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# Leveraging regulatory monitoring data for quantitative microbial risk assessment of *Legionella pneumophila* in cooling towers

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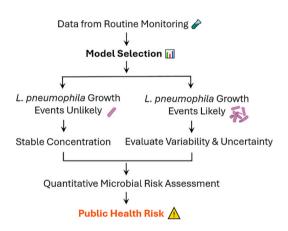
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#### HIGHLIGHTS

- Models predicted *L. pneumophila* concentration variability in cooling towers.
   Results were integrated into QMRA to
- predict human health risks.

   Cooling towers at risk of exceeding
- $\bullet$  Cooling towers at risk of exceeding health targets at concentrations  ${>}10^5$  CFU  ${\rm L}^{-1}.$
- Mitigating peak concentrations is critical for controlling public health risks.

#### G R A P H I C A L A B S T R A C T



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# ABSTRACT

Cooling towers are critical engineered water systems for air conditioning and refrigeration but can create favorable conditions for *Legionella pneumophila* growth and aerosolization. Human exposure to *L. pneumophila*-contaminated aerosols can cause Legionnaire's disease. Routine monitoring of *L. pneumophila* in cooling towers offers possibilities to develop quantitative microbial risk assessment (QMRA) models to guide system design, operation, control, and maintenance. Here, we used the regulatory monitoring database from Quebec, Canada, to develop statistical models for predicting *L. pneumophila* concentration variability in cooling towers and integrate these models into a screening-level QMRA model to predict human health risks. Analysis of 105,463 monthly *L. pneumophila* test results revealed that the exceedance rate of the 10<sup>4</sup> colony-forming unit (CFU) per liter threshold was constant at 10 % from 2016 to 2020, emphasizing the need to better validate the efficacy of corrective measures following the threshold exceedances. Among 2852 cooling towers, 51.2 % reported no detections, 38.5 % had up to nine positives, and 10.2 % over ten. The gamma or the lognormal distributions adequately described site-specific variations in *L. pneumophila* concentrations, but parametric uncertainty was very high for the lognormal distribution. We showed that rigorous model comparison is essential to predict peak

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concentrations accurately. Using QMRA, we found that an average  $\it L.$  pneumophila concentration below  $1.4 \times 10^4$  CFU  $\it L^{-1}$  should be maintained in cooling towers to meet a health-based target of  $10^{-6}$  DALY/pers.-year for clinical severity infections. We identified 137 cooling towers at risk of exceeding this limit, primarily due to the observation or prediction of rare peak concentrations above  $10^5$  CFU  $\it L^{-1}$ . Effective mitigation of those peaks is critical to controlling public health risks associated with  $\it L.$  pneumophila.

#### 1. Introduction

Exposure to Legionella pneumophila through inhalation of aerosols produced by engineered water systems, such as cooling towers, is a common cause of Legionnaire's disease. This disease can result in a severe form of pneumonia, particularly threatening individuals who are older, have weakened immune systems, or suffer from chronic lung diseases (National Academies of Sciences and Medicine, 2020). Cooling towers, engineered water systems designed to remove excess heat from buildings by cooling water, are critical for air conditioning and refrigeration but can provide ideal conditions for the growth and aerosolization of L. pneumophila. These systems operate using evaporative cooling. Warm water from cooling systems — usually between 29 and 35 °C, a temperature range favorable for L. pneumophila growth — is sprayed as fine droplets onto packing or honeycomb material. As ambient air is drawn into the cooling tower, either by natural or mechanical ventilation, a small portion of the water evaporates, reducing its temperature but also generating aerosol droplets that may carry L. pneumophila. While cooling towers should be equipped with drift eliminators designed to minimize aerosol droplets released into the atmosphere, the efficiency of these devices is not absolute (ASHRAE, 2008). The release of contaminated droplets into the atmosphere can pose public health risks for neighboring communities.

The growth of L. pneumophila colonies in cooling towers is influenced by various factors, such as the presence of protozoa (which serve as hosts), biofilms, nutrient availability, water temperature, and manufacturing materials (Kusnetsov et al., 1993; Paniagua et al., 2020; Türetgen and Cotuk, 2007). To minimize growth, the primary strategy involves chemical water disinfection (Kim et al., 2002). However, predicting the presence and survival of L. pneumophila in cooling towers solely from design and operational parameters is challenging. Various risk management guidelines and guidance documents based on the hazard analysis and critical control point (HACCP) method recommend routine monitoring of L. pneumophila in bulk water to validate water treatment efficiency (ASHRAE, 2018, 2020; Cooling Technology Institute (CTI), 2000; World Health Organization (WHO), 2011). Legal requirements for routine monitoring of L. pneumophila have been established across numerous countries (Radziminski and White, 2023; Van Kenhove et al., 2019), often necessitating corrective actions when specified L. spp or L. pneumophila concentrations are exceeded. For example, Quebec's regulation requires intervention and confirmation of the effectiveness of corrective measures when the L. pneumophila concentration exceeds 10<sup>4</sup> colonies forming unit (CFU) L<sup>-1</sup> (Gouvernment du Québec, 2014).

A quantitative microbial risk assessment (QMRA) framework has been proposed to assess health risks associated with exposure to *L. pneumophila*-laden aerosols from cooling towers (Hamilton et al., 2018). This framework incorporates dose–response models to predict human health effects from the deposition of *L. pneumophila* in the lungs (Armstrong and Haas, 2007) and uses a Gaussian plume model to simulate the fate and transport of contaminated aerosol droplets (Hardy et al., 2006). Applying QMRA makes it possible to estimate public health risks based on concentrations of *L. pneumophila* in the bulk water from cooling towers. These risk predictions can be compared with health-based targets to provide a foundation for risk-based criteria to guide system design, operation, control, and maintenance (National Academies of Sciences and Medicine, 2020).

Within the QMRA framework proposed by Hamilton et al. (2018),

the health risk is directly proportional to the concentrations of *L. pneumophila* in the bulk water of the cooling tower. Extensive routine *L. pneumophila* monitoring data from programs designed to validate treatment efficiency offer a significant opportunity to develop statistical models to investigate temporal variations in these concentrations. Parametric models, such as mixed Poisson distributions, have been widely used to model temporal variations in microbial concentrations in surface water sources (Haas et al., 1999; Masago et al., 2004; Teunis et al., 1997). These distributions have also been used in ecology to describe variations in the abundance of populations governed by an environmental carrying capacity (Dennis and Patil, 1988; Dennis and Patil, 1984). Despite the apparent potential of such models, their application to model routine monitoring *L. pneumophila* data remains unexplored. Bridging this gap could facilitate the development of more transparent, risk-based strategies for *L. pneumophila* risk assessment and management.

The objectives of our study are to i) develop candidate statistical models to predict the variability and uncertainty in *L. pneumophila* concentrations obtained from routine monitoring of bulk water in cooling towers, ii) establish a framework for model comparison and selection and implement it for an extensive database, and iii) incorporate selected models within a screening-level QMRA model to predict human health risks associated with exposure to droplet aerosols generated by a representative cooling tower. We applied this novel framework to model *L. pneumophila* test results from 2852 cooling towers in Quebec, Canada, routinely monitored from 2016 to 2020.

### 2. Methodology

# 2.1. Database: regulatory monitoring of cooling towers in Quebec

We accessed the Quebec regulatory database, which contains *L. pneumophila* monitoring results for 2852 cooling towers across 1960 buildings in Quebec, Canada. These routine monitoring activities are mandated by a provincial regulation for the maintenance of cooling towers (Gouvernement du Québec, 2014). The provincial reference laboratory (CEAEQ) oversees the accreditation of municipal, institutional, and private laboratories providing these analytical services. CEAEQ specifies the analytical methods and sample treatment protocols that laboratories must follow, sets and verifies mandatory QA/QC requirements, and periodically conducts blind proficiency testing. As all accredited laboratories are qualified to analyse *Legionella* in water using recognized analytical methods, the resulting data is comparable, allowing us to merge it into one dataset.

For each cooling tower, the database consists of results from monthly monitoring of L. pneumophila concentrations in bulk water obtained yearly or on a seasonal basis from 2016 to 2020. It also includes general system information, including the building type, usage type, service period, and location. Specific information on water treatment (types of biocides used, application frequencies, dosages, etc.) was unavailable. The regulation defines an action level of  $10^4$  colony-forming units (CFU)  $L^{-1}$  and a human health risk level of  $10^6$  CFU  $L^{-1}$ . Upon exceedance of the action level, the regulation requires immediate corrective intervention. A strict decontamination procedure must be applied when the concentration of L. pneumophila is equal to or  $>10^6$  CFU  $L^{-1}$ . Additional L. pneumophila results following a decontamination procedure were included in the database for some cooling towers, following testing requirements. However, these results could not be differentiated from

routine monitoring results.

# 2.2. Sample collection and quantification of L. pneumophila with culture-based methods

Each sample was collected and stored following Standard DR-09-11 (Centre d'expertise en analyse environnementale du Québec, 2022). L. pneumophila enumeration was performed using culture-based methods adapted from AFNOR NF T90-431 or ISO 11731:2017, depending on the laboratory conducting the analyses. Diluted samples were spread on glycine vancomycin polymyxin cycloheximide agar medium (GVPC) or BMPA, with or without acid (pH 2; 5 min) or heat (50 °C; 30 min) pre-treatment or a combination of acid and heat. Samples were then incubated at 36 °C for 8–11 days, and presumptive colonies of Legionella spp. were grown with buffered charcoal yeast extract (BCYE) supplemented or not with cysteine at 36  $\pm$  1.5 °C for 3–5 days. L. pneumophila species were identified, in most cases, by a latex agglutination test, and all results were expressed as CFU. Results at the detection limit were reported in CFU L $^{-1}$  calculated as 1 CFU per volume of sample tested.

#### 2.3. Rationale for the selection of statistical models

To assess temporal variations in L. pneumophila concentrations in cooling towers, three discrete parametric distributions — Poisson, Poisson gamma, and Poisson lognormal — were selected. We chose these discrete distributions to avoid replacing non-detects with a specific concentration (e.g., one organism per analyzed volume). Instead, we treated the sample concentration as a random variable that can be estimated using a Poisson distribution. This approach requires knowledge of the number of CFUs and the water volume analyzed per sample. However, laboratories do not typically report this critical information. We introduced a method to estimate the number of CFUs and the analyzed volume of each sample from reported concentrations, although the potential bias in these estimates could not be assessed. A direct comparison between the fit of these distributions to actual and estimated results would provide insights into the limitations of our approach. Ideally, laboratories should report raw data (i.e., the number of CFUs and the analyzed volume) to facilitate the modelling of temporal variations for risk assessment.

We developed an algorithm to identify the best modelling approach based on monitoring results. Using a goodness-of-fit test for the Poisson distribution, we determined whether the concentration was statistically stable or variable in cooling towers when at least one positive result was recorded. The distribution of microorganisms in water typically exhibits more dispersion than the Poisson distribution predicts, meaning that the variance of organism counts exceeds their average number. Our analysis attributes overdispersion relative to the Poisson distribution solely to temporal variations in concentrations. Yet, other heterogeneity sources such as organism aggregation in samples or variable recovery rates from enumeration methods — can also contribute to overdispersion (Haas and Heller, 1986). Although these other heterogeneity sources have not been analyzed for L. pneumophila, they are well-documented for other bacteria (El-Shaarawi et al., 1981; Haas and Heller, 1986; Pipes et al., 1977). Examining subsamples from individual water samples from cooling towers could elucidate the impact of *L. pneumophila* aggregation on concentration variation.

Gamma and lognormal distributions were chosen as mixture distributions to characterize temporal variations in *L. pneumophila* concentrations. The theoretical foundation for selecting these distributions lies in their ability to approximate random variations in population size, constrained by an environmental carrying capacity. The gamma distribution can be derived from population growth following the logistic function (Dennis and Patil, 1984), while the lognormal distribution emerges from the Gompertz function (Dennis and Patil, 1988). Both the logistic and Gompertz models yield sigmoid functions representing

population growth in three stages: initial slow growth, followed by optimal growth, which eventually slows down as the population approaches its carrying capacity. However, the Gompertz curve tapers off more gradually than the logistic curve. Thus, the lognormal distribution may indicate that *L. pneumophila* has not reached its carrying capacity, whereas the gamma distribution suggests that the carrying capacity has been reached. This carrying capacity might be affected by operational factors, including water treatment practices and the efficacy of interventions following regulatory threshold exceedances.

#### 2.4. Estimation of L. pneumophila counts from reported concentrations

We developed an approximation method to infer colony counts and tested volumes from reported concentrations. This method reduces potential bias associated with fixed detection limits. Statistical inference was not made directly from reported concentrations since treating detection limits as actual microbial concentrations or as censored data can significantly bias statistical analyses (Chik et al., 2018).

Initially, for concentrations reported at the detection limit, we calculated the tested volume  $(V_i)$  for each instance (i) using the formula:

$$V_i = \frac{1}{G_{\rm ID\,i}} \text{ for } i = 1, ..., j$$
 (1)

where  $C_{LD,i}$  represents the detection limit and j is the count of these instances in the database.

Subsequently, for each detected concentration ( $C_{\text{detected,m}}$ ), we computed the corresponding number of colonies ( $k_{i,m}$ ) by applying the previously determined volume ( $V_i$ ) in the equation:

$$k_{i,m} = \frac{V_i}{C_{\text{detected i.m}}} \text{ for } i = 1, ..., j \text{ and for } m = 1, ..., n$$
 (2)

Here, n is the total number of distinct detected concentrations. This calculation resulted in j pairs of colony numbers and volumes, each associated with a specific detected concentration.

To approach the statistical analysis conservatively and avoid underestimating the sampling uncertainty, we selected the minimum integer of colonies ( $k_{min}$ ) along with its corresponding volume (V). This approach minimizes the colony count to ensure the broadest possible confidence intervals for the average number of colonies per sample.

# 2.5. Estimation of L. pneumophila concentration from counts in individual water samples

Applying Poisson principles to the inferred colony counts, we used maximum likelihood estimators to derive L. pneumophila concentrations for individual water samples. To do this, the distribution of organisms within a specific volume of water is characterized as a discrete random variable. Assuming that these organisms are randomly distributed in the water sample, the probability of finding a specific count of organisms (k) within a sample of known concentration (c) and volume (V) is given by the Poisson distribution:

$$P(k;c,V) = \frac{cV^k}{k!} exp(-cV)$$
 (3)

For a sample adhering to a Poisson distribution, the maximum likelihood estimate for the concentration (*c*) is the sample arithmetic mean given by:

$$\bar{c} = \frac{\sum_{i=1}^{n} k_i}{\sum_{i=1}^{n} V_i} \tag{4}$$

This estimator is only valid if organisms are distributed "randomly," i.e., the bulk solution is well-mixed, and organisms are not aggregated.

#### 2.6. Stability or fluctuation of L. pneumophila concentrations over time

We employed a likelihood ratio test, applied individually to cooling tower datasets, to determine whether *L. pneumophila* concentrations measured monthly remain statistically stable over time or exhibit significant temporal fluctuations. This test compares the fit of a single Poisson distribution with a constant concentration (the null hypothesis) to an alternative hypothesis where each sample has its own Poisson distribution. (Haas et al., 1999).

The null hypothesis of the test posits that the data set is adequately described by a single Poisson distribution with a constant concentration,  $\bar{c}$ . The likelihood of the null hypothesis is given by:

$$L^{0} = \prod_{i=1}^{n} \frac{\overline{c} \ V_{i}^{k}}{k!} exp(-\overline{c} \ V_{i})$$
 (5)

Conversely, the alternative hypothesis suggests that each sample has its concentration  $\bar{c}$  at the time of sampling. Therefore, the likelihood for the alternative hypothesis is:

$$L^{A} = \prod_{i=1}^{n} \frac{k_{i}^{k_{i}}}{k_{i}!} exp(-k_{i})$$
 (6)

The test statistic ( $\land$ ) can be simplified to:

$$-\ln(\wedge) = \sum_{i=1}^{n} \left[ (\overline{c} V_i - k_i) - k_i \ln\left(\frac{\overline{c}V_i}{k_i}\right) \right]$$
 (7)

The null hypothesis is rejected when the value of  $-2ln(\ \land\ )$  exceeds the upper  $1-\alpha$  percentile of a  $\chi^2$  distribution with n-1 degrees of freedom. The degrees of freedom represent the difference in the number of parameters between the alternative hand null hypotheses. The error risk  $\alpha$  was set at 5 %. This analysis was applied to each cooling tower dataset with fewer than 10 detects, where Poisson gamma and Poisson lognormal models can have poor performance due to a high proportion of non-detects.

# 2.7. Characterizing fluctuation of L. pneumophila concentrations over time

To address overdispersion and capture a broader range of variability, we introduced gamma and lognormal mixture distributions for modelling *L. pneumophila* counts in cooling towers. Distributions of microorganisms in water are often more dispersed than what a Poisson distribution would predict; that is, the variance of the number of organisms exceeds the mean. This overdispersion relative to the Poisson distribution can result from spatial or temporal heterogeneities. This heterogeneity can be accounted for by a continuous random variable representing the concentration of each sample. The marginal distribution of the number of organisms is then obtained by the following integral:

$$P(k;c,V,\delta) = \int_{0}^{\infty} P_{p}(k;cV)h(c;\delta)dc$$
 (8)

where h is a continuous distribution with parameters  $\delta$  representing the temporal variations in concentrations. Gamma and lognormal distributions have been selected to model temporal variations. The general properties of these distributions are presented in Table 1.

#### 2.8. Bayesian inference for model parameter estimation

Using hierarchical Bayesian models and Markov Chain Monte Carlo (MCMC) simulations, we estimated the parameters of Poisson, Poisson gamma, and Poisson lognormal distributions. The hierarchical model is structured as follows: At the first level, the observed number of colonies per volume is distributed according to a Poisson distribution. At the second level, concentration c is modeled as a latent variable (i.e., not directly observable) following a continuous distribution (either gamma or lognormal for this study), making the first level conditional on c. The hierarchical structures for Poisson gamma and Poisson lognormal models are:

- Poisson gamma:  $x_i | c_i, V_i \sim \text{Poisson}(c_i V_i), c_i \sim \text{Gamma}(\alpha, \beta)$
- Poisson lognormal:  $x_i | c_i, V_i \sim \text{Poisson}(c_i V_i), c_i \sim \text{Lognormal}(\mu, \sigma)$

Prior distributions were chosen to ensure Markov chain stationarity while minimizing the prior influence on the posterior distribution. A conjugate Gamma  $(\alpha,\beta)$  prior was assigned to parameter c, with  $\alpha$  and  $\beta$  set to 0.01. Uniform (min, max) priors were used to infer gamma distribution parameters  $\alpha$  and  $\beta$ , with bounds set at 0 and 10 for  $\alpha$ , and  $10^{-12}$  and  $10^{-1}$  for  $\beta$ . A uniform (min, max) prior was selected for  $\mu$ , with bounds at - 10 and 10. An  $exp(\lambda)$  prior was allocated to  $\sigma$  as suggested by McElreath (2018), with  $\lambda$  set at 0.1, assuming the logarithm of the standard deviation was significantly below 50 for all cooling towers.

Models were fitted using MCMC simulations with rjags (v4-10) in R (v4.1.0). For each parameter, three Markov chains were run for  $10^5$  iterations following a burn-in of  $10^3$  iterations. The Brooks-Gelman-Rubin scale reduction factor was applied to assess chain convergence, and the effective sample size (the ratio of sample size to autocorrelation in Markov chains) was evaluated to ensure comprehensive exploration of the posterior distribution, deemed well-estimated at an effective size over 10,000. Brooks-Gelman-Rubin reduction factors and effective sample sizes were calculated using the diagMCMC function (Kruschke, 2014).

# 2.9. Model comparison and selection using a deviance information criterion

We applied the deviance information criterion (DIC) (Spiegelhalter et al., 2002) to compare competing Poisson and mixed Poisson models, identifying the best fit for each cooling tower. The DIC is computed as follows:

$$DIC = \overline{D} - (\overline{D} - \widehat{D}) = \overline{D} + p_D \tag{9}$$

where  $\overline{D}$  is the mean deviance across the sampled parameter values from the posterior distribution, and  $\widehat{D}$  is a penalty term related to the risk of model overfitting. A model with a lower DIC value is considered superior. Generally, a model with a DIC at least three points lower than that of other models is significantly better (Spiegelhalter et al., 2002). For hierarchical models with a latent variable (e.g., mixed Poisson distributions), different DICs can be calculated for the model's hierarchical levels. The fit of a Poisson distribution should be assessed with a conditional DIC (cDIC), while that of a mixed Poisson distribution should be evaluated with a marginal DIC (mDIC) (Millar, 2009). The cDIC can be directly calculated using the current version of rjags in R. However,

**Table 1**Density functions describing the variation of a concentration c, mean, and standard deviation for the gamma and lognormal distributions.

Distribution	Density function	Arithmetic mean	Standard deviation
Gamma $(\alpha, \beta)$	$rac{eta^{lpha}}{\Gamma(lpha)}c^{lpha-1}e^{-eta c}$	lpha/eta	$lpha/eta^2$
Lognormal $(\mu, \sigma)$	$\frac{1}{c\sigma\sqrt{2\pi}}exp\left[-\frac{\left[\ln c-\mu\right]^2}{2\sigma^2}\right]$	$expigg(\mu+rac{\sigma^2}{2}igg)$	$\sqrt{exp(\sigma^2)-1} \ expigg(\mu+rac{\sigma^2}{2}igg)$

computing the mDIC is more complex as it requires integrating the likelihood function of the distribution, which cannot be done directly in rjags. Thus, mDICs for mixed Poisson distributions were computed through numerical integration following the approach proposed by Quintero and Lesaffre (2018).

The cooling towers were classified using a decision-making algorithm identifying the best model describing *L. pneumophila* concentrations (Fig. 1). For cooling towers where the statistical distribution of data could be validated, we computed the median value and the 95 % uncertainty interval of the average *L. pneumophila* concentration predicted by the best-fit model.

#### 2.10. Screening-level quantitative microbial risk assessment model

A screening-level QMRA model was developed to estimate public health risks associated with exposure to aerosols emitted by a hypothetical cooling tower (Table 2). The model translates arithmetic mean L. pneumophila concentrations, predicted by the best-fit statistical models, into risk estimates. The QMRA uses point estimates for all parameters; it does not incorporate variability and uncertainty in model inputs. However, given that parametric uncertainty can be assessed using the statistical approach presented, we performed the OMRA twice: (1) using the best estimate of the predicted arithmetic mean L. pneumophila concentration and (2) using the upper 97.5 % uncertainty bound of the predicted arithmetic mean. This approach allows us to quantify the impact of uncertainty in L. pneumophila concentrations on the final risk estimates. To determine the dose of L. pneumophila that can deposit in the respiratory tract of the exposed subject, it is necessary to develop and integrate a series of sub-models that account for various stages of the process. These stages include the emission of contaminated droplets by the cooling tower, the transport and atmospheric dispersion of the droplets, the inactivation of L. pneumophila during atmospheric transport, and its inhalation and deposition in the human respiratory tract (Table 2). For our screening-level QMRA, a hypothetical cooling tower was considered, characterized by an effective head of 10 m and an average recirculation flow rate of  $10^3$  L s<sup>-1</sup>. The analysis also assumes that a drift eliminator reduces evaporative water loss to 0.003 % of the recirculation flow rate (ASHRAE, 2008).

2.10.1. Emission, fate, and transport of aerosol droplets discharged by the cooling tower

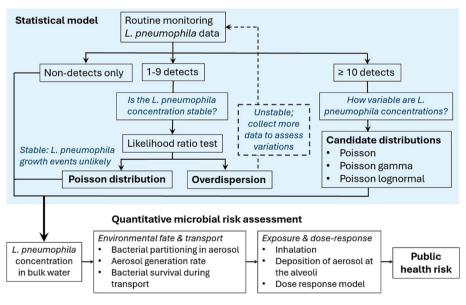
We focused our analysis only on aerosol droplets with diameters  $\leq\!150~\mu\text{m}$ , as they quickly evaporate once airborne and can be transported over long distances (Bourouiba, 2021). To estimate the massweighted fraction of these aerosols, we applied the lognormal distribution of aerosol mass modeled by Peterson and Lighthart (1977) from measurements obtained at the cooling tower outlet by Shofner and Thomas (1971). For the specific fractions of *L. pneumophila* bacteria transferred from water to aerosols with a diameter of 1–10  $\mu\text{m}$ , we relied on estimates from Hamilton et al. (2018) extracted from the empirical cumulative distribution function obtained by Allegra et al. (2016). These fractions are conservative for QMRA, given that the enumeration of *L. pneumophila* bacteria for each droplet diameter was determined using a qPCR method, which measures both viable and dead cells.

We employed a Gaussian model to model the transport and dispersion of these aerosols (Fig. 2). Based on results obtained at 66 meteorological stations in Quebec by Ilinca et al. (2003), we considered an arithmetic mean annual wind speed of 4.5 m/s. For environmental conditions, the Gaussian model predicts that the highest concentration of the pollutant emitted at an effective height of 10 m would be observed at a distance of 110 m from the point of emission.

Droplets were considered as evaporated immediately upon emission from the cooling tower. To evaluate the survival of *L. pneumophila* within these evaporated aerosol droplets, we used a first-order two-phase inactivation model developed from the data of Katz and Hammel (1987). This model predicts a high inactivation rate in the first 30 s ( $\lambda_{E1} = 0.12$  s<sup>-1</sup>), which then decreases ( $\lambda_{E2} = 7 \times 10^{-4}$  s<sup>-1</sup>). Inactivation rates for aqueous aerosols are expected to be very low ( $\lambda_A \sim 1 \times 10^{-4}$  s<sup>-1</sup>) compared to the rates for evaporated aerosols (first phase) (Hamilton et al., 2018).

## 2.10.2. Inhalation, deposition of aerosols, and health effects

We used deposition efficiencies for aerosols with diameters ranging from 1 to 10  $\mu$ m, asreported by Heyder et al. (1986), and incorporated their assumptions for an oral inhalation rate of 15 L air min<sup>-1</sup>, a respiratory cycle duration of 8 s, and an average respiratory volume of 1 L into our model. Our model assumed an exposure duration of one hour per day at a frequency of 365 days per year, as proposed by Hamilton et al. (2018), to assess the risks associated with residential exposure. The



**Fig. 1.** Decision framework for selecting the appropriate statistical model to assess *L. pneumophila* concentrations in bulk water from a cooling tower. The framework distinguishes between stable and variable concentration patterns, guiding model selection. The chosen model informs quantitative microbial risk assessment (QMRA), linking routine monitoring data to public health risks.

 Table 2

 Input parameters of the quantitative microbial risk assessment (QMRA) model.

Equation	Parameter	Unit	Value	Reference
	The concentration of the organism in	$ m CFU~L^{-1}$	Specific	-
1	water ( $C_{water}$ ) Detection method recovery rate ( $R$ )	Fraction	0.70	
$Q_{emission} = \frac{1}{R} C_{water} Q_{water} E \times q$	Detection method recovery rate (it)	Traction	0.70	Johnson et al. (2018)
				McCuin et al. (2021)
	Water recirculation rate $(Q_{water})$	m <sup>3</sup> /s	$10^{3}$	Hensley (2009)
	Effectiveness of drift eliminator (E)	%	0.003	ASHRAE (2008)
	Mass-weighted fraction of aerosols	Fraction	0.17	Peterson and
	with diameter $\leq 100 \ \mu m \ (q)$			Lighthart (1977
	Windows I ( )	/-	4.5	Shofner and Thomas (1971)
	Wind speed $(\mu)$	m/s	4.5	Ilinca et al. (2003)
$C_{air}(x, y, z) = rac{Q_{emission}}{2\pi\mu\sigma_y\sigma_z}exp\left[\left(-rac{y^2}{2\sigma_y^2} ight) ight]\left\{exp\left[-rac{(z-H_e)^2}{2\sigma_z^2} ight] + aexp\left[-rac{(z-H_e)^2}{2\sigma_z^2} ight] ight]$	Stability parameter $R_y$ (class C)	-	0.230	Seinfeld and Pandis (2016)
$\left[\frac{\left(z+H_{e} ight)^{2}}{2\sigma_{z}^{2}}\right]$ $exp\left[-\lambda\frac{x}{\mu}\right]$	Stability parameter $r_y$ (class C)	-	0.855	Seinfeld and Pandis (2016)
where $\sigma_y = R_y x^{r_y}$ $\sigma_z = R_z x^{r_z}$	Stability parameter $R_z$ (class C)	-	0.076	Seinfeld and
02 - 1124	Stability parameter $r_z$ (class C)	-	0.879	Pandis (2016) Seinfeld and
	Center distance from emission source	m	110	Pandis (2016) Assumed
	(x) Lateral distance from emission source (y)	m	0	Assumed
	Height of inhalation zone (z)	m	1.5	Assumed
	Effective height of CT $(H_e)$	m	10	Assumed
	Reflection term $(\alpha)$ Inactivation rate of $L$ . pneumophila in	$s^{-1}$	-1 ≤ 30 s: 0.12 >	Assumed
	evaporated aerosols ( $\lambda$ )	3	30 s: $7 \times 10^{-4}$	Katz and Hammel (1987
$D = C_{air}It\sum_{i}^{n}F_{i}DE_{i}$	Inhalation rate (I)	${ m m}^3$ ${ m minute}^{-1}$	0.015	Heyder et al. (1986)
				Epa (2011)
	Duration of exposure (t)	Minute	60	Assumed
	Fraction of organisms in aerosols of diameter $i$ for $i=1-10 \mu m$ ( $F_i$ )	%		Allegra et al.
	1		17.50	(2016)
	2		16.39	
	3		15.56	
	4 5		6.67 3.89	
			2.50	
	6		2.78	
	6 7		, -	
	7 8		5.00	
	7 8 9		5.00 5.28	
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i=1-10$	Fraction	5.00	Heyder et al.
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ )	Fraction	5.00 5.28 3.89	Heyder et al. (1986)
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ )	Fraction	5.00 5.28 3.89	
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ )	Fraction	5.00 5.28 3.89	
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ ) 1 2	Fraction	5.00 5.28 3.89 0.25 0.53	
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ ) 1 2 3 4 5	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52	
	7 8 9 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ ) 1 2 3 4 5 6	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52 0.40	-
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m $(DE_i)$ 1 2 3 4 5 6 7	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52 0.40 0.29	-
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m $(DE_i)$ 1 2 3 4 5 6 7	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52 0.40 0.29 0.19	
	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m ( $DE_i$ ) 1 2 3 4 5 6 7 8	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52 0.40 0.29 0.19 0.12	
(1) $P_{inf} = 1 - exp(-rD)$	7 8 9 10 Deposition efficiency of diameter aerosols $i$ on the alveolus for $i$ =1–10 $\mu$ m $(DE_i)$ 1 2 3 4 5 6 7	Fraction	5.00 5.28 3.89 0.25 0.53 0.62 0.61 0.52 0.40 0.29 0.19	

(continued on next page)

#### Table 2 (continued)

Equation	Parameter	Unit	Value	Reference
			41 10=5	Muller et al. (1983)
	Dose-response parameter for L. pneumophila, infection with clinical severity (r <sub>isc</sub> )	_	$4.1\times10^{-5}$	Armstrong and Haas (2007)
				Fitzgeorge et al. (1983)
	DALY for Legionnaires' disease (DALY/infection)	Fraction	0.97	van Lier et al. (2016)
	Exposure frequency (N)	Day	365	Assumed

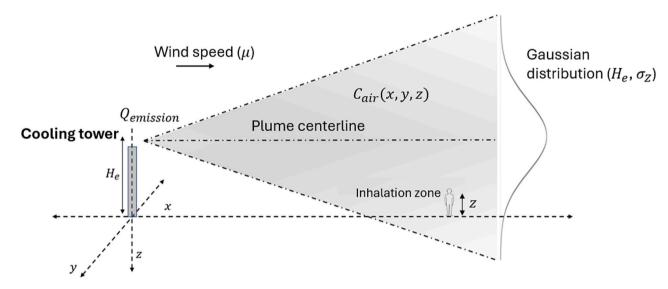


Fig. 2. Gaussian dispersion of microbial aerosols from a cooling tower. The diagram illustrates the airborne transport of <u>Legionella</u>-containing aerosols emitted from a cooling tower. The plume disperses downwind due to wind speed, following a Gaussian distribution characterized by vertical and lateral dispersion parameters. The concentration of airborne *L. pneumophila* at any location is governed by the dispersion model. The inhalation zone represents potential human exposure, relevant for the quantitative microbial risk assessment (QMRA) model.

dose–response models developed by Armstrong and Haas (2007) based on animal data from Muller et al. (1983) and Fitzgeorge et al. (1983) were used to predict infection with subclinical severity and infection with clinical severity. Clinical severity was defined as an infection requiring medical attention or seeking health services. The health impact of a clinically severe infection was assessed using the disability-adjusted life years (DALY) factor calculated by van Lier et al. (2016) based on surveillance data for Legionnaires' disease in the Netherlands (Dijkstra et al., 2010; Dijkstra et al., 2008).

## 2.10.3. Risk characterization

The health outcome target corresponds to the tolerable risk associated with exposure to L. pneumophila in contaminated aerosols a cooling tower produces. Infection risks and DALYs specific to each cooling tower in the database were assessed based on the arithmetic mean concentration of L. pneumophila in the water, estimated by parametric modelling. Risk estimates were compared to health-based targets of 1)  $10^{-4}$  infections per person per year (Macler and Regli, 1993) and 2)  $10^{-6}$  DALYs per person per year (Havelaar and Melse, 2003; World Health Organization (WHO), 2008).

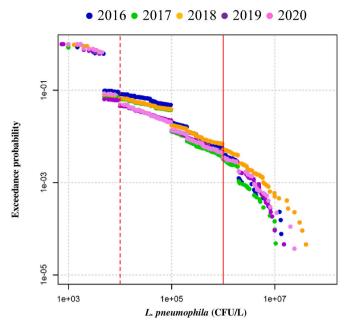
### 3. Results

The database aggregated 105,463 monitoring results documenting *L. pneumophila* concentrations in 2852 cooling towers from 1960 buildings in Quebec, Canada. Empirical complementary cumulative distribution functions (CCDFs) of monitoring results grouped by year

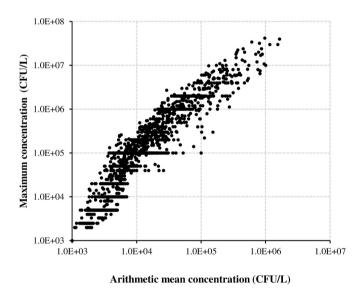
show exceedances of *L. pneumophila* concentration thresholds (Fig. 3). The  $10^4~{\rm CFU~L}^{-1}$  threshold has a steady exceedance probability of 10~% over the 2016–2020 period. The exceedance probability of the  $10^6~{\rm CFU~L}^{-1}$  threshold also remains constant at around 0.5 % for the whole period. Notably, the  $10^7~{\rm CFU~L}^{-1}$  threshold was exceeded more often in 2018 than in the other years.

The 5-year arithmetic mean concentration and the maximum concentration for each cooling tower were computed to assess their relationship. The log-log relationship between the maximum and the arithmetic mean concentrations demonstrates that peak concentrations can substantially impact the 5-year arithmetic mean (Fig. 4). The arithmetic mean is typically about 1.0 log lower than the sample maximum.

For 1461 cooling towers (about half of the total), all monitoring results are non-detects (Table 3). For 1099 cooling towers, 1 to 9 positive results are obtained. Results from the Poisson test reveal that *L. pneumophila* concentrations are statistically stable for roughly 50 % of these cooling towers, equivalent to around 20 % of the total. For the remaining 50 %, the limited number of positive results hinders accurate parameter estimation of the Poisson Gamma and Poisson lognormal distributions. Temporal variations could be assessed for the 292 cooling towers where the positive results count is ten or more (about 10 % of the total). Within this subset, the marginal deviance information criterion (mDIC) suggests the Poisson distribution as the most suitable model for ten cooling towers, indicating stable concentrations despite consistent positive findings. For the remaining 282 cooling towers, the mDIC can discriminate between the Poisson gamma and Poisson lognormal for 145



**Fig. 3.** Empirical complementary cumulative distribution function of *L. pneumophila* concentrations in bulk water from 2852 cooling towers in Quebec, Canada, from 2016 to 2020. The dotted and solid lines indicate concentrations of  $10^4$  and  $10^6$  CFU L $^{-1}$ , respectively.



**Fig. 4.** Relationship between the sample arithmetic mean concentration and the maximum concentration of *L. pneumophila* in bulk water from 2852 cooling towers in Quebec, Canada.

cooling towers, often favoring the Poisson lognormal. For the remainder, the mDIC indicates a similar quality of fits.

When results are Poisson distributed, arithmetic mean concentrations are consistently below  $10^4\, \text{CFU L}^{-1}$  (Table 4). Predictions from the

Poisson gamma generally indicate arithmetic mean concentrations below 10<sup>5</sup> CFU L<sup>-1</sup>. Arithmetic mean concentrations surpass 10<sup>6</sup> CFU L<sup>-1</sup> in 30 of the 90 cooling towers with optimal Poisson lognormal fits. This discrepancy underscores the significant impact of distribution selection on average concentration estimates. Moreover, the span of the 95 % uncertainty interval of the arithmetic mean predicted by the Poisson lognormal generally extends 1.0- to 2.0-log more than that of the Poisson gamma (Table S1, Fig. 5). The CCDFs show that the predictions of the lognormal distribution can extrapolate beyond the maximum observation, unlike the gamma distribution. The uncertainty interval of the CCDF remains stable for the gamma distribution but not for the lognormal distribution, expanding as exceedance probabilities diminish. For certain cooling towers, like Cooling tower D, the CCDF exhibits distinct tail behaviors at exceedance probabilities below 1 %. In such cases, the limited data size fails to adequately capture the upper tail's behavior, explaining the often-similar mDICs for Poisson gamma and Poisson lognormal fits.

QMRA results indicate that achieving a health-based target of 10<sup>-4</sup> inf./person-year requires maintaining the arithmetic mean L. pneumophila concentrations below 1.0E+03 CFU  $L^{-1}$  for the subclinical infection severity model and below 1.4E+06 CFU L<sup>-1</sup> for the clinical infection severity model (Table 5). The stricter health-based target of 10<sup>-6</sup> DALY/pers.-year results in a critical arithmetic mean concentration of 1.4E+04 CFU L<sup>-1</sup>. Results from the screening-level QMRA of 844 cooling towers are shown in Table 6. For both the arithmetic mean risk and its 97.5 % uncertainty bound, this table documents the number of cooling towers that achieve the target or not. For cooling towers where the discrimination between the Poisson gamma and Poisson lognormal was not possible, risks were estimated for both models. These results illustrate how uncertainty and statistical model choices can substantially influence risk assessment outcomes. Approximately 77 % of those 844 cooling towers meet the  $10^{-6}$  DALYs/pers.-year target. Using the 97.5 % uncertainty bound as an input for QMRA lowers compliance to about 70 %. Among the 562 cooling towers with stable concentration (Poisson distributed), the arithmetic mean risk and its upper uncertainty bound are generally below targets. For the Poisson gamma, roughly half of the cooling towers meet the 10<sup>-6</sup> DALYs/pers.-year. The remaining cooling towers exhibit arithmetic mean risks about 1.0-log above this target. Risks predicted by the Poisson lognormal tend to surpass 10<sup>-6</sup> DALYs/ pers.-year, with 60 cooling towers with risks exceeding the target by >2.0-log and 137 cooling towers with the uncertainty bound exceeding the target by >2.0-log.

#### 4. Discussion

# 4.1. Evolution of L. pneumophila concentration exceedance rates in Ouehec

In Quebec, the period from July 2014 to June 2017 saw a documented decrease in exceedance rates of  $10^4~\rm CFU~L^{-1}$  for approximately 300 cooling towers (Racine et al., 2019), coinciding with the introduction of the new regulation (Gouvernement du Québec, 2014). The average annual exceedance rate of  $10^4~\rm CFU~L^{-1}$  decreased from 15 % in 2014–2015 to 9 % in 2016–2017. For the 2016–2020 period, our results indicate stable exceedance rates of about 10 % for concentrations of  $10^4~\rm CFU~L^{-1}$  for the combined results of the 2852 cooling towers included in

**Table 3**Classification of cooling towers based on the decision algorithm to determine the best-fit model to assess *L. pneumophila* concentrations in bulk water, as shown in Fig. 1. Models are the Poisson distribution (Pois.), the Poisson gamma distribution (PGA), and the Poisson lognormal distribution (PLN).

Model	No positives $1-9$ positives $\geq 10$ positives				Total			
	n/a	Pois.	No Pois.	Pois.	PGA	PLN	Mixed Poisson <sup>A</sup>	
Total cooling towers	1461	552	547	10	55	90	137	2852
Fraction of total	51.2 %	19.3 %	19.1 %	0.3 %	1.9 %	3.1 %	4.8 %	100 %

A Discrimination between the fit of the PGA and PLN is not possible because the difference in DIC between the two models is less than three points.

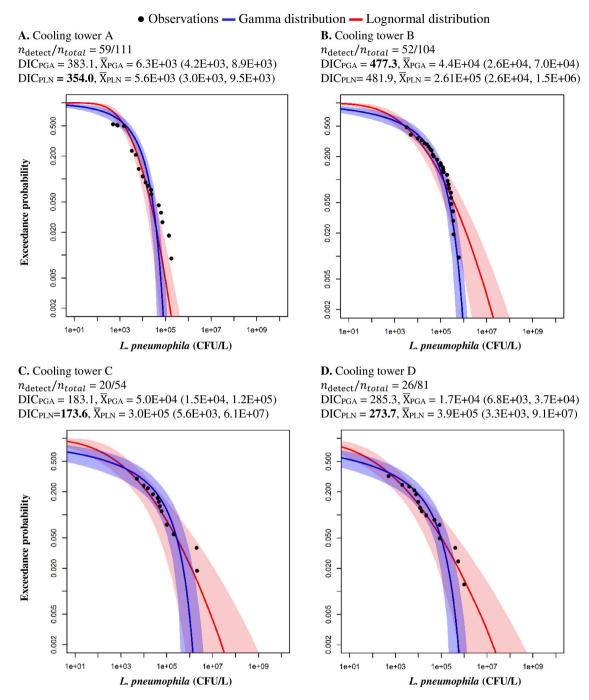


Fig. 5. Complementary cumulative distribution function of the Poisson-gamma (PGA) and Poisson lognormal (PLN) distributions fitted to L. pneumophila data from four cooling towers from the Quebec database. The dark blue and red lines represent the best fits of the gamma and lognormal distributions, respectively. The blue and red areas represent the 95 % uncertainty intervals of the gamma and lognormal distributions, respectively. The values of the marginal deviance information criterion (mDIC) and the arithmetic mean concentration (with 95 % uncertainty interval) in the bulk water of the cooling tower (CFU  $L^{-1}$ ) predicted by each model are listed. mDICs in bold indicate the best-fit models.

the database. The exceedance rates for concentrations of  $10^5$  CFU L $^{-1}$  and  $10^6$  CFU L $^{-1}$  also remained stable during this period, except for a notable increase in the exceedance rate of  $10^7$  CFU L $^{-1}$  in 2018. This increase may have been influenced by the extreme heat waves of summer 2018, potentially leading to increased cooling tower usage and operating conditions favorable to *L. pneumophila* growth. That summer was recorded as the hottest in 146 years of meteorological observations in southern Quebec (Lebel et al., 2019). Analyzing risk factors, such as air temperature, humidity level, and precipitation, could provide insights into the conditions that resulted in these extreme concentrations.

Overall, the introduction of Quebec's regulation appears to have initially reduced the high concentrations of L. pneumophila in Quebec's cooling towers but did not reduce the exceedance rates of  $10^4\,\mathrm{CFU\,L}^{-1}$  to the lower values of around  $1\,\%$  found in the Canadian federal cooling tower databases (data not shown). Gathering specific information on the water treatment strategies used in these cooling towers, including the types of biocides, application frequencies, and dosages, could shed light on the causes of the differences observed between these datasets.

Table 4
Classification of 844 cooling towers based on their arithmetic mean *L. pneumophila* concentration in bulk water. The average concentration was predicted using the best-fit model determined using the decision algorithm shown in Fig. 1. The models are the Poisson distribution, the Poisson gamma distribution (PGA), and the Poisson lognormal distribution (PLN).

Model	Arithmetic mean L. pr	Total			
	<104	10 <sup>4</sup> -10 <sup>5</sup>	$10^5 – 10^6$	>10 <sup>6</sup>	
Poisson (1–9 positives)	552 (100 %)	0 (0.0 %)	0 (0.0 %)	0 (0.0 %)	552 (100 %)
Poisson (≥10 positives)	10 (100 %)	0 (0.0 %)	0 (0.0 %)	0 (0.0 %)	10 (100 %)
PGA	11 (20.0 %)	35 (63.6 %)	9 (16.3 %)	0 (0.0 %)	55 (100 %)
PLN	15 (16.6 %)	26 (28.8 %)	19 (21.1 %)	30 (33.3 %)	90 (100 %)
Mixed Poisson <sup>A</sup> - PGA	81 (59.1 %)	48 (35.0 %)	8 (5.8 %)	0 (0.0 %)	137 (100 %)
Mixed Poisson <sup>A</sup> - PLN	45 (32.8 %)	34 (24.8 %)	26 (18.9 %)	32 (23.3 %)	137 (100 %)

A Discrimination between the fit of the PGA and PLN is not possible because the difference in DIC between the two models is less than three points.

#### 4.2. Navigating model uncertainty in L. pneumophila risk assessment

These findings highlight the importance of addressing uncertainty and carefully selecting the appropriate statistical model when predicting *L. pneumophila* concentrations for risk assessment. In particular, the wide uncertainty intervals observed with lognormal models underscore how inadequate characterization of tail behaviors can lead to underestimating public health risk. Selecting an empirical point estimate or a single distribution without sufficient comparison or validation can overlook rare but high-magnitude concentration events that can significantly impact public health risks. Consequently, risk management decisions may rely on model assumptions that mask the true variability in *L. pneumophila* concentrations. This study demonstrates how systematically evaluating and comparing different models allows risk assessors to make the most of available monitoring data, better account for uncertainty, and improve the reliability of QMRA outcomes.

Our analysis reveals that neither the gamma nor the lognormal distribution consistently outperforms the other in predicting variations, and in many cases, identifying the most accurate model was not feasible, likely due to limited sample sizes. When discrimination between the two distributions is not possible, we recommend selecting the lognormal to conservatively predict peak concentrations. The presence of these two distinct underlying generation processes (i.e., gamma and lognormal) suggests that L. pneumophila growth in cooling towers may follow either a logistic function or a Gompertz function (as detailed in Section 2.3). However, the apparent upper bound on maximum concentrations predicted by the gamma distribution may not necessarily represent a true biological carrying capacity of the logistic growth model but rather reflect the impact of interventions or adjustments to water treatment regimes following the exceedance of regulatory thresholds. Further research integrating statistical modelling with microbial growth dynamics could provide valuable insights for quantifying the impact of environmental and biological factors on L. pneumophila proliferation in full-scale engineered water systems.

Applying static parametric distributions to cooling tower data has limitations. Using such distributions for modelling *L. pneumophila* 

Table 5
Critical arithmetic mean *L. pneumophila* dose deposited at the alveoli and critical arithmetic mean *L. pneumophila* concentration in water of the cooling water to achieve three different annual health-based targets. Parameter values of the QMRA model used to calculate the doses and concentrations are presented in Table 2.

Health-based target	Average dose deposited at the alveoli (CFU)	Average $L$ . $pneumophila$ concentration in water (CFU $L^{-1}$ )
10 <sup>-4</sup> inf./persyear (subclinical severity)	4.8E-06	1.0E+03
10 <sup>-4</sup> inf./persyear (clinical severity)	6.8E-03	1.4E+06
10 <sup>-6</sup> DALY/persyear (clinical severity)	7.0E-05	1.4E+04

concentrations assumes the stochastic process is stationary (i.e., the process's structure is constant over time) and ergodic (i.e., the sample size is sufficiently large to reflect the process's structure). However, fulfilling these conditions in cooling towers presents challenges. A shift in the average concentration due to changes in water treatment may render the process non-stationary, potentially requiring different analyses for pre- and post-intervention data. Additionally, monthly monitoring over five years may not adequately represent the process. This is evidenced by the considerable parametric uncertainty in the L. pneumophila concentration distribution of some cooling towers. This result highlights the importance of accounting for short-term variations to predict public health risks. The causes of these fluctuations, such as L. pneumophila growth events or biofilm detachment, remain uncertain, limiting precise recommendations for monitoring. Nevertheless, our analysis suggests that the monitoring guidelines and regulations require revision for the risk management of high-risk cooling towers (e.g., those exceeding the 10<sup>4</sup> CFU L<sup>-1</sup> threshold). A more frequent monitoring interval, potentially bi-weekly or weekly, notably during periods of higher risk like warmer months, could be key to managing public health risks associated with these cooling towers.

### 4.3. Advancing QMRA for systems with high variability

The field of QMRA was initially developed to predict health risks associated with enteric pathogen concentrations in surface water. In such systems, the variability in concentrations is generally low enough to accurately estimate the long-term average pathogen concentration with low-frequency (e.g., monthly) monitoring over a set period (e.g., two years) (USEPA, 2010). Evaluating compliance with an annual health-based target can make sense for these systems. However, for systems with higher variability in pathogen concentrations, such as L. pneumophila in cooling towers, demonstrating compliance with an annual health-based target is challenging. For such systems, a short-term health-based risk target may be preferable, as previously recommended for drinking water safety management by Signor and Ashbolt, 2009. Our reverse-QMRA model indicates the critical annual average concentration of *L. pneumophila* should be  $1.4 \times 10^4$  CFU L<sup>-1</sup> to achieve a healthbased target of  $10^{-6}$  DALY/pers.-year, aligning closely with Quebec's threshold of 10<sup>4</sup> CFU L<sup>-1</sup>. Although the Quebec regulatory threshold was established "per sample," our findings demonstrate that concentrations exceeding  $10^4$  CFU  $\rm L^{-1}$  can substantially increase the annual average concentration. Therefore, maintaining "per sample" concentrations below 10<sup>4</sup> CFU L<sup>-1</sup> could be an effective strategy to maintain a critical annual average concentration of L. pneumophila of  $1.4 \times 10^4$  $CFU L^{-1}$ .

However, ensuring concentrations consistently remain below a set threshold with a defined confidence level demands detailed statistical analyses to determine the necessary monitoring frequency to capture *Legionella* growth, bloom, and sloughing events. The validity of a risk-based threshold also depends on the assumptions underlying the QMRA model. Our study's assumptions largely align with those of

Table 6
Classification of 844 cooling towers (CTs) based on their arithmetic mean infection risk and their upper uncertainty bound (97.5 %) of the arithmetic mean infection risk. Risks have been calculated using a screening-level quantitative microbial risk assessment (QMRA). Risks are compared to three different annual health-based targets. Models are the Poisson distribution, the Poisson gamma distribution (PGA), and the Poisson lognormal distribution (PLN).

Model (total cooling tower)	Infection risk level				Upper uncerta infection risk	Upper uncertainty bound (97.5 %) of the arithmetic mean infection risk			
		10 <sup>-4</sup> inf./ persyear	10 <sup>-4</sup> inf./persyear (clinical severity)	10 <sup>-6</sup> DALY/ persyear	10 <sup>-4</sup> inf./ persyear	10 <sup>-4</sup> inf./persyear (clinical severity)	10 <sup>-6</sup> DALY/ persyear		
Poisson 1–9 positives	Target achieved	552	552	552	544	552	552		
(552 CT)	Exceeded <1.0- log	0	0	0	8	0	0		
	Exceeded 1.0–2.0-log	0	0	0	0	0	0		
	Exceeded >2.0- log	0	0	0	0	0	0		
Poisson (≥10 positives)	Target achieved	10	10	10	7	10	10		
(10 CT)	Exceeded <1.0- log	0	0	0	3	0	0		
	Exceeded 1.0–2.0-log	0	0	0	0	0	0		
	Exceeded >2.0- log	0	0	0	0	0	0		
PGA (55 CT)	Target achieved	0	55	20	0	55	5		
	Exceeded <1.0- log	11	0	29	4	0	34		
	Exceeded 1.0–2.0-log	35	0	6	34	0	16		
	Exceeded >2.0- log	9	0	0	17	0	0		
PLN (90 CT)	Target achieved	0	60	20	0	27	3		
	Exceeded <1.0- log	14	13	23	1	11	8		
	Exceeded 1.0–2.0-log	26	6	17	7	12	16		
	Exceeded >2.0- log	50	11	30	82	40	63		
Mixed Poisson <sup>A</sup> - PGA	Target achieved	1	137	85	1	133	66		
(137 CT)	Exceeded <1.0- log	79	0	44	48	4	54		
	Exceeded 1.0–2.0-log	48	0	8	64	0	13		
	Exceeded >2.0- log	9	0	0	24	0	4		
Mixed Poisson <sup>A</sup> - PLN	Target achieved	3	107	52	1	62	19		
(137 CT)	Exceeded <1.0- log	42	12	34	13	17	22		
	Exceeded 1.0–2.0-log	34	10	21	23	4	22		
	Exceeded >2.0- log	58	8	30	100	54	74		

A Discrimination between the fit of the PGA and PLN is not possible because the difference in DIC between the two models is less than three points.

Hamilton et al. (2018). We diverged in assuming negligible reflection of viable bacteria from surfaces. Also, we found a fraction of aerosol mass with diameters of  $100~\mu m$  or smaller of 17~%, based on the distribution of Peterson and Lighthart (1977), which is above the 1.4~% reported by Hamilton et al. (2018). Assessing aerosol mass distributions emitted by more recently constructed cooling towers could enhance the accuracy of exposure models. Furthermore, developing dynamic QMRA models that account for temperature- and humidity-dependent L. pneumophila decay in aerosols, droplet evaporation, season-specific exposure patterns, and site-specific L. pneumophila concentrations in cooling towers could further improve the overall accuracy of exposure assessments.

# 4.4. Proactive strategies to reduce L. pneumophila exceedance thresholds

Under Quebec's regulations, exceeding the  $10^4~{\rm CFU~L^{-1}}$  threshold requires identifying the causes of the increase, implementing corrective measures, and assessing the efficacy of these measures. However, the effectiveness of these interventions appears limited, given the recurring exceedances of the  $10^4~{\rm CFU~L^{-1}}$  and  $10^6~{\rm CFU~L^{-1}}$  thresholds since 2016. To address this issue, adopting high-frequency  $\it L.$   $\it pneumophila$ 

monitoring following exceedances could enable early detection of peak concentrations, allowing for a better evaluation of the corrective measures. The implementation of comprehensive water management plans could shift focus from reactive to preventive measures, potentially reducing costs and resources associated with frequent monitoring following threshold exceedances.

#### 5. Conclusions

The analysis of an regulatory database, including  $\it L.$   $\it pneumophila$  concentrations monitored monthly from 2852 cooling towers across Quebec, Canada, between 2016 and 2020, led to the following conclusions:

The analysis of the 105,463 monitoring results shows that exceedance rates of the 10<sup>4</sup> and the 10<sup>6</sup> CFU L<sup>-1</sup> thresholds have remained constant at 10 % and 0.5 %, respectively. While the introduction of Quebec regulations in 2014 initially reduced threshold exceedances, this trend was not sustained from 2016 to 2020. Establishing validation procedures for corrective actions to prevent recurring

threshold exceedances could be an effective risk management strategy to address this issue.

- For the 2852 cooling towers, 51.2 % reported exclusively non-detects, 38.5 % reported between one to nine positives, and 10.2 % recorded more than ten positives. For this latter group, site-specific temporal variations in concentrations of *L. pneumophila* were often well described by either the gamma or the lognormal distribution. Due to the distinct behaviors of their upper tails, these distributions predicted considerably divergent arithmetic mean concentrations for some data sets. Implementing rigorous model comparison and selection approaches is essential to reliably predict peak concentrations.
- Our screening-level QMRA model suggests that maintaining a yearly average L. pneumophila concentration of  $1.4 \times 10^4$  CFU  $L^{-1}$  or lower is necessary to achieve a health-based target of  $10^{-6}$  DALY/pers. year for infections of clinical severity. QMRA identified 137 cooling towers at risk of exceeding this health-based target, primarily because of observed or predicted concentrations above  $10^5$  CFU  $L^{-1}$ . Therefore, maintaining "per sample" concentrations below  $10^4$  CFU  $L^{-1}$  could be an effective strategy for managing short-term clinical infection risks. Increasing the monitoring frequency would be necessary to enhance the identification and mitigation of L. pneumophila growth periods in these cooling towers.

Detailed information on water treatment practices at cooling towers (e.g., biocide types, application frequencies, and dosages) would be valuable in investigating design and operational factors contributing to *L. pneumophila* proliferation within cooling towers.

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### CRediT authorship contribution statement

Émile Sylvestre: Writing – review & editing, Writing – original draft, Visualization, Validation, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. Dominique Charron: Writing – review & editing, Validation, Project administration. Xavier Lefebvre: Writing – review & editing, Validation, Emilie Bedard: Writing – review & editing, Validation, Project administration, Funding acquisition. Michèle Prévost: Writing – review & editing, Validation, Supervision, Project administration, Methodology, Funding acquisition.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Data availability

The data that has been used is confidential.

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