-viable cells (Collins et al., 2015). Furthermore, qPCR results are not always correlated with culture-based methods (Whiley and Taylor, 2014). While EMA-qPCR produces estimates of *Legionella* cells with intact membranes that can be a good indication of cell viability, some authors argue the use of EMA-qPCR may not be appropriate for testing for regulatory purposes for biofilms, or when high levels of background bacteria are present (Taylor et al., 2014). EMA-based methods will need to be improved in terms of reliability and robustness in order to increase

their routine use, however, there are algorithms available to aid in their interpretation and comparison with qPCR-based data (Ditomaso et al., 2015).

The most important factor identified in the sensitivity analysis for nearly all models was the concentration of *Legionella* in the reclaimed water. This analysis did not account for dilution of reclaimed water with other types of water. Typically, reclaimed water can have up to 20% v/v dilution (Dungan, 2014). Dilution with water of higher microbiological quality could provide significant water savings while still reducing the need for additional risk management options. Additionally, changes in reclaimed water quality within a cooling tower or premise plumbing were not considered. *Legionella* could grow in these systems and result in potentially higher risks, although the net effect during premise plumbing under specific conditions is a research gap.

Another important parameter identified in the sensitivity analysis is the aerosolization efficiency or fraction that ultimately is in the respirable range by the time it reaches a downwind receptor. Determining the aerosol size distribution and downwind proportion of *Legionella*-containing aerosols in the respirable range remains a substantial challenge for QMRA models. It is challenging to model the evolution of aerosol size distributions over time due to co-occurring and interrelated dynamic rate physical phenomena of settling, evaporation, condensation, coalescence, and secondary aerosol formation due to bubble burst and film collapse (Hinds, 1999; Lighthart et al., 1991). The fraction of aerosols in the respirable range is therefore not likely to remain constant over time and the current model may underestimate the impact of varying meteorological parameters as a constant fraction of respirable droplets over downwind distances is assumed. This approach has been applied in other *Legionella* QMRA models that considered aerosol size distributions (Armstrong and Haas, 2007b; Nygård et al., 2008), however, a model that considers these factors is therefore recommended for further development to more accurately determine *Legionella* risks from systems with the potential for large scale dispersion. Finally, risks were not computed using disability adjusted life years (DALY), which are useful for comparing health burdens from various infection scenarios; however, although DALY measures are available for the Netherlands (van Lier et al., 2016), standard DALY values for Legionnaires Disease in US-based populations are not currently available and are recommended for further development to aid in interpretation of risk values.

5. Conclusions

- *Legionella* median annual infection risks and annual clinical severity infection risks for toilet flushing can exceed a 10^{-4} annual risk of infection benchmark for some aerosol exposure estimation methods and the 95th percentile risk exceeded the benchmark for all aerosol exposure estimation methods.
- *Legionella* annual infection risks and annual clinical severity infection risks are non-trivial at potentially large distances away from cooling towers and sprinklers operating under typical conditions.
- Ranked according to their influence on annual risk estimates, the dose response model chosen (infection or clinical severity infection), the population at risk (residential or occupational), the detection method (culture-based, EMA-qPCR, or qPCR), operating conditions (drift eliminator performance or stack height for cooling towers only), and meteorological conditions (cooling towers and sprinklers) were the most important factors identified in the scenario analyses.
- The concentration of *Legionella* present in reclaimed water was the most influential parameter within all individual risk

simulations, highlighting the importance of efforts to control occurrence for managing risks.

- Management practices such as closing toilet lids, using more efficient drift eliminators for cooling towers, or using wind breaks for cooling towers and sprinklers could contribute to public health risk mitigation.

Acknowledgements

This work was supported by grant WRF12-05 “Development of a Risk Management Strategy for *Legionella* in recycled water systems”.

Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.watres.2017.12.022>.

References

- Adams, A., Garbett, M., Rees, H., Lewis, B., 1978. Bacterial aerosols from cooling towers. *J. Water Pollut. Control Feder.* 2362–2369.
- Adams, A., Garbett, M., Rees, H., Lewis, B., 1980. Bacterial aerosols produced from a cooling tower using wastewater effluent as makeup water. *J. Water Pollut. Control Feder.* 498–501.
- Ahmadrajabi, R., Shakibaie, M.R., Iranmanesh, Z., Mollaei, H.R., Sobhanipoor, M.H., 2016. Prevalence of mip virulence gene and PCR-base sequence typing of *Legionella pneumophila* from cooling water systems of two cities in Iran. *Virulence* 7 (5), 602–609.
- Allegra, S., Leclerc, L., Massard, P.A., Girardot, F., Riffard, S., Pourchez, J., 2016. Characterization of aerosols containing *Legionella* generated upon nebulization. *Sci. Rep.* 6.
- Allestam, G., de Jong, B., Långmark, J., 2006. *Legionella*. American Society of Microbiology, pp. 493–496.
- Amemura-Maekawa, J., Kikukawa, K., Helbig, J.H., Kaneko, S., Suzuki-Hashimoto, A., Furuhashi, K., Chang, B., Murai, M., Ichinose, M., Ohnishi, M., 2012. Distribution of monoclonal antibody subgroups and sequence-based types among *Legionella pneumophila* serogroup 1 isolates derived from cooling tower water, bath water and soil in Japan. *Appl. Environ. Microbiol. AEM*, 06869–06811.
- Aoki, K., Memon, M., Mabuchi, H., 2005. Water and Wastewater Reuse: an Environmentally Sound Approach for Sustainable Urban Water Management. United Nations Environmental Program.
- Armstrong, T., Haas, C.N., 2007a. A quantitative microbial risk assessment model for Legionnaires' Disease: animal model selection and dose-response modeling. *Risk Anal.* 27 (6), 1581–1596.
- Armstrong, T.W., Haas, C.N., 2007b. Quantitative microbial risk assessment model for Legionnaires' disease: assessment of human exposures for selected spa outbreaks. *J. Occup. Environ. Hyg.* 4 (8), 634–646.
- Armstrong, T.W., Haas, C.N., 2008. Legionnaires' disease: evaluation of a quantitative microbial risk assessment model. *J. Water Health* 6 (2), 149–166.
- ASHRAE, 2004. HVAC Systems and Equipment, Atlanta, GA.
- ASHRAE, 2015. Legionellosis: Risk Management for Building Water Systems.
- Azuma, K., Uchiyama, I., Okumura, J., 2013. Assessing the risk of Legionnaires' disease: the inhalation exposure model and the estimated risk in residential bathrooms. *Regul. Toxicol. Pharmacol.* 65 (1), 1–6.
- Barker, J., Jones, M., 2005. The potential spread of infection caused by aerosol contamination of surfaces after flushing a domestic toilet. *J. Appl. Microbiol.* 99 (2), 339–347.
- Baron, P.A., Willeke, K., 1986. Respirable droplets from whirlpools: measurements of size distribution and estimation of disease potential. *Environ. Res.* 39 (1), 8–18.
- Beer, K.D., Gargano, J.W., Roberts, V.A., Hill, V.R., Garrison, L.E., Kutty, P.K., Hilborn, E.D., Wade, T.J., Fullerton, K.E., Yoder, J.S., 2015. Surveillance for waterborne disease outbreaks associated with drinking water—United States, 2011–2012. *MMWR Morb. Mortal. Wkly. Rep.* 64, 842–848.
- Berendt, R., 1980. Survival of *Legionella pneumophila* in aerosols: effect of relative humidity. *J. Infect. Dis.* 141 (5), 689–689.
- Berendt, R., 1981. Influence of blue-green algae (cyanobacteria) on survival of *Legionella pneumophila* in aerosols. *Infect. Immun.* 32 (2), 690–692.
- Bhopal, R., Barr, G., 1990. Maintenance of cooling towers following two outbreaks of Legionnaires' disease in a city. *Epidemiol. Infect.* 104 (1), 29–38.
- Blanchard, D.C., Syzdek, L.D., 1982. Water-to-air transfer and enrichment of bacteria in drops from bursting bubbles. *Appl. Environ. Microbiol.* 43 (5), 1001–1005.
- Blatny, J.M., Ho, J., Skogan, G., Fykse, E.M., Aarskaug, T., Waagen, V., 2011. Airborne *Legionella* bacteria from pulp waste treatment plant: aerosol particles characterized as aggregates and their potential hazard. *Aerobiologia* 27 (2), 147–162.
- Blatny, J.M., Reif, B.A.P., Skogan, G., Andreassen, O., Høiby, E.A., Ask, E., Waagen, V., Aanonsen, D., Aaberge, I.S., Caugant, D.A., 2008. Tracking airborne *Legionella* and *Legionella pneumophila* at a biological treatment plant. *Environ. Sci. Technol.* 42 (19), 7360–7367.
- Borgen, K., Aaberge, L., Werner-Johansen, O., Gjosund, K., Storsrud, B., Haugsten, S., Nygard, K., Krogh, T., Høiby, E., Caugant, D., Kanestrom, A., Simonsen, O., Blystad, H., 2008. Cluster of Legionnaires disease linked to an industrial plant in southeast Norway, June – July 2008. *Euro Surveill.* 13 (38).
- Bound, W., Atkinson, R., 1966. Bacterial aerosol from water closets: a comparison of two types of pan and two types of cover. *The Lancet* 287 (7451), 1369–1370.
- Brooks, J., Tanner, B., Josephson, K., Gerba, C.P., Haas, C., Pepper, I.L., 2005a. A national study on the residential impact of biological aerosols from the land application of biosolids. *J. Appl. Microbiol.* 99 (2), 310–322.
- Brooks, J.P., McLaughlin, M.R., Gerba, C.P., Pepper, I.L., 2012. Land application of manure and class B biosolids: an occupational and public quantitative microbial risk assessment. *J. Environ. Qual.* 41 (6), 2009–2023.
- Brooks, J.P., Tanner, B.D., Gerba, C.P., Haas, C.N., Pepper, I.L., 2005b. Estimation of bioaerosol risk of infection to residents adjacent to a land applied biosolids site using an empirically derived transport model. *J. Appl. Microbiol.* 98 (2), 397–405.
- Castilla, J., Barricarte, A., Aldaz, J., Garcia, C., Ferrer, T., Pelaz, C., Pineda, S., Baladron, B., Martin, I., Goni, B., Aratajo, P., Chamorro, J., Lameiro, F., Torroba, L., Dorronsoro, L., Martinez-Artola, V., Esparza, M., Gastaminza, M., Fraile, P., Aldaz, P., 2008. A large Legionnaires' disease outbreak in Pamplona, Spain: early detection, rapid control and no case fatality. *Epidemiol. Infect.* 136 (6), 823–832.
- CDC, 2016. Developing a Water Management Program to Reduce Legionella Growth & Spread in Buildings: a Practical Guide to Implementing Industry Standards.
- Chan, E., Iseman, M., 2013. Underlying host risk factors for nontuberculous mycobacterial lung disease. *Semin. Respir. Crit. Care Med.* 34 (1), 110–123.
- Chaudhry, R.M., Hamilton, K.A., Haas, C.N., Nelson, K.L., 2017. Drivers of Microbial Risk for Direct Potable Reuse and de Facto Reuse Treatment Schemes: the Impacts of Source Water Quality and Blending. *Int. J. Environ. Res. Publ. Health* 14 (6), 635.
- Chen, N.C., Hanna, S.R., 1978. Drift modeling and monitoring comparisons. *Atmos. Environ.* 12 (8), 1725–1734 (1967).
- Chhipi-Shrestha, G., Hewage, K., Sadiq, R., 2017. Fit-for-purpose wastewater treatment: conceptualization to development of decision support tool (1). *Sci. Total Environ.* 607, 600–612.
- Collins, S., Jorgensen, F., Willis, C., Walker, J., 2015. Real-time PCR to supplement gold-standard culture-based detection of *Legionella* in environmental samples. *J. Appl. Microbiol.* 119 (4), 1158–1169.
- Cooper, I., Hanlon, G., 2010. Resistance of *Legionella pneumophila* serotype 1 biofilms to chlorine-based disinfection. *J. Hosp. Infect.* 74 (2), 152–159.
- Darlow, H., Bale, W., 1959. Infective hazards of water-closets. *The Lancet* 273 (7084), 1196–1200.
- Delgado-Viscogliosi, P., Simonart, T., Parent, V., Marchand, G., Dobbelaere, M., Pierlot, E., Pierzo, V., Menard-Szczepara, F., Gaudard-Ferveur, E., Delabre, K., 2005. Rapid method for enumeration of viable *Legionella pneumophila* and other *Legionella* spp. in water. *Appl. Environ. Microbiol.* 71 (7), 4086–4096.
- Delignette-Muller, M.L., Dutang, C., 2015. fitdistrplus: an R package for fitting distributions. *J. Stat. Software* 64 (4), 1–34.
- Dennis, P., Lee, J., 1988. Differences in aerosol survival between pathogenic and non-pathogenic strains of *Legionella pneumophila* serogroup 1. *J. Appl. Bacteriol.* 65 (2), 135–141.
- Department of Health and Human Services, 2015. A Guide to Developing Risk Management Plans for Cooling Tower Systems. Victoria State Government Melbourne, Australia.
- Ditommaso, S., Ricciardi, E., Giacomuzzi, M., Rivera, S.R.A., Zotti, C.M., 2015. *Legionella* in water samples: how can you interpret the results obtained by quantitative PCR? *Mol. Cell. Probes* 29 (1), 7–12.
- Dowd, S.E., Gerba, C.P., Pepper, I.L., Pillai, S.D., 2000. Bioaerosol transport modeling and risk assessment in relation to biosolid placement. *J. Environ. Qual.* 29 (1), 343–348.
- Dungan, R., 2010. Board-invited review: fate and transport of bioaerosols associated with livestock operations and manures. *J. Anim. Sci.* 88 (11), 3693–3706.
- Dungan, R.S., 2014. Estimation of infectious risks in residential populations exposed to airborne pathogens during center pivot irrigation of dairy wastewaters. *Environ. Sci. Technol.* 48 (9), 5033–5042.
- Durand, R., Schwebach, G., 1989. Gastrointestinal effects of water reuse for public park irrigation. *Am. J. Publ. Health* 79 (12), 1659–1660.
- Fernandez-Cassi, X., Silvera, C., Cervero-Aragó, S., Rusiñol, M., Latif-Eugeni, F., Bruguera-Casamada, C., Civit, S., Araujo, R., Figueras, M., Girones, R., 2016. Evaluation of the microbiological quality of reclaimed water produced from a lagooning system. *Environ. Sci. Pollut. Res.* 23 (16), 16816–16833.
- Fewtrell, L., Kay, D., 2007. Quantitative microbial risk assessment with respect to *Campylobacter* spp. in toilets flushed with harvested rainwater. *Water Environ. J.* 21 (4), 275–280.
- Fields, B.S., Benson, R.F., Besser, R.E., 2002. Legionella and Legionnaires' disease: 25 years of investigation. *Clin. Microbiol. Rev.* 15 (3), 506–526.
- Fisman, D.L.S., Wellenius, G.A., Johnson, C., Britz, P., Gaskins, M., Maher, J., Mittleman, M.A., Spain, V., Haas, C.N., Newbern, C., 2005. It's not the heat, it's the humidity: wet weather increases legionellosis risk in the greater Philadelphia metropolitan area. *J. Infect. Dis.* 192, 2066–2073.
- Fitzgeorge, R., Baskerville, A., Broster, M., Hambleton, P., Dennis, P., 1983. Aerosol infection of animals with strains of *Legionella pneumophila* of different virulence: comparison with intraperitoneal and intranasal routes of infection. *Epidemiol. Infect.* 90 (1), 81–89.
- Fliermans, C., Cherry, W., Orrison, L., Thacker, L., 1979. Isolation of *Legionella*

- pneumophila from non-epidemic-related aquatic habitats. *Appl. Environ. Microbiol.* 37 (6), 1239–1242.
- Fossum, H., Reif, B.P., Tutkun, M., Gjesdal, T., 2012. On the use of computational fluid dynamics to investigate aerosol dispersion in an industrial environment: a case study. *Boundary-Layer Meteorol.* 144 (1), 21–40.
- Galada, H., Gurian, P., Joe, A., Kumar, A., Olson, B., Olson, M., Richter, E., Teng, J., Zhang, H., Xagorarakis, I., 2012. Site Specific Risk Assessment Tool for Land Applied Biosolids. Water Environment Research Foundation, Alexandria, VA, USA.
- Garner, E., Zhu, N., Strom, L., Edwards, M., Pruden, A., 2016. A human exposome framework for guiding risk management and holistic assessment of recycled water quality. *Environ. Sci. Water Res. Technol.* 2 (4), 580–598.
- George, F., Shivaji, T., Pinto, C.S., Serra, L.A.O., Valente, J., Albuquerque, M.J., Vicêncio, P.C.O., San-Bento, A., Diegues, P., Nogueira, P.J., 2016. A Large Outbreak of Legionnaires' Disease in an Industrial Town in Portugal. *Revista Portuguesa de Saúde Pública*, 34(3), 199–208.
- Gerba, C.P., Wallis, C., Melnick, J.L., 1975. Microbiological hazards of household toilets: droplet production and the fate of residual organisms. *Appl. Microbiol.* 30 (2), 229–237.
- Goldstein, R.E.R., Micallef, S.A., Gibbs, S.G., He, X., George, A., Sapkota, A., Joseph, S.W., Sapkota, A.R., 2014. Occupational exposure to *Staphylococcus aureus* and *Enterococcus* spp. among spray irrigation workers using reclaimed water. *Int. J. Environ. Res. Publ. Health* 11 (4), 4340–4355.
- Gregersen, P., Grunnet, K., Uldum, S.A., Andersen, B.H., Madsen, H., 1999. Pontiac fever at a sewage treatment plant in the food industry. *Scand. J. Work. Environ. Health* 291–295.
- Haas, C.N., Rose, J.B., Gerba, C.P., 1999. *Quantitative Microbial Risk Assessment*. Wiley.
- Haas, C.N., Rose, J.B., Gerba, C.P., 2014. *Quantitative Microbial Risk Assessment*. John Wiley & Sons, Inc.
- Hambleton, P., Broster, M., Dennis, P., Henstridge, R., Fitzgeorge, R., Conlan, J., 1983. Survival of virulent *Legionella pneumophila* in aerosols. *J. Hyg.* 90 (03), 451–460.
- Hamilton, K.A., Ahmed, W., Toze, S., Haas, C.N., 2017. Human health risks for *Legionella* and *Mycobacterium avium* complex (MAC) from potable and non-potable uses of roof-harvested rainwater. *Water Res.* 119, 288–303.
- Hamilton, K.A., Haas, C.N., 2016. Critical review of mathematical approaches for quantitative microbial risk assessment (QMRA) of *Legionella* in engineered water systems: research gaps and a new framework. *Environ. Sci. Water Res. Technol.* 2 (4), 599–613.
- Hardy, R., J. S., Fromm, X., Cook, M., 2006. Technical Background Document: Microbial Risk Assessment and Fate and Transport Modeling of Aerosolized Microorganisms at Wastewater Land Application Facilities in Idaho. Idaho Department of Environmental Quality, Boise, ID.
- Haupt, T.E., Heffernan, R.T., Kazmierczak, J.J., Nehls-Lowe, H., Rheineck, B., Powell, C., Leonhardt, K.K., Chitnis, A.S., Davis, J.P., 2012. An outbreak of Legionnaires disease associated with a decorative water wall fountain in a hospital. *Infect. Contr.* 33 (02), 185–191.
- Heyder, J., Gebhart, J., Rudolf, G., Schiller, C.F., Stahlhofen, W., 1986. Deposition of particles in the human respiratory tract in the size range 0.005–15 µm. *J. Aerosol Sci.* 17 (5), 811–825.
- Hinds, W.C., 1999. *Aerosol Technology: Properties, Behavior, and Measurement of Airborne Particles*. Wiley-Interscience, New York.
- Hines, S.A., Chappie, D.J., Lordo, R.A., Miller, B.D., Janke, R.J., Lindquist, H.A., Fox, K.R., Ernst, H.S., Taft, S.C., 2014. Assessment of relative potential for *Legionella* species or surrogates inhalation exposure from common water uses. *Water Res.* 56, 203–213.
- Holmes, N.S., Morawska, L., 2006. A review of dispersion modelling and its application to the dispersion of particles: an overview of different dispersion models available. *Atmos. Environ.* 40 (30), 5902–5928.
- Jahne, M.A., Rogers, S., Holsen, T.M., Grimberg, S.J., Ramler, I., 2015. Emission and Dispersion of Bioaerosols from Dairy Manure Application Sites: Human Health Risk Assessment.
- Jahne, M.A., Rogers, S.W., Holsen, T.M., Grimberg, S.J., 2014. Quantitative microbial risk assessment of bioaerosols from a manure application site. *Aerobiologia* 31 (1), 73–87.
- Jiménez, B., Asano, T., 2008. *Water Reuse: an International Survey of Current Practice, Issues and Needs*. IWA publishing.
- Jjemba, P.K., Johnson, W., Bukhari, Z., LeChevallier, M.W., 2015. Occurrence and control of *Legionella* in recycled water systems. *Pathogens* 4 (3), 470–502.
- Jjemba, P.K., Weinrich, L.A., Cheng, W., Giraldo, E., LeChevallier, M.W., 2010. Regrowth of potential opportunistic pathogens and algae in reclaimed-water distribution systems. *Appl. Environ. Microbiol.* 76 (13), 4169–4178.
- Johnson, D., Lynch, R., Marshall, C., Mead, K., Hirst, D., 2013. Aerosol generation by modern flush toilets. *Aerosol. Sci. Technol.* 47 (9), 1047–1057.
- Johnson, W., Jjemba, P., Bukhari, Z., LeChevallier, M., 2017. Occurrence of *Legionella* in non-potable reclaimed water. *J. Am. Water Works Assoc.* 110 (3). <https://doi.org/10.5942/jawwa.2018.110.0021>.
- Katz, S.M., Hammel, J.M., 1987. The effect of drying, heat, and pH on the survival of *Legionella pneumophila*. *Ann. Clin. Lab. Sci.* 17 (3), 150–156.
- Kincaid, D., Solomon, K., Oliphant, J., 1996. Drop size distributions for irrigation sprinklers. *Trans. ASAE* 39 (3), 839–845.
- Kirschner Jr., R.A., Parker, B.C., Falkinham III, J.O., 1992. Epidemiology of infection by nontuberculous mycobacteria: *Mycobacterium avium*, *Mycobacterium intracellulare*, and *Mycobacterium scrofulaceum* in acid, brown-water swamps of the Southeastern United States and their association with environmental variables. *Am. Rev. Resp. Dis.* 145 (2), 271–275.
- Kohl, R., 1974. Drop size distributions from medium-sized agricultural sprinklers. *Trans. ASAE* 17 (4), 690–693.
- Kusnetsov, J., Neuvonen, L.-K., Korpio, T., Uldum, S.A., Mentula, S., Putus, T., Minh, N.N.T., Martimo, K.-P., 2010. Two Legionnaires' disease cases associated with industrial waste water treatment plants: a case report. *BMC Infect. Dis.* 10 (1), 343.
- LeChevallier, M.W., Bukhari, Z., Jjemba, P., Johnson, W., Haas, C.N., Hamilton, K.A., 2017. Development of a Risk Management Strategy for *Legionella* in Recycled Water Systems (WRF12–05). WaterReuse Research Foundation, Alexandria, VA.
- Levine, A.D., Asano, T., 2004. Peer reviewed: recovering sustainable water from wastewater. *Environ. Sci. Technol.* 38 (11), 201A–208A.
- Li, H., Chien, S.-H., Hsieh, M.-K., Dzombak, D.A., Vidic, R.D., 2011. Escalating water demand for energy production and the potential for use of treated municipal wastewater. *Environ. Sci. Technol.* 45 (10), 4195–4200.
- Lighthart, B., 1994. *Atmospheric Microbial Aerosols*. Springer, pp. 285–303.
- Lighthart, B., Mohr, A., 1987. Estimating downwind concentrations of viable airborne microorganisms in dynamic atmospheric conditions. *Appl. Environ. Microbiol.* 53 (7), 1580–1583.
- Lighthart, B., Shaffer, B., Marthi, B., Ganio, L., 1991. Trajectory of aerosol droplets from a sprayed bacterial suspension. *Appl. Environ. Microbiol.* 57 (4), 1006–1012.
- Lim, K.-Y., Hamilton, A.J., Jiang, S.C., 2015. Assessment of public health risk associated with viral contamination in harvested urban stormwater for domestic applications. *Sci. Total Environ.* 523, 95–108.
- Lucas, M., Martínez, P., Viedma, A., 2012. Experimental determination of drift loss from a cooling tower with different drift eliminators using the chemical balance method. *Int. J. Refrig.* 35 (6), 1779–1788.
- Mansi, A., Amori, I., Marchesi, I., Marcelloni, A., Proietto, A., Ferranti, G., Magini, V., Valeriani, F., Borella, P., 2014. *Legionella* spp. survival after different disinfection procedures: comparison between conventional culture, qPCR and EMA-qPCR. *Microchem. J.* 112, 65–69.
- Mayer, P.W., DeOreo, W.B., 1999. *Residential End Uses of Water*. American Water Works Association.
- Medema, G., Wullings, B., Roelveland, P., Van Der Kooij, D., 2004. Risk assessment of *Legionella* and enteric pathogens in sewage treatment works. *Water Supply* 4 (2), 125–132.
- Metcalfe, Eddy, 2007. *Water Reuse : Issues, Technologies, and Applications: Issues, Technologies, and Applications*. McGraw-hill, Michigan, USA.
- Montero, J., Tarjuelo, J., Carrión, P., 2003. Sprinkler droplet size distribution measured with an optical spectrophotometer. *Irrigat. Sci.* 22 (2), 47–56.
- Moore, G., Hewitt, M., Stevenson, D., Walker, J.T., Bennett, A.M., 2015. Aerosolization of respirable droplets from a domestic spa pool and the use of MS-2 coliphage and *Pseudomonas aeruginosa* as markers for *Legionella pneumophila*. *Appl. Environ. Microbiol.* 81 (2), 555–561.
- Muller, D., Edwards, M.L., Smith, D.W., 1983. Changes in iron and transferrin levels and body temperature in experimental airborne legionellosis. *J. Infect. Dis.* 147 (2), 302–307.
- Nguyen, T.M.N., Illef, D., Jarraud, S., Rouil, L., Campese, C., Che, D., Haeghebaert, S., Ganiayre, F., Marcel, F., Etienne, J., 2006. A community-wide outbreak of legionnaires disease linked to industrial cooling towers—how far can contaminated aerosols spread? *J. Infect. Dis.* 193 (1), 102–111.
- NRMMC, E., 2008. AHMC, Australian Guidelines for Water Recycling: Managing Health and Environmental Risks (Phase 2): Augmentation of Drinking Water Supplies. Environment Protection and Heritage Council, National Health and Medical Research Council. Natural Resource Management Ministerial Council, Canberra.
- Nygård, K., Werner-Johansen, Ø., Rønsen, S., Caugant, D.A., Simonsen, Ø., Kanestrøm, A., Ask, E., Ringstad, J., Ødegård, R., Jensen, T., 2008. An outbreak of Legionnaires disease caused by long-distance spread from an industrial air scrubber in Sarpsborg, Norway. *Clin. Infect. Dis.* 46 (1), 61–69.
- O'Toole, J., Keywood, M., Sinclair, M., Leder, K., 2009. Risk in the mist? Deriving data to quantify microbial health risks associated with aerosol generation by water-efficient devices during typical domestic water-using activities. *Water Sci. Technol.* 60 (11), 2913–2920.
- Olsen, J.S., Aarskaug, T., Thrane, I., Pourcel, C., Ask, E., Johansen, G., Waagen, V., Blatny, J.M., 2010. Alternative routes for dissemination of *Legionella pneumophila* causing three outbreaks in Norway. *Environ. Sci. Technol.* 44 (22), 8712–8717.
- OSHA, 2017. *Extended Unusual Work Shifts*.
- Paez-Rubio, T., Peccia, J., 2005. Estimating solar and nonsolar inactivation rates of airborne bacteria. *J. Environ. Eng.* 131 (4), 512–517.
- Paez-Rubio, T., Ramarui, A., Sommer, J., Xin, H., Anderson, J., Peccia, J., 2007. Emission rates and characterization of aerosols produced during the spreading of dewatered class B biosolids. *Environ. Sci. Technol.* 41 (10), 3537–3544.
- Palmer, C.J., Bonilla, G.F., Roll, B., Paszko-Kolva, C., Sangermano, L.R., Fujioka, R.S., 1995. Detection of *Legionella* species in reclaimed water and air with the EnviroAmp *Legionella* PCR kit and direct fluorescent antibody staining. *Appl. Environ. Microbiol.* 61 (2), 407–412.
- Pascual, L., Pérez-Luz, S., Yáñez, M.A., Santamaría, A., Gibert, K., Salgot, M., Apraiz, D., Catalán, V., 2003. Bioaerosol emission from wastewater treatment plants. *Aerobiologia* 19 (3–4), 261–270.
- Peterson, E.W., Lighthart, B., 1977. Estimation of downwind viable airborne microbes from a wet cooling tower—including settling. *Microb. Ecol.* 4 (1), 67–79.
- Pouillot, R., Delignette-Muller, M.-L., 2010. Evaluating variability and uncertainty in

- microbial risk assessment using two R packages. *Int. J. Food Microbiol.* 142 (3), 330–340.
- Qin, T., Tian, Z., Ren, H., Hu, G., Zhou, H., Lu, J., Luo, C., Liu, Z., Shao, Z., 2012. Application of EMA-qPCR as a complementary tool for the detection and monitoring of *Legionella* in different water systems. *World J. Microbiol. Biotechnol.* 28 (5), 1881–1890.
- Rouil, L., Gardenas, G., Marcel, F., 2004. Évaluation de la dispersion atmosphérique d'aérosols potentiellement contaminés lors de l'épidémie de légionellose de la région de Lens. *NUMÉRO SPÉCIAL CONSACRÉ À LA LÉGIONELLOSE 2004* (36/37), 182–184.
- Sales-Ortells, H., Medema, G., 2014. Screening-level microbial risk assessment of urban water locations: a tool for prioritization. *Environ. Sci. Technol.* 48 (16), 9780–9789.
- Sales-Ortells, H., Medema, G., 2015. Microbial health risks associated with exposure to stormwater in a water plaza. *Water Res.* 74, 34–46.
- Sánchez-Monedero, M., Aguilar, M., Fenoll, R., Roig, A., 2008. Effect of the aeration system on the levels of airborne microorganisms generated at wastewater treatment plants. *Water Res.* 42 (14), 3739–3744.
- Schoen, M.E., Ashbolt, N.J., 2011. An in-premise model for *Legionella* exposure during showering events. *Water Res.* 45 (18), 5826–5836.
- Seinfeld, J.H., 1986. *Atmospheric Chemistry and Physics of Air Pollution*. John Wiley & Sons.
- Selby, K.A., Puckorius, P.R., Helm, K.R., 1996. The use of reclaimed water in electric power stations and other industrial facilities. *Water Air Soil Pollut.* 90 (1–2), 183–193.
- Sharvelle, S., Ashbolt, N., Clerico, E., Hultquist, R., Leverenz, H., Olivieri, A., 2017. Risk-based Framework for the Development of Public Health Guidelines for Decentralized Non-potable Water Systems, Prepared by the National Water Research Institute for the Water Environment & Reuse Foundation. WE&RF Project No. SIWM10C15, Alexandria, VA.
- Sheikh, B., Cort, R.P., Kirkpatrick, W.R., Jaques, R.S., Asano, T., 1990. Monterey wastewater reclamation study for agriculture. *Res. J. Water Pollut. Contr. Fed.* 216–226.
- Ssematimba, A., Hagenaars, T.J., De Jong, M.C., 2012. Modelling the wind-borne spread of highly pathogenic avian influenza virus between farms. *PLoS One* 7 (2), e31114.
- Tanner, B.D., Brooks, J.P., Gerba, C.P., Haas, C.N., Josephson, K.L., Pepper, I.L., 2008. Estimated occupational risk from bioaerosols generated during land application of class B biosolids. *J. Environ. Qual.* 37 (6), 2311–2321.
- Taylor, M.J., Bentham, R.H., Ross, K.E., 2014. Limitations of using propidium monoazide with qPCR to discriminate between live and dead *Legionella* in biofilm samples. *Microbiol. Insights* 7, 15.
- Teltsch, B., Shuval, H., Tadmor, J., 1980. Die-away kinetics of aerosolized bacteria from sprinkler application of wastewater. *Appl. Environ. Microbiol.* 39 (6), 1191–1197.
- Teng, J., Kumar, A., Gurian, P.L., Olson, M.S., 2013. A spreadsheet-based site specific risk assessment tool for land-applied biosolids. *Open Environ. Eng. J.* 6, 7–13.
- Thomas, V., McDonnell, G., Denyer, S.P., Maillard, J.-Y., 2010. Free-living amoebae and their intracellular pathogenic microorganisms: risks for water quality. *FEMS Microbiol. Rev.* 34 (3), 231–259.
- Thomson, R.M., Carter, R., Tolson, C., Coulter, C., Huygens, F., Hargreaves, M., 2013a. Factors associated with the isolation of Nontuberculous mycobacteria (NTM) from a large municipal water system in Brisbane, Australia. *BMC Microbiol.* 13 (1), 1.
- Thomson, R.M., Carter, R., Tolson, C., Coulter, C., Huygens, F., Hargreaves, M., 2013b. Factors associated with the isolation of Nontuberculous mycobacteria (NTM) from a large municipal water system in Brisbane, Australia. *BMC Microbiol.* 13 (1), 1–8.
- USEPA, 1982. Estimating Microorganism Densities in Aerosols from Spray Irrigation of Wastewater. US Environmental Protection Agency.
- USEPA, 2011. Exposure Factors Handbook, Washington, DC.
- Van Leuken, J., Swart, A., Havelaar, A., Van Pul, A., Van der Hoek, W., Heederik, D., 2015. Atmospheric dispersion modelling of bioaerosols that are pathogenic to humans and livestock—A review to inform risk assessment studies. *Microb. Risk Anal.* 1, 19–39.
- van Lier, A., McDonald, S.A., Bouwknegt, M., Kretzschmar, M.E., Havelaar, A.H., Mangen, M.-J.J., Wallinga, J., de Melker, H.E., 2016. Disease burden of 32 infectious diseases in The Netherlands, 2007–2011. *PLoS One* 11 (4), e0153106.
- Viau, E., Bibby, K., Paez-Rubio, T., Peccia, J., 2011. Toward a consensus view on the infectious risks associated with land application of sewage sludge. *Environ. Sci. Technol.* 45 (13), 5459–5469.
- Wallis, L., Robinson, P., 2005. Soil as a source of *Legionella pneumophila* serogroup 1 (Lp1). *Aust. N. Z. J. Publ. Health* 29 (6), 518–520.
- Walser, S.M., Brenner, B., Wunderlich, A., Tuschak, C., Huber, S., Kolb, S., Niessner, R., Seidel, M., Höller, C., Herr, C.E., 2017. Detection of *Legionella*-contaminated aerosols in the vicinity of a bio-trickling filter of a breeding sow facility—A pilot study. *Sci. Total Environ.* 575, 1197–1202.
- Walser, S.M., Gerstner, D.G., Brenner, B., Höller, C., Liebl, B., Herr, C.E., 2014. Assessing the environmental health relevance of cooling towers—a systematic review of legionellosis outbreaks. *Int. J. Hyg Environ. Health* 217 (2), 145–154.
- Ward, R.L., Knowlton, D.R., Stober, J., Jakubowski, W., Mills, T., Graham, P., Camann, D.E., 1989. Effect of wastewater spray irrigation on rotavirus infection rates in an exposed population. *Water Res.* 23 (12), 1503–1509.
- Weiss, D., Boyd, C., Rakeman, J.L., Greene, S.K., Fitzhenry, R., McProud, T., Musser, K., Huang, L., Kornblum, J., Nazarian, E.J., 2017. A large community outbreak of Legionnaires' disease associated with a cooling tower in New York City, 2015. *Publ. Health Rep.* 132 (2), 241–250.
- Whiley, H., Taylor, M., 2014. *Legionella* detection by culture and qPCR: comparing apples and oranges. *Crit. Rev. Microbiol.* 42 (1), 65–74.
- Zhang, M., Liu, W., Nie, X., Li, C., Gu, J., Zhang, C., 2012. Molecular analysis of bacterial communities in biofilms of a drinking water clearwell. *Microb. Environ.* 27 (4), 443–448.