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Declaration of competing interest

The authors declare no competing financial interests.

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Quantitative microbial risk assessment and sensitivity analysis for workers exposed to pathogenic bacterial bioaerosols under various aeration modes in two wastewater treatment plants
ABSTRACT

Wastewater treatment plants (WWTPs) could emit a large amount of bioaerosols containing pathogenic bacteria. Assessing the health risks of exposure to these bioaerosols by using quantitative microbial risk assessment (QMRA) is important to protect workers in WWTPs. However, the relative impacts of the stochastic input variables on the health risks determined in QMRA remain vague. Hence, this study performed a Monte Carlo simulation-based QMRA case study for workers exposing to *S. aureus* or *E. coli* bioaerosols and a sensitivity analysis in two WWTPs with various aeration modes. Results showed that when workers equipped without personal protective equipment (PPE) were exposed to *S. aureus* or *E. coli* bioaerosol in the two WWTPs, the annual probability of infection considerably exceeded the U.S. EPA benchmark (≤10E-4 pppy), and the disease burden did not satisfy the WHO benchmark (≤10E-6 DALYs pppy) (except exposure to *E. coli* bioaerosol for disease health risk burden). Nevertheless, the use of PPE effectively reduced the annual infection health risk to an acceptable level and converted the disease health risk burden to a highly acceptable level. Referring to the sensitivity analysis, the contribution of mechanical aeration modes to the variability of the health risks was absolutely dominated in the WWTPs. On the aeration mode that showed high exposure concentration, the three input exposure parameters (exposure time, aerosol ingestion rate, and breathing rate) had a great impact on health risks. The health risks were also prone to being highly influenced by the various choices of the dose—
response model and related parameters. Current research systematically delivered new data and a novel perspective on the sensitivity analysis of QMRA. Then, management decisions could be executed by authorities on the basis of the results of this sensitivity analysis to reduce related occupational health risks of workers in WWTPs.

**Keywords**

Quantitative microbial risk assessment; Sensitivity analysis; *Staphylococcus aureus*; *Escherichia coli*; Monte Carlo simulation; Personal protective equipment
1. Introduction

Wastewater treatment plants (WWTPs) could emit a large amount of bioaerosols containing pathogenic bacteria (Szyłak-Szydłowski et al., 2016). Compared with other workers, workers in WWTPs have a particularly higher prevalence of the so-called “sewage worker’s syndrome,” characterized by fatigue, headache, dizziness, gastrointestinal symptoms, and respiratory symptoms (Hung et al., 2010). These symptoms could be caused by work-related exposure to various bacterial bioaerosols that were liberated in wastewater treatment processes (Kowalski et al., 2017). Staphylococcus aureus and Escherichia coli bioaerosols, which had been frequently found in domestic wastewaters, are widely used as target indicator pathogens (Ikehata, 2013; Szyłak-Szydłowski et al., 2016; Shi et al., 2018; Kozajda et al., 2019). These bacteria from wastewater or sludge can infect people through inhalation (Szyłak-Szydłowski et al., 2016). Direct exposure to these bioaerosols causes gastrointestinal infection through bioaerosol capture in the upper respiratory tract by inhalation, where pathogenic bacterial bioaerosols move by ciliary action and pass into the digestive tract through the pharynx (Peccia et al., 2008). In general, the exposure of humans to WWTPs with pathogenic bacterial bioaerosols has significant health risks (Kozajda et al., 2019). Therefore, assessing the health risks of exposure to pathogenic bacterial bioaerosols is important to protect workers in WWTPs. In addition, the aeration mode in WWTPs and using personal protective equipment (PPE) could considerably influence the health risks of workers (Brandi et al., 2000; Teixeira et al., 2013). Pasalari et al. (2019) measured high health risks for workers exposed to Rotavirus and Norovirus bioaerosols in a WWTP equipped with various aeration tanks. Jones (2020) reported an increased contribution to health risks for patient care exposed to COVID-19 of the inhalation scenario equipped without PPE.
Health risk is usually quantified by the annual probability of infection ($P_{(a)inf}$) and disease burden (DB) (Haas et al., 2014). The $P_{(a)inf}$ and DB of bioaerosol exposure could be estimated by quantitative microbial risk assessment (QMRA) (Haas et al., 2014; Shi et al., 2018; Esfahanian et al., 2019). QMRA commonly follows four classical working steps: (a) hazard identification, (b) exposure assessment, (c) dose–response assessment, and (d) risk characterization (Haas et al., 2014). Moreover, QMRA is often estimated from Monte Carlo simulations to assess the range and likelihood of the health risk quantitatively (Lim et al., 2013; Shi et al., 2018; Liu et al., 2019). Furthermore, for risk characterization, the two most widely used health risk benchmarks are the acceptable annual infection risk level proposed by the U.S. EPA ($\leq 10E^{-4}$ pppy) (2005) and the acceptable disability-adjusted life years (DALYs) by the WHO ($\leq 10E^{-6}$ DALYs pppy) (2008). They are widely used in interpreting the magnitude of risk assessment outcomes (Lim et al., 2015; Fuhrimann et al., 2016; Shi et al., 2018). These two benchmarks were built around the concept of health-based targets that were grounded on well-defined health metrics (e.g., DALYs) and a level of tolerable health burdens (Fuhrimann et al., 2016).

In the QMRA, the importance of all input variables could be identified through a sensitivity analysis, which tests the relative impacts of stochastic input variables on health risks (Abhishek et al., 2008; Federigi et al., 2019). Sensitivity analysis is usually performed in QMRA to: (a) identify the most influential input variables on the output so as to propose feasible management recommendations to the authorities; (b)
improve the understanding and interpretation of the QMRA framework in order to extent of its analysis methodology; and (c) recognize input variable gaps and then prioritize future research priorities (Tesson et al., 2020). Haas et al. (2017) demonstrated health risk of *Ebolavirus* for sewer worker with or without PPE from inhalation exposure and sensitivity analysis from Monte Carlo simulation. Kowalski et al. (2017) analyzed the emission characterization of the bacteria and fungi bioaerosols collected in different aeration modes of WWTPs in Poland. Carducci et al. (2018) reported that the health risk for workers in WWTPs exposed to the human adenovirus (*HAdV*) was estimated by QMRA and the sensitivity analysis was employed to examine the impact of input parameters (breathing rate and concentration) on health risk.

However, given the ranking, significance, and contribution of these relative impacts remain vague, a series of open questions have been raised about the QMRA and its sensitivity analysis for stochastic input variables associated with workers using PPE and exposing under various aeration modes in WWTPs. Therefore, this research systematically investigates a Monte Carlo simulation-based QMRA case study for workers exposing to *S. aureus* and *E. coli* bioaerosols in two WWTPs. After that, the health risks (*P_{a,inf}*, and DB) of the workers without or with PPE exposed to bioaerosols under various aeration modes in two WWTPs were discussed. Then, it focuses on the rank correlation coefficient values and contribution to variance of each input variable in QMRA which were assessed by sensitivity analysis. The current
research can enrich the knowledge bases of the sensitivity analysis of QMRA for workers with PPE exposed to bioaerosols under various aeration modes in WWTPs and then provide an advanced understanding of the rank correlation coefficient values and contributions to variance of each input variable in QMRA framework. These results can inform efforts to establish rational management recommendations for reducing occupational health risks of workers in WWTPs.

2. Materials and methods

2.1 Description of the WWTPs

This study was performed in two WWTPs (plants A and B), which were located in the central part of P.R. China. Their drainage pipe systems were similar. The collected domestic wastewater (occasionally mixed with a little industrial wastewater) was distributed into the WWTP by a series of variable-frequency pump stations. Plant A had a parallel wastewater treatment system equipped with a rotating disc aeration tank (RD) and a microporous aeration tank (M), treating 50,000 tons of wastewater per day, respectively. Similarly, plant B was also a parallel system. It had an inverted umbrella aeration tank (IU) and a microporous aeration tank (M), treating 100,000 tons of wastewater per day, respectively. Thus, there were three modes for aeration tanks (RD, IU, and M) in this research.

2.2 Sampling and analysis

2.2.1 Sampling procedure

Six bioaerosol sampling campaigns were conducted on 21th November 2019, 5th December 2019, 16th December 2019, 23rd December 2019, 7th January 2020, and 8th January 2020 in plants A and B by using an Andersen six-stage cascade impactor
Sterile agar media Egg-Yolk Mannitol Salt Agar Base and MacConkey-Agar-Medium were used as the collection media for culturing and colony enumeration of *S. aureus* and *E. coli*, respectively (Oppliger et al., 2005; Szyłak-Szydlowski et al., 2016; Nasir et al., 2018; Wang et al., 2019). A 27 mL aliquot of this sterile agar media (autoclaved at 121 °C for 15 min) was pipetted into sterile glass Petri dishes equipped with the cascade impactor (Jahne et al., 2015; Jahne et al., 2016).

The sampling point was set at 1.5 m above each aeration tank’s ground (Szyłak-Szydlowski et al., 2016). The cascade impactor was operated for 10 min at a flow rate of 28.3 L/min (Hung et al., 2010; Kowalski et al., 2017). Each stage of the Andersen six-stage cascade impactor was decontaminated with 75% alcohol before and after use for air sampling on site (Hung et al., 2010). All samples were in triplicate and transported to the laboratory in a cold box before being cultivated in incubators for 24–48 h at 37 °C.

2.2.2 Colony enumeration

After cultivation, the samples were enumerated as colony-forming unit (CFU) by using an automatic colony enumeration instrument (HICC-B, Wanshen Inc., Hangzhou, China). The positive hole method was used to correct and then obtain the actual number of colonies measured at the each Petri dish stage on the basis of the enumeration results (Hung et al., 2010; Delort et al., 2017). Bioaerosol concentrations of *S. aureus* and *E. coli* in CFU/m³ were estimated by dividing the number of colonies in CFU by the sampled air volume in m³ (Hung et al., 2010). Then, the bioaerosol concentration was the sum of the concentrations of the six Petri dish stages of the Andersen six-stage cascade impactor (Katsivela et al., 2017).
2.3 Quantitative microbial risk assessment framework

2.3.1 Hazard identification

The indicator pathogens of concern in this research were *S. aureus* and *E. coli* bioaerosols in the two WWTPs. So, the workers in the WWTP aeration tanks were exposed to serious *S. aureus* and *E. coli* bioaerosols-related health risks.

[Table 1 inserts here]

[Figure 1 inserts here]

2.3.2 Exposure assessment

The parameters and flow chart for the exposure assessment referring to the QMRA calculation framework are presented in Table 1 and Figure 1, respectively. This research had eight exposure scenarios (Fig. 1): (a) workers without PPE exposed to *S. aureus* bioaerosol in plant A, (b) workers without PPE exposed to *S. aureus* bioaerosol in plant B, (c) workers with PPE exposed to *S. aureus* bioaerosol in plant A, (d) workers with PPE exposed to *S. aureus* bioaerosol in plant B, (e) workers without PPE exposed to *E. coli* bioaerosol in plant A, (f) workers without PPE exposed to *E. coli* bioaerosol in plant B, (g) workers with PPE exposed to *E. coli* bioaerosol in plant A, and (h) workers with PPE exposed to *E. coli* bioaerosol in plant B. The exposure concentrations (*ec*) of *S. aureus* and *E. coli* bioaerosols are calculated and shown in Supplementary Materials Table 1. The aerosol ingestion rate (*ag*) is shown in Supplementary Materials Table 2.

The removal fraction by employing PPE (*F_{PPE}*) was conducted in two situations (the two exposure groups in Fig. 1): (a) workers in WWTPs used no face protection
(i.e., workers without PPE) and (b) workers in WWTPs wore a properly fitted N-95 respirator at all times (i.e., workers with PPE) (Haas et al., 2017).

The dose of pathogens (Dose) per person per day was calculated in Equation (1) (Dungan, 2014; Jahne et al., 2015; Haas et al., 2017):

\[
Dose = 10^{ec} \times br \times t \times ag \times (1 - F_{PPE})/1000,
\]

(1)

where Dose represents the dose of bioaerosol inhaled per person per day (CFU/day), \( ec \) is the exposure concentration (Supplementary Materials Table 1), \( br \) is the breathing rate, \( ag \) is the aerosol ingestion rate (Supplementary Materials Table 2), and \( F_{PPE} \) is the removal fraction by employing PPE (Table 1).

2.3.3 Dose–response models

For *S. aureus* bioaerosol, the exponential dose–response model as a dose–infection model was used to determine the relationship between the dose and the infection risks (Equation (2)) (Esfahanian et al., 2019):

\[
P_{(d)inf} = 1 - \exp(-k \times Dose),
\]

(2)

where \( P_{(d)inf} \) is the estimated daily probability of infection, and \( k \) is the parameter of the model (Table 1).

For *E. coli* bioaerosol, the Beta–Poisson dose–response model as a dose–infection model was used to determine the relationship between the dose and the infection risks, which is shown in Equation (3) (Shi et al., 2018):

\[
P_{(d)inf} = 1 - (1 + Dose^{2\alpha - 1}/N_{50})^{-\alpha},
\]

(3)

where \( P_{(d)inf} \) is the estimated daily probability of infection, and \( \alpha \) and \( N_{50} \) are the parameters of the model (Table 1).
2.3.4 Risk characterization

Risk characterization was carried out on the basis of the contaminant concentration to which individuals were exposed. Annual probability was estimated considering the number of exposure events per year with Equation (4) (Haas et al., 2014; Salesortells et al., 2014):

$$P_{(a)inf} = 1 - (1 - P_{(d)inf})^n,$$

(4)

where $P_{(a)inf}$ is the annual probability of infection per person per year (pppy), and $n$ is the annual exposure frequency (Table 1).

For $S. \text{aureus}$ bioaerosol, the probability of infection was assumed equal to the probability of illness ($P_{aill} = 1$). The probability of illness, as a conditional of infection, was calculated in Equation (5) (Busgang et al., 2018; Carducci et al., 2018):

$$P_{(a)ill} = P_{(a)inf} \times P_{ill/inf},$$

(5)

where $P_{(a)ill}$ is the annual probability of illness, and $P_{ill/inf}$ is the specific conditional probability of illness given an infection (i.e., prevalence) (Table 1).

For $E. \text{coli}$ bioaerosol, the exponential dose–response model was used as a dose–illness model to calculate the probability of illness, which was defined in Equation (6) (Shi et al., 2018):

$$P_{(a)ill} = 1 - \exp(-k \times Dose),$$

(6)

where $P_{(a)ill}$ is the annual probability of illness, and $k$ is the parameter of the model, which are listed in Table 1.

The specific potential disease burden attributable to illness caused by exposure to $S. \text{aureus}$ or $E. \text{coli}$ bioaerosol was estimated in Equation (7) (Havelaar et al., 2012; Shi et al., 2018):
\[ DB = HB \times P_{(a)\text{ill}}, \] (7)

where \( DB \) is the disease burden and expressed in DALYs pppy, and \( HB \) is the health burden and expressed in DALYs per illness case (DALYs/case) (Table 1).

2.4 Monte Carlo simulation

Monte Carlo simulation was used to represent the propagation of variability in QMRA (Lim et al., 2015). It was run with 10,000 trials by using Oracle Crystal Ball and Microsoft Excel 2010 (Devleesschauwer et al., 2014; Wass et al., 2017; Liu et al., 2019). All inputted variables (exposure concentration, three input exposure parameters (exposure time, aerosol ingestion rate, and breathing rate), the removal fraction by employing PPE, and the model parameters of the dose–response model) were randomly selected from their probability distributions. Output health risks were computed over 10,000 iterations so that the distributions would reach a steady state (Lim et al., 2013; Shi et al., 2018). The results of Monte Carlo simulation were shown by a box-and-whiskers chart. The lower whisker in the box chart represented optimistic estimate. The non-conservative estimate was originated from 25th percentile values to the lower whisker.

2.5 Sensitivity analysis

The rankings of each inputted variables were assessed using a sensitivity analysis with Oracle Crystal Ball. The significance of each parameter was characterized by its correlation coefficient values with the health risks, where a higher value (i.e. high ranking) indicated greater contribution (i.e., great impact) to the variability of the health risks and vice versa (Hamilton et al., 2006; Lim et al., 2013; Vásquez et al., 2014; Pang et al., 2017; Pasalari et al., 2019).
Contribution to variance was calculated by squaring the rank correlation coefficient values and normalizing them to 100% (Zhou et al., 2014; Haas et al., 2017; Shi et al., 2018). Contribution to variance showed sensitivities as values that range from 0 to 100% and indicated relative importance by showing the percentage of the variance of the predicted variable contributed by each dose–response model input variable (Zhou et al., 2014; Haas et al., 2017; Shi et al., 2018).

3. Results and discussion

3.1 Dose–response assessment and risk characterization

Figure 2 demonstrates the annual infection risks ($P_{(a)\text{inf}}$) and the disease burdens (DB) that were estimated from the Monte Carlo simulations with 10,000 iterations under the eight exposure scenarios where workers (without or with PPE) were exposed to $S.\text{aureus}$ or $E.\text{coli}$ bioaerosols in the two WWTPs.

For exposing to $S.\text{aureus}$ bioaerosol, the $P_{(a)\text{inf}}$ of the workers in plant A were always much higher than that of the workers in plant B (Fig. 2a). This finding could be explained by the theory that the different aeration modes between the two WWTPs lead to huge differences in the concentration of $S.\text{aureus}$ bioaerosol emissions, which would largely affect the annual infection health risks for workers (Haas et al., 2014; Dungan, 2014; Jahne et al., 2015). Nevertheless, the $P_{(a)\text{inf}}$ of the workers without PPE in plants A and B both considerably exceeded the U.S. EPA benchmark ($\leq 10E-4$ pppy) (Fig. 2a). However, the $P_{(a)\text{inf}}$ of the workers with PPE in plant A (median = 6.04E-04) was on the same order of magnitude as the benchmark, and the $P_{(a)\text{inf}}$ of the workers with PPE in plant B clearly satisfied the benchmark. These results indicated that using
PPE can effectively reduce the annual infection health risks of *S. aureus* bioaerosol to an acceptable level (Ikehata, 2013; Hass et al., 2017; Carducci et al., 2018).

For *E. coli* bioaerosol, the $P_{(a)inf}$ of the workers without or with PPE in plant A slightly differed from that of the workers in plant B (Fig. 2a). Furthermore, the $P_{(a)inf}$ of the workers without PPE in the two plants both considerably exceeded the U.S. EPA benchmark ($\leq 10^{-4}$ pppy). However, the $P_{(a)inf}$ values of the workers with PPE in plant A (median = 4.36E-04) and plant B (median = 2.88E-04) were just on the same order of magnitude as the benchmark. The annual infection health risk of the workers was even acceptable under the optimistic estimate because the lower whisker of the $P_{(a)inf}$ of the workers with PPE satisfied the benchmark. This result disclosed that the use of PPE reduced the annual infection health risks of *E. coli* bioaerosol in the two plants to an acceptable level, but the risks were still far from negligible.

For *S. aureus* bioaerosol, the DB of the workers in plant A was much higher than that of the workers in plant B (Fig. 2b). The DB of the workers without PPE in plant A significantly exceeded the WHO benchmark ($\leq 10^{-6}$ DALYs pppy). However, the DB values of the workers with PPE in plant A (median = 1.57E-06) and that of the workers without PPE in plant B (median = 1.67E-06) were roughly on the same order of magnitude as the benchmark. Under non-conservative estimate, the disease health risk burden of those workers was even acceptable since the benchmark was satisfied by the DB from the 25th percentile values to the lower whisker. Furthermore, the DB of the workers with PPE in plant B completely satisfied the WHO benchmark. Therefore, wearing of PPE improved the disease health risk burden of workers exposed to *S. aureus* bioaerosol from low acceptable level to high acceptable level.
For *E. coli* bioaerosol, the DB of the workers in plant A was similar to that of the workers in plant B. However, all DBs of the workers without or with PPE in the two plants satisfied the WHO benchmark. This result can be ascribed to the fact that the dose–illness model used in *E. coli* bioaerosol QMRA made the calculated DB demonstrate a non-conservative health risk estimate and therefore fulfilled the WHO benchmark (Shi et al., 2018). Thus, even without PPE, the disease health risk burden of the workers exposed to *E. coli* bioaerosol was still acceptable. Similar result had been reported. Shi et al. (2018) found that even in the worst-case scenario, where all *E. coli* bioaerosols were assumed to be pathogenic, the health risks were still far below the benchmark.

What was noteworthy was that, as expected, the health risks (P_{(a)inf} and DB) of the workers exposed to *S. aureus* bioaerosol with PPE reduced by approximately two orders of magnitude compared with those of the workers without PPE in plants A and B. This result was because the N-95 respirators utilized in this research were engineered to filter at least 95% of the particles that would be inhaled (Hass et al., 2017). The results of the reduction of health risk of workers with PPE exposed to *E. coli* bioaerosol were similar.

P_{(a)inf} and DB were adopted as health risk indicators throughout the analysis in this research, considering that the DALYs approach can add values to health risk management (Haas et al., 2014; Lim et al., 2015; Shi et al., 2018). However, DALYs might be blighted by the lack of data to support its development in China because of its rare local practical application. Therefore, DALYs data specific to China were
thought to be less readily available. Lim et al. (2015) put forth that the U.S. EPA $P_{(a)_{inf}}$ benchmark is regionally bounded because it was proposed according to the disease surveillance data only in the U.S. Therefore, this benchmark might not be representative of the whole world. Moreover, the WHO DB benchmark should be treated cautiously in a similar manner to the U.S. EPA $P_{(a)_{inf}}$ benchmark, and these two indicators ought to be used as complements rather than opposites in health risk assessment (Lim et al., 2015). In addition, the U.S. EPA $P_{(a)_{inf}}$ benchmark and the WHO DB benchmark are considered to be overly conservative (Lim et al., 2015).

In this research, the $P_{(a)_{inf}}$ and DB were calculated by using different dose–response models for the QMRA of *S. aureus* and *E. coli* bioaerosols (Shi et al., 2018). For *S. aureus* bioaerosol QMRA, the metrics used for $P_{(a)_{inf}}$ and DB were directly related to each other, and the DB was calculated via $P_{(a)_{inf}}$ and DALYs (Havelaar et al., 2012; Busgang et al., 2018). By contrast, the correlation of the dose–response models for *E. coli* bioaerosol QMRA led to the variability of the health risk calculations. $P_{(a)_{inf}}$ was calculated using the Beta–Poisson dose–response model (Equations (3) and (4)), and the DB was calculated using the exponential dose–response model (Equations (6) and (7)). Thus, this research implied that the health risks ($P_{(a)_{inf}}$ and DB) were prone to being highly influenced by the various dose–response models of choice. In general, accurate health risk estimation called for additional field studies and clinical infection data (Shi et al., 2018). But there remain also need to understand that an efficient and rigorous validation of the dose–response model and its relevant parameters for QMRA is warranted (Haas, 2015).

[Figure 3 inserts here]

[Figure 4 inserts here]
3.2 Sensitivity analysis for quantitative microbial risk assessment results

Figure 3 shows the input variable ranking of the sensitivity of the exposure concentration, the three input exposure parameters (exposure time, aerosol ingestion rate, and breathing rate), the removal fraction by employing PPE, and the dose-response model parameters to the health risks ($P_{\text{a} \text{inf}}$ and DB). Each aeration mode was individually analyzed in the Figure 3. Figure 4 demonstrates the contribution to variance of the input variables that impact the output value of the health risks.

For *S. aureus* bioaerosol, the exposure concentration for workers (with or without PPE) on mechanical aeration modes (the rotating disc aeration tank (RD) in plant A or the inverted umbrella aeration tank (IU) in plant B) was the most sensitive to the health risks (Figs. 3a, 3b, 3c, and 3d). On the RD aeration tank in plant A, the exposure concentration for workers contributed the maximum variability of health risks associated with *S. aureus* bioaerosol. Among workers without or with PPE, the contribution to variance of the exposure concentration accounted for 43.62% or 30.06%, respectively (Figs. 4a and 4c). The exposure time, aerosol ingestion rate, and breathing rate on the RD aeration tank showed lower input variables ranking than the exposure concentration (Figs. 3a and 3c). On the RD aeration tank, the fraction of the contribution to variance of the exposure concentration was approximately 2, 6, and 7 times as large as the exposure time, the aerosol ingestion rate, and the breathing rate, respectively (Figs. 4a and 4c). The exposure concentration and the three input exposure parameters on the RD aeration tank were all more sensitive than those on the microporous aeration tank (M) in plant A (Figs. 3a and 3c). This result disclosed that on the aeration mode, which characterized high exposure concentration, the three
input exposure parameters had a great impact on the workers’ health risks. In plant B, the exposure concentration for the workers on the IU aeration tank showed a large contribution to the health risks, accounting for 31.22% (without PPE) or 22.6% (with PPE) (Figs. 4b and 4d). Moreover, the contribution to variance of the exposure concentration for the workers on the M aeration tank exerted minor effect on the health risk with fraction >10% (Figs. 4b and 4d). The three input exposure parameters (exposure time, aerosol ingestion rate, and breathing rate) for the workers on the IU aeration tank all showed a slightly higher ranking than those on the M aeration tank (Figs. 3b and 3d). These results reflected that the contribution of mechanical aeration modes to the variability of the health risks was absolutely dominated in the two WWTPs, especially for the contribution of the RD aeration tank in plant A. The ranking of the dose–response model parameters was just lower than those of the exposure concentration, the exposure time, and the removal fraction by employing PPE. The contribution of choice of the dose–response model parameters to health risks was far from negligible.

For *E. coli* bioaerosol, the rankings of the input variables were nearly the same as those for *S. aureus* bioaerosol. The exposure concentration on mechanical aeration modes, rather than mode of the microporous aeration tanks, accounted for most of the health risk’s variability, with the fraction >40% (without PPE) or >30% (with PPE) (Figs. 4e, 4f, 4g, and 4h).

This result could be due to the fact that the microporous aeration mode completely differs from the mechanical aeration mode (Li et al., 2016). Several studies reported similar results. Stellacci et al. (2010) detected bioaerosols in the WWTP, which is often related to the surface mechanical aeration bioreactors. Another study in Poland obtained comparable results with this research, and they explained that the blast
aeration technology with microporous aerators does not cause any large turbulence because it is situated at the bottom of the tank and accordingly does not generate large amounts of bioaerosols as the mechanical aeration mode did (Gotkowska-Plachta et al., 2013; Li et al., 2016).

When the PPE employed workers exposing to the mechanical aeration tanks in plants A and B, the input variable “removal fraction by employing with PPE” contributed the second ranking for health risks. This result illustrated that the PPE can largely affect health risks associated with bioaerosol (Haas et al., 2017; Carducci et al., 2018). Therefore, workers exposed to the mechanical aeration modes are strongly suggested to wear PPE. However, the effects of employing PPE on the M aeration tank in plants A and B showed weaker impact on the variability of the health risks. This result disclosed that the microporous aeration mode did not exert obvious effects on the health risks of the workers wearing PPE as large as that on the mechanical aeration modes. This finding is consistent with previous studies that QMRA could be used to indicate the most suitable scenario to employ PPE by considering its efficiency of protection (Carducci et al., 2018). In addition, the effective use of PPE can significantly decrease the worker’s health risks (Ikehata, 2013; Haas et al., 2017).

4. Conclusion

The P_{a,inf} of the workers equipped without PPE exposed to S. aureus or E. coli bioaerosols in the two WWTPs considerably exceeded the U.S. EPA benchmark (≤10E-4 pppy), and the DB also did not satisfy the WHO benchmark (≤10E-6 DALYs pppy) except exposure to E. coli bioaerosol for disease health risk burden. However, the use of PPE can effectively reduce the annual infection health risk to an acceptable level and convert the disease health risk burden to a high acceptable level. In general,
the health risks ($P_{\text{adjinf}}$ and DB) of the workers with PPE were reduced approximately two orders of magnitude compared with those of the workers without PPE. The PPE could largely affect the health risk associated with bioaerosol, especially on the mechanical aeration modes. In addition, the different aeration modes between the two WWTPs led to the higher health risks of the workers in plant A than those of the workers in plant B. Under exposure to *S. aureus* bioaerosol, the contribution of mechanical aeration modes to the variability of the health risks was absolutely dominated in the WWTPs, especially the contribution of the KD aeration tank in plant A. The exposure concentration of the workers exposed to *E. coli* bioaerosol on the mechanical aeration modes, rather than the microporous aeration mode, accounted for most of the health risk variability. Therefore, the mechanical aeration should be managed as priority. On the aeration mode characterized with high exposure concentration, the three input exposure parameters (exposure time, aerosol ingestion rate, and breathing rate) had a great impact on health risks. Of note, the health risks were also prone to being highly influenced by the various choices of the dose–response model and related parameters. Therefore, accurate health risk estimation called for additional field studies and clinical infection data, and the dose–response model should be chosen discreetly.

This research systematically delivered new data and a novel perspective on the sensitivity analysis of QMRA for workers with PPE exposed to pathogenic bacterial bioaerosols under various aeration modes. Furthermore, it significantly aided in advancing the understanding of the rank correlation coefficient values and contributions to variance of each input variable in QMRA. Then, management decisions can be implemented by authorities on the basis of the results of the sensitivity analysis for the workers to abate the related occupational health risks.
Generally, this research could be an educational tool to fill the gap between the QMRA framework and feasible rational management recommendations and to offer proposals that could be executed by authorities to protect public health.
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**CRediT author statement**

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Declaration of interests

☒ 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.

☐ The authors declare the following financial interests/personal relationships which may be considered as potential competing interests:
Figure 1 Flow chart of quantitative microbial risk assessment

Figure 2 Box-and-Whiskers Diagram showing (a) annual infection risks ($P_{\text{a,inf}}$) and (b) disease burdens (DBs) under eight exposure scenarios of workers (without or with PPE) exposed to *S. aureus* or *E. coli* bioaerosols in the two wastewater treatment plants.

The bottom and top of the box represent the first and third quartiles (25th and 75th percentile values), the band inside the box represents the second quartile (median), and the tetragon inside the box represents the average value. The whiskers extend 1.5 interquartile ranges (75th percentile value–25th percentile value) from each end of the box, and markers plotted outside each whisker are considered as outliers.

*S. aureus*= *Staphylococcus aureus*

*E. coli*= *Escherichia coli*

PPE= Personal Protective Equipment

U.S. EPA= United States Environmental Protection Agency

WHO= World Health Organization
Figure 3 Tornado graphs to display the ranking of input variables that impact the output value for workers (without or with PPE) exposed to *S. aureus* or *E. coli* bioaerosols in various aeration tanks of the two wastewater treatment plants referring to (a) workers without PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant A, (b) workers without PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant B, (c) workers with PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant A, (d) workers with PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant B, (e) workers without PPE exposed to *E. coli* bioaerosol in wastewater treatment plant A, (f) workers without PPE exposed to *E. coli* bioaerosol in wastewater treatment plant B, (g) workers with PPE exposed to *E. coli* bioaerosol in wastewater treatment plant A, and (h) workers with PPE exposed to *E. coli* bioaerosol in wastewater treatment plant B.

Correlation coefficient values were obtained from @ Oracle Crystal Ball sensitivity analyses and are shown next to each bar.

*S. aureus* = *Staphylococcus aureus*

*E. coli* = *Escherichia coli*

PPE = Personal Protective Equipment
Figure 4 Pie charts showing the contribution to variance of input variables that impact the output value for workers (without or with PPE) exposed to *S. aureus* or *E. coli* bioaerosols in various aeration tanks of the two wastewater treatment plants referring to (a) workers without PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant A, (b) workers without PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant B, (c) workers with PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant A, (d) workers with PPE exposed to *S. aureus* bioaerosol in wastewater treatment plant B, (e) workers without PPE exposed to *E. coli* bioaerosol in wastewater treatment plant A, (f) workers without PPE exposed to *E. coli* bioaerosol in wastewater treatment plant B, (g) workers with PPE exposed to *E. coli* bioaerosol in wastewater treatment plant A, and (h) workers with PPE exposed to *E. coli* bioaerosol in wastewater treatment plant B.

Contribution to variance values were obtained from Oracle Crystal Ball sensitivity analyses and are shown next to each pie.

*S. aureus* = *Staphylococcus aureus*

*E. coli* = *Escherichia coli*

PPE = Personal Protective Equipment

RD = Rotating disc aeration tank

IU = Inverted umbrella aeration tank

M = Microporous aeration tank
| Description                          | Unit                  | Value                        | Reference                  |
|-------------------------------------|-----------------------|------------------------------|----------------------------|
| Exposure concentrations (ec)        | \(\log_{10}\text{CFU/m}^3\) | Supplementary Materials Table 1 Uniform distribution (Min=9.8; Max=13.0) | -                          |
| Breathing rate (br)                 | L/min                 | (Min=9.8; Max=13.0)          | MEP-P RC, 2013             |
| Exposure time (t)                   | min                   | Uniform distribution (Min=8; Max=20) | According to the field survey in this research. |
| Aerosol ingestion rate (ag)         | Unitless              | Supplementary Materials Table 2 Uniform distribution (Min=0.95; Max=0.99) | -                          |
| Removal fraction by employing PPE (F_{PPE}) | Unitless              | Uniform distribution (Min=0.95; Max=0.99) | Haas et al., 2017          |
| Annual exposure frequency (n)       | Number of times       | 183                          | According to the field survey in this research. |
| Staphylococcus aureus bioaerosol    | Exponential dose–response model (dose–k) | Uniform (Min=6.46E−8; Max=1.00E−7) | Esfahani an et al., 2019    |
| Parameter                          | Formula | Unit     | Value     | Reference          |
|-----------------------------------|---------|----------|-----------|--------------------|
| Prevalence \( P_{il} \)          |         | Unitless | 1         | Busgang et al., 2018 |
| Health burden (HB) \( \text{DALYs/case} \) | \( 2.60 \times 10^{-3} \) | Unitless | 1         | Havelaar et al., 2012 |
| Beta-Poisson dose–response model \( \alpha \) |         | Unitless | 1.55 \times 10^{-1} | Shi et al., 2018 |
| Exponential dose–response model \( N_{50} \) |         | Unitless | \( 2.11 \times 10^6 \) | Shi et al., 2018 |
| Health burden (HB) \( \text{DALYs/case} \) | \( 4.55 \times 10^{-2} \) | Unitless | 1         | Shi et al., 2018 |

*Escherichia coli* bioaerosol
Even without PPE, DB of workers exposed to *E. coli* bioaerosol was still acceptable. The use of PPE effectively reduced the health risks to an acceptable level. With high exposure concentration, input exposure parameters highly impact health risk. Mechanical aeration modes’ contribution to health risks’ variability was dominated. Health risks were highly influenced by the various choices of the dose–response model.
1. Hazards identification

*Staphylococcus aureus* bioaerosol  *Escherichia coli* bioaerosol

2. Exposure assessment

| Exposure groups                  | Wastewater treatment plant A          | Wastewater treatment plant B          |
|----------------------------------|--------------------------------------|--------------------------------------|
| Worker without personal protective equipment (PPE) | Rotating disc aeration tank | Microporous aeration tank  |
| Worker with PPE                  | Inverted umbrella aeration tank      | Microporous aeration tank  |

| Exposure sites                  | Wastewater treatment plant A          | Wastewater treatment plant B          |
|---------------------------------|--------------------------------------|--------------------------------------|
| Exposure concentrations (ec)     | Workers exposed to the bioaerosols in the exposure sites |  |
| Breathing rate (br)              | Uniform distribution (Min=9.8 L/min; Max=13.0 L/min) |  |
| Exposure duration (t)            | Uniform distribution (Min=8 min; Max=20 min) |  |
| Aerosol ingestion rate (ag)      | Uniform distribution (Min=proportion of 3-6 stages; Max=1) |  |
| Removal fraction by employing PPE (F_PPE) | Uniform distribution (Min=0.95; Max=0.99) |  |
| Annual exposure frequency (n)    | 183 number of times                  |  |

3. Dose–response model with Monte Carlo simulation

*Staphylococcus aureus* bioaerosol

Dose–infection model: Exponential dose–response model  
Prevalence (P_{ill/inf})

*Escherichia coli* bioaerosol

Dose–infection model: Beta–Poisson dose–response model  
Dose–illness model: Exponential dose–response model

4. Risk characterization

Annual probability of infection (per person per year)  
U.S. EPA benchmark=10^{-4} pppy

Disease burden (DALYs per person per year)  
WHO benchmark=10^{-6} DALYs pppy

Figure 1
Figure 4