

Composition, dispersion, and health risks of bioaerosols in wastewater treatment plants: A review

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HIGHLIGHTS

- Bioaerosols are produced in the process of wastewater biological treatment.
- The concentration of bioaerosol indoor is higher than outdoor.
- Bioaerosols contain large amounts of potentially pathogenic biomass and chemicals.
- Inhalation is the main route of exposure of bioaerosol.
- Both the workers and the surrounding residents will be affected by the bioaerosol.

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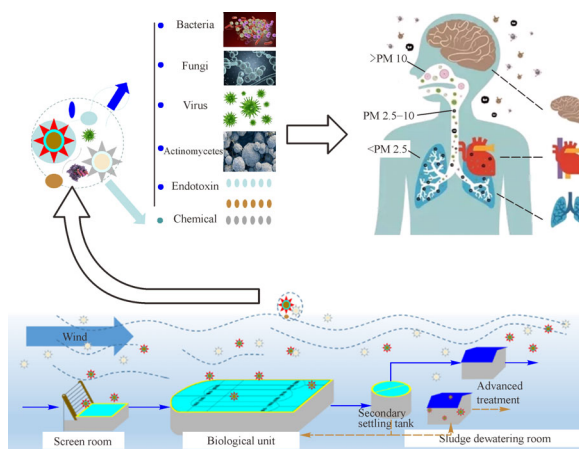
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GRAPHIC ABSTRACT



ABSTRACT

Bioaerosols are defined as airborne particles (0.05–100 μm in size) of biological origin. They are considered potentially harmful to human health as they can contain pathogens such as bacteria, fungi, and viruses. This review summarizes the most recent research on the health risks of bioaerosols emitted from wastewater treatment plants (WWTPs) in order to improve the control of such bioaerosols. The concentration and size distribution of WWTP bioaerosols; their major emission sources, composition, and health risks; and considerations for future research are discussed. The major themes and findings in the literature are as follows: the major emission sources of WWTP bioaerosols include screen rooms, sludge-dewatering rooms, and aeration tanks; the bioaerosol concentrations in screen and sludge-dewatering rooms are higher than those outdoors. WWTP bioaerosols contain a variety of potentially pathogenic bacteria, fungi, antibiotic resistance genes, viruses, endotoxins, and toxic metal(loid)s. These potentially pathogenic substances spread with the bioaerosols, thereby posing health risks to workers and residents in and around the WWTP. Inhalation has been identified as the main exposure route, and children are at a higher risk of this than adults. Future studies should identify emerging contaminants, establish health risk assessments, and develop prevention and control systems.

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1 Introduction

In recent years, wastewater production has increased with an increase in the population and continuous urbanization (Qu et al., 2019). More than 300 km³ of wastewater is produced globally each year (Wu et al., 2019). Out of this,

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approximately 60% is treated before release. Therefore, the number of wastewater treatment plants (WWTPs) also shows an increasing trend each year. By the end of 2018, there were approximately 4436 municipal WWTPs in operation in China alone (Yang et al., 2019d). Owing to their stable operation, low cost, and high treatment efficiency, more than 95% of these WWTPs adopt biological treatment processes such as anaerobic/anoxic/aerobic (A²O), oxidation ditch (OD), and sequencing batch reactor (SBR) processes. Approximately one billion species of active microorganisms exist in wastewater biological treatment systems, and the total number of cells in activated sludge is approximately $2.3 \pm 0.4 \times 10^9 \text{ mL}^{-1}$. These microorganisms play a crucial role in current wastewater treatment processes. Simultaneously, to support the growth and reproduction of these microorganisms, aeration, stirring, compression, and other mechanical devices are widely used in the sewage treatment process. The disturbance action of these mechanical devices promotes the particles in the water tank to break through the water–air interface and enter the atmosphere, thereby forming bioaerosols (Yang et al., 2020).

The term bioaerosols is a short form of biological aerosols. Bioaerosols are defined as airborne particles of biological origin (size range: 0.05–100 μm). They consist of bacteria, fungi, viruses, fungi, endotoxins, and toxic substances, among others (Schlosser, 2019). Many studies have summarized the properties and health risks of bioaerosols from various sources, including dairy processing facilities, agricultural facilities, farms, hospitals, compost, and landfills (Walser et al., 2014; Heldal et al., 2016; Straumfors et al., 2016; Stockwell et al., 2019; Madhwal et al., 2020). The effect of bioaerosols on human health mainly depends on their concentration, particle size distribution, composition, and dispersion processes (Mandal and Brandl, 2011). Owing to their microscale or nanoscale size, bioaerosols can easily be deposited throughout the human body (especially the respiratory system) and cause a range of diseases (Pastuszka et al., 2000; Liu et al., 2013; Gao et al., 2015). Particles with an aerodynamic diameter $>5 \mu\text{m}$ easily adhere to the upper respiratory tract and can cause rhinitis. Particles with an aerodynamic diameter $<5 \mu\text{m}$ can easily reach the alveoli. The finer particles, such as those at the nanoscale, are particularly toxic to cells (Nel, 2005; Nel et al., 2006). Thus, it is essential to understand the characteristics and health risks associated with bioaerosols.

A large number of microbes in wastewater treatment processes are potentially pathogenic and could harm human health (Wu et al., 2019). Therefore, bioaerosols from WWTPs also contain a variety of potentially pathogenic active substances (Han et al., 2018) and may also contain chemical components and polluted water (Han et al., 2018, 2019). During bioaerosol transmission, chemical components and moisture provide conditions for the long-term survival of potentially pathogenic

microorganisms in bioaerosols, thereby increasing the health risks that they pose. An illness among workers at WWTPs, termed “sewage worker’s syndrome,” is largely caused by the bioaerosol dispersal from WWTPs (Han et al., 2020). Given the significance of these health risks, we sought to summarize the existing research results on bioaerosols from WWTPs to advance the recognition, prevention, and control of such bioaerosols.

This article presents a short review of the current state of knowledge about the risks associated with WWTP bioaerosols. Further, the available literature on the concentrations, size distribution, and potential pathogenic components of WWTP bioaerosols are summarized, and relevant microbial genera, such as bacteria and fungi genera, populations are discussed. Further, emission characteristics and exposure risks of WWTP bioaerosols are also reviewed. Finally, challenges associated with WWTP bioaerosol prevention and control strategies are discussed, along with future research directions.

2 Wastewater treatment plant bioaerosol emissions

Concentration and size are among the most influential characteristics for determining the health risks of WWTP bioaerosols.

It is widely recognized that bioaerosols are produced in every stage of wastewater treatment. However, their emission levels can vary widely (Fig. 1). Bacterial aerosol concentrations of $0.00\text{--}5.16 \times 10^4 \text{ CFU/m}^3$ and fungal aerosol concentrations of $0.00\text{--}1.19 \times 10^4 \text{ CFU/m}^3$ were observed in WWTPs located in areas including Poland, the Middle East, and the People’s Republic of China (PR China) (Niazi et al., 2015; Silini et al., 2016; Szyłak-Szydłowski et al., 2016; Han et al., 2018; Han et al., 2019). The differences in bioaerosol concentrations found in different studies are closely related to the treatment scale, adopted processes, equipment selection, sampling time, and other factors including environmental conditions and WWTP location.

Overall, the pretreatment sections (coarse screen, fine screen, settling tanks, etc.), biological treatment sections (A²O, OD, and SBR), and sludge treatment sections (sludge thickening, sludge dehydration, etc.) of wastewater treatment are regarded as the main sources of bioaerosols (Fernando and Fedorak, 2005; Sánchez-Monedero et al., 2008; Han et al., 2018). The results in south-eastern Spain showed that the main detection zones of bioaerosols from WWTPs were the screen room (1787 CFU/m^3), biological treatment zones (4580 CFU/m^3), and sludge-thickening room (1050 CFU/m^3) (Sánchez-Monedero et al., 2008). The results of another study in Beijing, Hefei, Yixing, and Guangzhou in PR China found that the zones with the highest bioaerosol concentrations also comprised the screen room ($51\,590 \text{ CFU/m}^3$), biological treatment

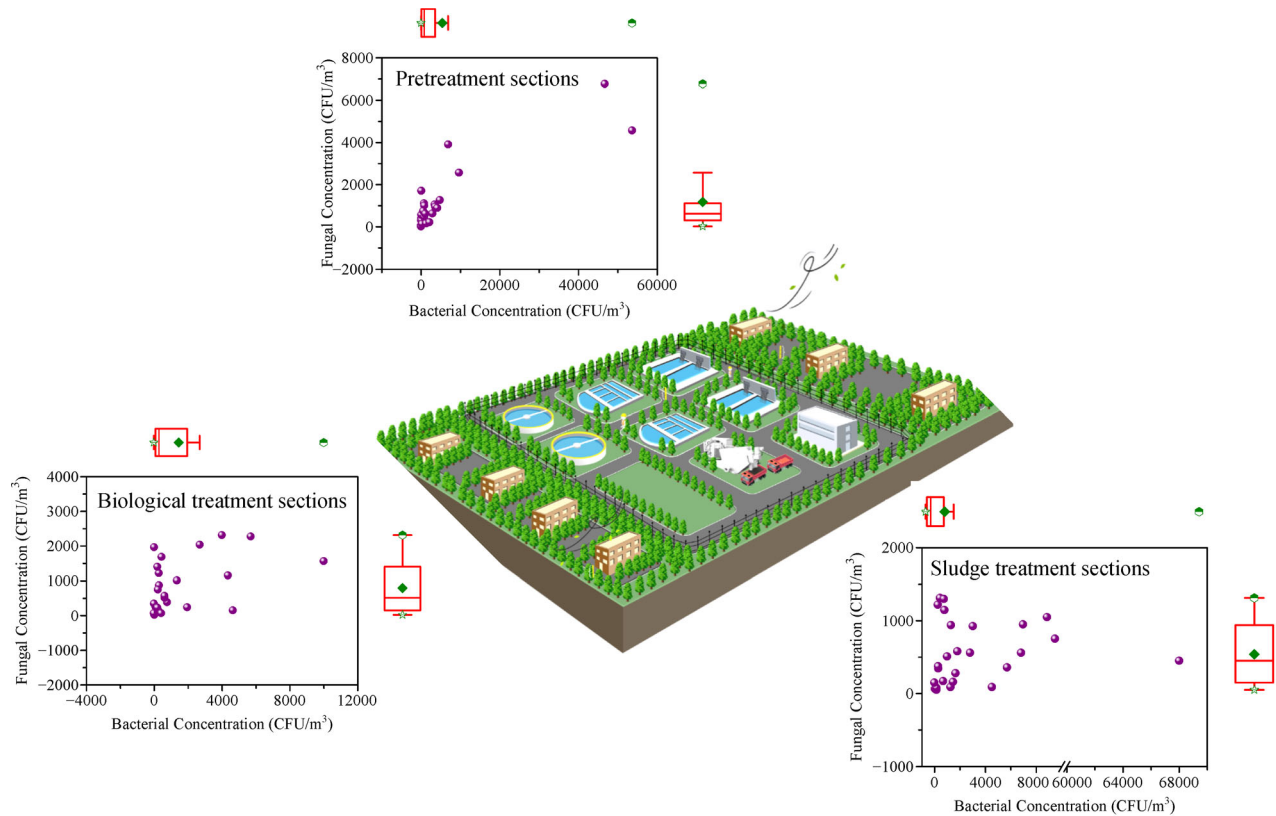


Fig. 1 Bioaerosol concentration at different operation sections of wastewater treatment plants (Data Sources: Fernando and Fedorak, 2005; Sánchez-Monedero et al., 2008; Niazi et al., 2015; Silini et al., 2016; Szyłak-szydłowski et al., 2016; Xu et al., 2018; Han et al., 2019; Wang et al., 2019a).

zones (554 CFU/m^3), and sludge-dewatering room (1525 CFU/m^3) (Han et al., 2018; Xu et al., 2018; Wang et al., 2019a).

In addition, some studies found that the bioaerosols from WWTPs are mainly small particles. For example, bacterial (52%), fungal (62%), and actinomycetes (65%) bioaerosols from the OD process were within the respirable size range (less than $3.3 \mu\text{m}$) (Li et al., 2013a). Other studies also found that more than half of the airborne bacteria from A^2O and airborne fungi from anoxic/aerobic treatment processes are attached to fine particles (smaller than $2.1 \mu\text{m}$ and $2.1\text{--}3.3 \mu\text{m}$, respectively) (Maharia and Srivastava, 2015; Wang et al., 2019a). Ding et al. (2016) further investigated an indoor WWTP and found that airborne bacteria and fungi were primarily distributed below $3.3 \mu\text{m}$ under more environmentally friendly operating conditions. Some studies have found that the size of bioaerosol particles in some WWTPs is large; however, in general, such particles are mainly distributed below $4.7 \mu\text{m}$ (Li et al., 2016; Kowalski et al., 2017; Wang et al., 2018a). The difference in bioaerosol size is mainly caused by the different properties of activated sludge, parameters of mechanical facilities, and environmental conditions in WWTPs.

The above research results were mainly obtained by collecting samples and then storing and transporting them back to the laboratory for testing and analysis. Ghosh et al. (2015) summarized in detail the sampling and analysis of bioaerosol samples based on both culture and culture-independent methods. The results showed that different research methods should be adopted for different research objectives. In recent years, the development of real-time detection has provided conditions for long-term continuous monitoring of the characteristics of bioaerosols in WWTPs and has revealed the spatial and temporal variation in bioaerosols. Nasir et al. (2019) compared the total mean concentration and fluorescence particle peak of bioaerosols in five different outdoor environments using a spectral intensity bioaerosol sensor. The results showed that the fluorescence peaks of bioaerosols in WWTPs are mainly $0.25 \mu\text{m}$ and $1.03 \mu\text{m}$. Another study confirmed that the fluorescence peaks of bioaerosols in WWTPs are distinct from those in the background and that the temporal variation in the average concentration of total particles and fluorescence particles is high (Tian et al., 2020). Thus, this information also revealed that the combination of multiple methods can provide a more comprehensive and profound understanding of the concentration, particle size distribu-

tion, composition, and temporal and spatial variation characteristics of bioaerosols in WWTPs and provide basic data for the understanding of their risks.

3 Bioaerosol composition characteristics

WWTP bioaerosols contain a large amount of biomass and chemicals (Fig. 2). Source analyses have indicated that most of the components of bioaerosols originate from wastewater/sludge (Han et al., 2019; Wang et al., 2019a). Therefore, it is necessary to identify and examine the characteristics of different WWTP bioaerosol components with a particular focus on identifying the main disease-causing components.

3.1 Bacteria

Bacteria are a type of prokaryotic microorganism abundant in all environments on the Earth, including extreme environments (van den Burg, 2003). Air has been widely proven to be an important carrier for many pathogenic bacteria (GBD Mortality and Causes of Death Collaborators, 2014). Most bacteria in WWTP bioaerosols are affiliated with the phyla Proteobacteria, Bacteroidetes, and Firmicutes (Table 1). These bacterial species are closely associated with different regions, seasons, technologies, and WWTP processes (Liu et al., 2013; Kowalski et al., 2017; Han et al., 2018; Yang et al., 2019a, 2019b).

Among these bacteria, many pathogenic or opportunistic-pathogenic bacteria such as *Staphylococcus aureus*,

Aeromonas hydrophila, *Comamonas testosteroni*, *Moraxella osloensis*, *Pseudomonas stutzeri*, and *Pseudomonas aeruginosa* have been detected in WWTP bioaerosols (Pascual et al., 2003; Fernando and Fedorak, 2005; Fracchia et al., 2006; Gangamma et al., 2011; Li et al., 2016; Wang et al., 2018b; Zhang et al., 2018; Yang et al., 2019b). These bacteria can affect humans via bioaerosol transmission and can cause respiratory infections, pyogenic infections, and septicemia in severe cases (Yang and Wang, 2006; Jang et al., 2009; Wang et al., 2011; Kim et al., 2018). In addition, many Enterobacteriaceae bacteria were found in WWTP bioaerosols, especially in screen rooms (Gotkowska-Płachta et al., 2013; Xu et al., 2018). These intestinal bacteria have been found to be closely related to certain respiratory diseases (such as toxic pneumonia, chronic bronchitis, and asthma) (Espigares et al., 2006).

3.2 Fungi

Fungal spores can also survive in a variety of environments on the Earth. Previous studies detected several fungi in WWTP bioaerosols (Table 2), including *Geotrichum candidum*, *Cladosporium lignicola*, and *Alternaria alternate* (Prazmo et al., 2003; Cyprowski et al., 2008; Park et al., 2011). In addition, these dominant fungal species present close regional and seasonal correlations (Han et al., 2019). Among the detected airborne fungi, *Penicillium*, *Aspergillus*, and *Cladosporium* are associated with respiratory infections and allergic reactions (Vujanovic et al., 2001; Kanaani et al., 2008). *G. candidum* is currently

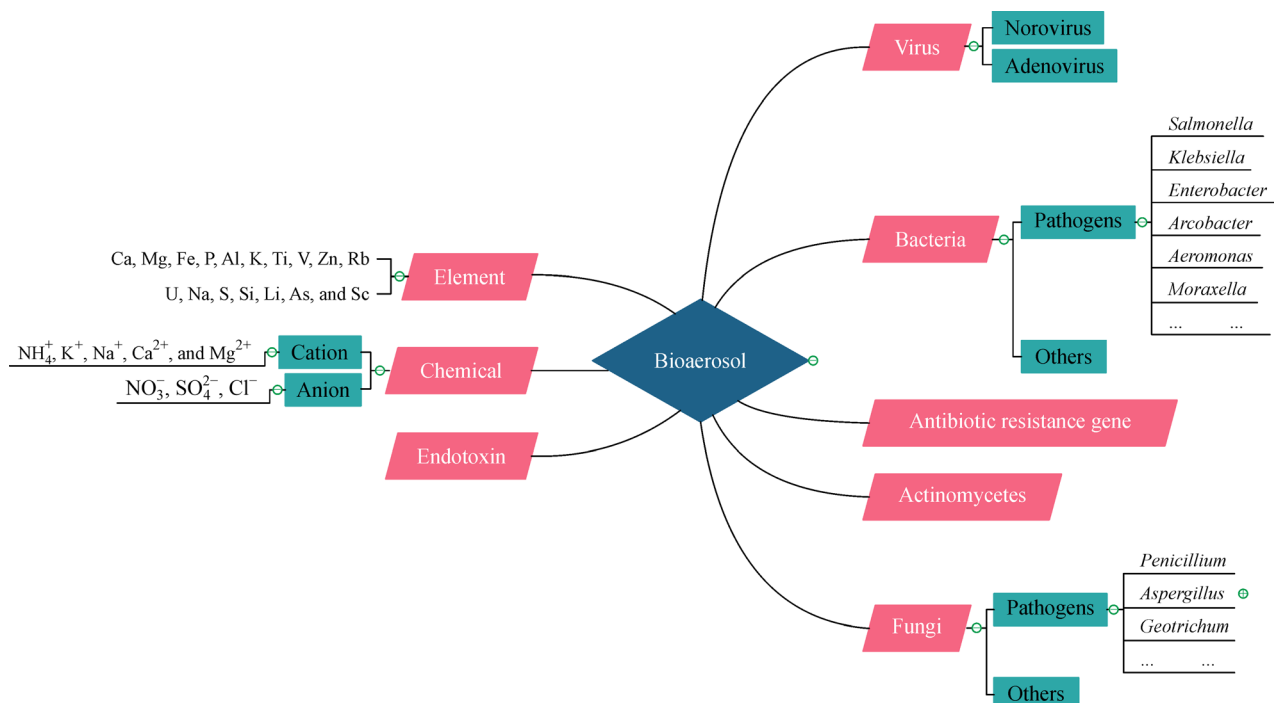


Fig. 2 Schematic diagram of main components of bioaerosol in wastewater treatment plant.

Table 1 Predominant bacteria in bioaerosols from WWTPs

Number	Phylum	Genus	Species	Sampling sites	Wastewater property	Treatment technology	Country or region	References
1	<i>Firmicute</i>	<i>Bacillus</i>	/	Grille	Municipal wastewater	A/O	Beijing, China	Liu et al. (2013)
	<i>Proteobacteria</i>	<i>Escherichia</i>	<i>Escherichia coli</i>					
		<i>Pseudomonas</i>	/					
	<i>Firmicute</i>	<i>Staphylococcus</i>	/	Aeration tank				
		<i>Brevibacterium</i>	/					
	<i>Firmicute</i>	<i>Bacillus</i>	/					
		<i>Staphylococcus</i>	/	Sludge treatment				
	<i>Actinobacteria</i>	<i>Corynebacterium</i>	/					
	<i>Proteobacteria</i>	<i>Pseudomonas</i>	/					
	<i>Escherichia</i>	<i>Escherichia coli</i>						
<i>Firmicute</i>	<i>Bacillus</i>	/						
2	<i>Firmicute</i>	<i>Bacillus</i>	/	Oxidation ditch	Municipal wastewater	Oxidation ditch	Beijing, China	Yang et al. (2019b)
		<i>Lysinibacillus</i>	/					
	<i>Proteobacteria</i>	<i>Sphingomonas</i>	/					
3	<i>Firmicute</i>	<i>Bicillus</i>	/	Upwind direction, Mechanical treatment, Biological treatment, Downwind direction	Municipal wastewater	A ² O	Beijing, China	Han et al. (2019)
	<i>Actinobacteria</i>	<i>Arthrobacter</i>	/					
	<i>Firmicute</i>	<i>Micrococcus</i>	/					
4	<i>Acidobacteria</i>	<i>Geothrix</i>	/	Upwind direction	Municipal wastewater	Oxidation ditch	Hefei, China	Yang et al. (2019d)
	<i>Actinobacteria</i>	<i>Microthrix</i>	/					
	<i>Cyanobacteria</i>	<i>Cyanobacteria</i>	/	Aeration unit				
	<i>Bacteroidetes</i>	<i>Saprospiraceae</i>	/	Sludge dewatering room				
5	<i>Proteobacteria</i>	<i>Mitochondria</i>	/	Sludge dewatering room	Municipal wastewater	Activated sludge	Guangzhou, China	Han et al. (2018)
	<i>Firmicute</i>	<i>Peptostreptococaceae</i>	/					
	<i>Proteobacteria</i>	<i>Sphingomonas</i>	/					
6	<i>Firmicute</i>	<i>Staphylococcus</i>	/	Mechanical treatment, Aeration tank, Clarifier, Sludge treatment	Municipal wastewater, wastewater, Food processing wastewater,	Activated sludge	Poland	Kowalski et al. (2017)
		<i>Bacillus</i>	/					
7	<i>Actinobacteria</i>	<i>Corynebacterium</i>	/	Mechanical treatment, Biological treatment, Sludge treatment	Municipal wastewater	Activated sludge	Eastern Poland	Prazmo et al. (2003)
	<i>Actinobacteria</i>	<i>Arthrobacter</i>	/					
	<i>Firmicute</i>	<i>Brevibacterium</i>	/					
		<i>Staphylococcus</i>	/					
		<i>Micrococcus</i>	/					
	<i>Bacillus</i>	/						
8	<i>Firmicute</i>	<i>Bacillus</i>	/	Mechanical treatment, Biological treatment, office	Municipal wastewater	/	India	Gangamma et al. (2011)

Table 2 Predominant fungi in bioaerosols in WWTPs

Number	Phylum	Genus	Species	Sampling sites	Wastewater property	Treatment technology	Country or region	references
1	<i>Zygomycotina</i>	<i>Mucor</i>	/	Grille	Municipal wastewater	A/O	Beijing, China	Liu et al. (2013)
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					
	<i>Zygomycota</i>	<i>Rhizopus</i>	/	Aeration tank				
	<i>Ascomycota</i>	<i>Aspergillus</i>	/					
	<i>Deuteromycotina</i>	<i>Paecilomyces</i>	/	Sludge treatment				
	<i>Zygomycotina</i>	<i>Mucor</i>	/					
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					
	<i>Zygomycota</i>	<i>Rhizopus</i>	/					
	<i>Zygomycotina</i>	<i>Mucor</i>	/					
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					
2	<i>Deuteromycotina</i>	<i>Geotrichum</i>	<i>Geotrichum candidum</i>	Mechanical treatment, Biological treatment, Sludge treatment	Municipal wastewater	Activated sludge	Eastern Poland	Prazmo et al. (2003)
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					
	<i>Deuteromycotina</i>	<i>Cladosporium</i>	<i>Cladosporium lignicola</i>					
	<i>Ascomycota</i>	<i>Alternaria</i>	<i>Alternaria alternate</i>					
3	<i>Ascomycota</i>	<i>Aspergillus</i>	/	Mechanical treatment, Biological treatment, Sludge treatment	Municipal wastewater	/	Central Poland	Cyprowski et al. (2008)
	<i>Zygomycotina</i>	<i>Mucor</i>	/					
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					
	<i>Ascomycota</i>	<i>Alternaria</i>	/					
4	<i>Deuteromycotina</i>	<i>Cladosporium</i>	/	Aeration tank	Municipal wastewater, Feces-urine wastewater	/	Korea	Park et al. (2011)
	<i>Ascomycota</i>	<i>Alternaria</i>	/					
	<i>Ascomycotina</i>	<i>Penicillium</i>	/					

recognized as the most common fungal symbiont and pathogen in humans, and is also associated with infectious diseases under certain conditions (Sun and Lu, 2018). Moreover, some fungi are cytotoxic in specific environments, and the mycotoxins produced by them are toxic to humans.

3.3 Actinomycetes

Actinomycetes have also been widely detected in WWTP bioaerosols (Korzeniewska et al., 2009). The most prevalent actinomycetes genera are *Thermoactinomyces* and mesophilic actinomycetes. The dominant *Thermoactinomyces* species are *Thermoactinomyces thalophilus* (47.2%), *Thermoactinomyces vulgaris* (22.2%), *Saccharopolyspora rectivirgula* (13.9%), and *Thermomonospora fusca* (13.9%) (Prazmo et al., 2003). *Streptomyces* spp. and *Nocardia* spp. are the dominant mesophilic actinomycetes species (Kowalski et al., 2017). Although actinomycetes concentrations are relatively low in aerosols, the potential health risks caused by actinomycetes cannot be ignored. Studies have found that some species of *Thermoactinomyces* can be pathogenic, such as *T. vulgaris*, which can cause hypersensitivity pneumonia (Liu et al., 2012).

Exposure to other pathogenic actinomycete particles can lead to jaw tumors, lung infections, and even death (Lacey and Crook, 1988).

3.4 Viruses

Airborne viruses are also responsible for work-related symptoms in WWTP employees (Divizia et al., 2008). Studies have detected norovirus (NoV; 84%) and human adenovirus (AdV; 2.4%) in WWTP bioaerosols (Masclaux et al., 2014). NoV and AdV cause the two most common human infections transmitted by the fecal-oral route. NoV is an important pathogen that causes global outbreaks of non-bacterial acute gastroenteritis, and it is also a significant cause of food-borne infections. Approximately 90% of non-bacterial diarrhea is caused by this virus (Glass et al., 2009). AdV is a DNA virus that usually causes mild infections of the respiratory tract, gastrointestinal tract, or conjunctiva (Shimizu et al., 2007).

3.5 Antibiotic resistance genes

WWTPs are one of the main sources and sinks of antibiotic resistance genes (ARGs) (Rizzo et al., 2013; Gaviria-

Figuerola et al., 2019; Yang et al., 2019c). A few studies have shown that ARGs have been detected in WWTP bioaerosols. For example, researchers identified the *Sul2* and *intI1* genes in the screen rooms and biological treatment units of WWTPs, respectively (Li et al., 2016). These ARGs are thought to be closely related to human health risks; the *Sul2* gene is related to community-acquired respiratory tract pneumonia (Enne et al., 2002).

3.6 Endotoxins

In addition to microorganisms, microbial products such as endotoxins are also widely found in bioaerosols. According to the Dutch Expert Committee, exposure to endotoxins in bioaerosols in occupational environments should remain below 50 EU/m³. When concentrations of endotoxins are higher than 50 EU/m³, a significant dose-responsive relationship between endotoxin concentrations and lower respiratory tract infections, skin inflammation, influenza, and other symptoms has been observed. The concentration of endotoxins in WWTP bioaerosols has been found to exceed the relevant standards intended to protect occupational health (Helldal et al., 2017). In addition, the endotoxin concentrations in bioaerosols in indoor treatment units have been found to be significantly higher than those detected in outdoor treatment units (Uhrbrand et al., 2011).

3.7 Chemicals

Chemicals in bioaerosols can provide suitable microenvironments for the survival and growth of airborne microorganisms (Han et al., 2012). The main chemicals in WWTP bioaerosols include Total Carbon, Total Organic Carbon (TOC), water-soluble ions, and major elements. The proportion of TOC in WWTP bioaerosols is higher than that in the air, which may be related to the presence of organic or living matter (Yang et al., 2019b). Water-soluble ions in bioaerosols include anions (including NO₃⁻, SO₄²⁻, and Cl⁻) and cations (including NH₄⁺, K⁺, Na⁺, Ca²⁺, and Mg²⁺). Na⁺ has been found to be the dominant cation, followed by Ca²⁺. SO₄²⁻ was determined to be the dominant anion, followed by NO₃⁻ and Cl⁻ (Han et al., 2018). The major elements in bioaerosols vary according to different functional areas in WWTPs. Ca, Mg, Fe, P, Al, K, Ti, V, Zn, Rb, and U are the main elements of bioaerosols in the aeration unit, whereas Na, S, Si, Li, and Sc are the most important elements in bioaerosols in the sludge-dewatering room (Yang et al., 2019b).

4 Formation

The agitation of open water, such as in WWTP aeration tanks, is regarded as a primary source of bioaerosols released in outdoor areas (Fannin et al., 1985; Sánchez-

Monedero et al., 2008). Wastewater biological treatment technology degrades pollutants through the metabolism of microorganisms to achieve wastewater purification. Oxygen dissolved in water is supplied by diffusers installed at the bottom of tanks (submerged aeration) or by rotor aeration systems (surface aeration) facilitated on the surface of the water to accelerate substrate metabolism. Aeration is a mass transfer process in which oxygen molecules are exchanged between water and the air via a gas/liquid interface, which plays an important role in the purification of wastewater. Bioaerosols formed by submerged aeration are different to those produced by surface aeration because of the differences in the mechanism and parameters during the generation processes.

4.1 Surface aeration

Paddle wheel aerators are typical devices for surface aeration that are widely adopted in WWTPs. At present, there are few reports on the processes and mechanism of bioaerosol formation generated by surface aeration. Existing research has mainly focused on paddle wheel aeration.

In a study conducted by Han et al. (2020) blades installed in ODs were rotated at a certain speed during aeration. They disturbed the water body and sprayed water into the atmosphere. A negative pressure zone was generated at the rear of the tank, which absorbed some of the air and caused the water to jump at the surface (Han et al., 2020). In front of the aerator, the lifted tiny droplets traveled in the air as para-curves. Atomization zones were generated when the droplets mixed with the air and lost moisture. Three stages are involved in the procedure of aerosolization in surface aeration, that is, (1) tiny droplets with microbes are thrown into the surrounding air, (2) tiny droplets entering the atmosphere produce bioaerosols after losing water, and (3) air is drawn into the water to form bioaerosols owing to the disturbance of the water surface caused by the liquid falling back (Wang et al., 2019b). The quantity and grain size depend on the amount of atomization, which can be calculated by determining the parameters such as droplet ejection speed, aerated water thickness, starting position of atomization, and wind speed (Liu et al., 2006; Sheng et al., 2006). Bioaerosol formation mechanisms were explored via a simulation experiment using laboratory-scale rotating blades as a surface aerator (Wang et al., 2019b). Droplet sedimentation was investigated in a fixed space, and the atomization quantity was calculated. When the rotation speed was 50 r/min, droplets settled in the fixed area reached 16 850 within 1 min, and over 30% of the droplets were greater than 2 mm. The atomization quantity in the experiment reached 6.3×10^{-5} m³/s.

Water aeration, mixing, and flow propulsion by aerators drive the transfer of microorganisms from the liquid into the surrounding air. These processes were explored in

another experiment conducted using brushes rotating at speeds of 40–60 r/min as a surface aerator (Li et al., 2011). The brushes stirred the water body and created a thin water curtain, which then burst into many tiny droplets. *Zoogloea* and activated sludge flocs, namely the surface-active granules, were dispersed in the air with the tiny droplets. It was found that microorganisms, mycelium, and sludge particles gathered in the resultant droplets, and the outer layer was covered with a water film (Han et al., 2012). Although this process enables mass transfer of atmospheric oxygen into the water droplets, the fine droplets containing *Zoogloea* act as a medium during bioaerosol formation and are transmitted to the atmosphere.

4.2 Submerged aeration

Submerged aeration, e.g., microporous aeration, forces air into the water body to provide oxygen and causes turbulence owing to the thorough mixing of the liquids (Frank et al., 2009). Bubble aeration, which is typically applied in WWTPs with A²O and SBR treatment processes, provides oxygen via air bubbles released from air diffusers arranged at the bottom of the tanks. The bubbles form at the tiny outlets of the diffusers and float upward to the water surface. They eventually burst and project a large number of droplets into the air at the water surface. Microorganisms present in water are released into the surrounding air with the droplets, thereby leading to bioaerosol emissions.

Bubbles can break into a single bubble or a group of bubbles with diverse sizes. In an experiment considering single-bubble bursting, the film droplets were colored and observed in a fixed area. When 10 bubbles burst, 1290 film droplets were observed to have formed. More film droplets were generated as the number of broken bubbles increased (Wang et al., 2019b). Over 85% of the particles were less than 2.0 mm in diameter. The amount of film droplets produced was found to be correlated with the size of the ruptured bubbles. Bubbles with 1.7 mm diameters produced 10–20 film droplets, which ranged from < 2 μm to over 30 μm in diameter. Half of the droplets were < 10 μm (Blanchard and Syzdek, 1982). Using holographic imaging techniques, Afeti and Resch (1990) demonstrated that the amount and particle size of the formed film droplets increase with an increase in the bubble size.

Bacteria can be enriched in droplets formed from

bubbles that burst at the surface of bacterial suspensions (Baylor et al., 1977; Hejkal et al., 1980). Enrichment factors (the ratio of the concentration of bacteria in a droplet to that in the bulk suspension) vary from 1 to more than 1000. To investigate the water-to-air transmission of pollutants via aerosols from submerged aeration in wastewater treatment processes, research was conducted to determine the generation of aerosols from bursting air bubbles with sizes of millimeters to centimeters. The results showed that the number and size of bioaerosols are mainly influenced by the diameter of bubbles. Larger bubbles resulted in the formation of thicker film droplets and the release of larger bioaerosols. The short-lived bubbles break down, thereby resulting in a thick rupture film and a small number of aerosols (Ke et al., 2017). The mass of released aerosols also increased with the increase in the bubble size, which means that compared with tiny bubbles, large bubbles contain more chemicals during water-to-gas transportation. Aerosol emissions from bursting bubbles are associated with the size, speed, and position of the film droplets, which are strongly affected by the water properties e.g., chemical oxygen demand, sludge concentration, and flocs size. The correlation between the particle mass and the chemicals in the water follows a linear function (Russell and Singh, 2006). A similar result was obtained for the chemicals and microbes in bioaerosols from six WWTPs (Wang et al., 2018b).

5 Dispersion

Microorganisms and chemicals initially existing in the water are transferred to the air through atomization droplets, thereby forming bioaerosols that are suspended in the atmosphere. The kinetic energy provided by air motion acts on bioaerosols and allows them to travel. The energy obtained by bioaerosols is considerable, which leads to their long-distance transmission in the air. Bioaerosol transportation can be defined in terms of time and distance, and the specific parameters are summarized in Table 3 (Pepper and Dowd, 2009). WWTP bioaerosols typically travel via sub-microscale and microscale transport.

5.1 Spatial variations

The concentration, particle size distribution, and microbial and chemical compositions of bioaerosols vary with the

Table 3 Transport of bioaerosols

Transport scale	Time	Distance	Occasions
Submicroscale	< 10 min	< 100 m	Common type within buildings or other confined spaces
Microscale	10 min–1 h	100 m to 1 km	Common type for bioaerosols transportation
Mesoscale	A few days	Up to 100 km	Seldom occur in WWTPs
Macroscale	Longer than a few days	>100km	No report related to WWTPs

distance from the bioaerosol source owing to dispersion. Table 4 shows the variations in the bioaerosol concentrations along a horizontal distance from their sources. Bioaerosol observations from municipal WWTPs showed that bacterial aerosols clearly decreased when the distance from the sources increased (Li et al., 2013a). Ding et al. (2016) conducted a bioaerosol emission survey at a wastewater treatment station, in which the rotary brush installed in the OD was identified as the source of bacteria and fungi in the air. There were 4155 CFU/m³ of airborne bacteria and 883 CFU/m³ of airborne fungi detected at a site 1.5 m from the source. When the distance to the source increased to 6.5 m, the concentration of bacterial aerosols decreased by 68% and that of fungal aerosols decreased by 77%. Moreover, Yang et al. (2019a) found that when the distance from the OD increased from 0.5 m to 25.0 m, 55.0 m, and 210.0 m, the levels of airborne bacteria in the air decreased gradually from 4536 CFU/m³ to 2042 CFU/m³,

1475 CFU/m³, and 1057 CFU/m³, respectively. In another study, over 34.6% of airborne bacteria were found in particle sizes below 1.1 μm at an area located 25 m downwind of the rotating brushes, whereas only 2.0% of airborne bacteria were observed in particles with the same size near the rotating brushes in OD. Large particles tended to fall down rather quickly (Li et al., 2011). Air samples were gathered at different distances from bioaerosol-generating sources in a WWTP. The microbial structure and chemicals of the bioaerosols demonstrated site-specific variations. Elements and water-soluble ions in the bioaerosols exhibited reduced concentrations at 20 m and 40 m downwind of the sources. Some species, such as Proteobacteria and Bacteroidetes, were not found at distances far downwind from the bioaerosol source (Han et al., 2012). Similarities appeared in the release of aerosols containing bacteria and viruses from activated sludge WWTPs (Fannin et al., 1985).

Table 4 Variation of bioaerosols concentrations along horizontal distance from the sources

Distance (m)	Airborne bacteria (CFU/m ³)	Airborne fungi (CFU/m ³)	<i>E. coli</i> (PFU/m ³)	Fecal coliform (PFU/m ³)	Fecal streptococci (PFU/m ³)	<i>E. coli</i> phage (PFU/m ³)	Rotavirus (viruses/m ³ h)	Norovirus (viruses/m ³ h)	Norovirus (GC/m ³)
0	2358 ^a	–	–	–	–	–	–	–	–
20	972 ^a	–	–	–	–	–	–	–	–
40	669 ^a	–	–	–	–	–	–	–	–
Upwind	824 ^b	–	–	–	–	–	–	–	–
0.5	4536 ^b	–	–	–	–	–	–	–	–
25	2042 ^b	–	–	–	–	–	–	–	–
55	1475 ^b	–	–	–	–	–	–	–	–
210	1057 ^b	–	–	–	–	–	–	–	–
1.5	4155 ^c	883 ^c	–	–	–	–	–	–	–
6.5	1313 ^c	203 ^c	–	–	–	–	–	–	–
Upwind	–	–	0.22 ^d	0.01 ^d	0.04 ^d	120 ^d	–	–	–
<150	–	–	6.81 ^d	1.67 ^d	0.29 ^d	730 ^d	–	–	–
150–250	–	–	0.86 ^d	0.18 ^d	0.15 ^d	490 ^d	–	–	–
>250	–	–	0.4 ^d	0.29 ^d	0.48 ^d	760 ^d	–	–	–
100	–	–	–	–	–	–	–	–	0.16 ^e
300	–	–	–	–	–	–	–	–	8.60E-03 ^e
500	–	–	–	–	–	–	–	–	2.10E-03 ^e
1000	–	–	–	–	–	–	–	–	2.90E-04 ^e
Upwind 250	106 ^f	67 ^f	–	–	–	–	–	–	–
100	166 ^f	322 ^f	–	–	–	–	–	–	–
250	198 ^f	394 ^f	–	–	–	–	–	–	–
500	181 ^f	189 ^f	–	–	–	–	–	–	–
0	–	–	–	–	–	–	27 ^g	3099 ^g	–
300	–	–	–	–	–	–	3.87E-06 ^g	1.75E-04 ^g	–
500	–	–	–	–	–	–	1.51E-06 ^g	1.74E-04 ^g	–
1000	–	–	–	–	–	–	4.56E-07 ^g	5.24E-05 ^g	–

Note: a: Li et al., 2013a; b: Yang et al., 2019b; c: Ding et al., 2016; d: Fannin et al., 1985; e: Courault et al., 2017; f: Fathi et al., 2017; g: Pasalari et al., 2019.

The concentrations of bioaerosols suspended at greater heights are demonstrated in Table 5. Vertical diffusion of bioaerosols was investigated at different heights above the water surface at ODS; 849 CFU/m³ of airborne bacteria was detected at the sampling point situated 0.1 m above water surface. When the height increased to 1.5 m and 3.0 m, the level of bacteria in the air decreased to 641 and 141 CFU/m³, respectively (Wang et al., 2018a). Similar results were found in a survey of other WWTPs, including plants using submerged aeration (Han et al., 2013; Yang et al., 2019a). Vertical sampling points were established at the biochemical reaction tanks 0.1 m, 1.5 m, and 3.0 m above the water surface to investigate the spatial variations in the concentrations of chemicals and microorganisms in the bioaerosols. The highest concentration of bioaerosols was observed at 0.1 m above the water surface. When the height increased to 3.0 m, the concentrations of bacteria and fungi decreased by 36.14% and 23.71%, respectively, while the concentrations of total suspended particles and total organics decreased by 38.74% and 10.72%, respectively (Wang et al., 2018b).

5.2 Dispersion factors

Bioaerosols are immediately influenced by the atmosphere when exposed to the environment. Wind speed affects the diffusion velocity and distance of bioaerosols. The emission and dispersal patterns of ARGs through bioaerosols released from a sewage treatment plant were simulated according to the parameters of discharge velocity and wind speed (Gaviria-Figueroa et al., 2019). Over 220 000 ARGs were scattered within a 10 000 m radius around the sewage plant when the wind speed was 1.39 m/s, while more than 100 000 and 200 000 ARGs were present in the air 120 000 m from the source at wind speeds of 2.78 m/s and 5.56 m/s, respectively. The results obtained from the report suggested that bioaerosols containing ARGs may be carried several kilometers from the sources in sewage plants under high wind speeds.

The action of gravity on aerosol particles is another factor that affects dispersion. Gravity acts on all particles heavier than air and pulls them down, which essentially provides spatial and temporal limits for the diffusion of airborne particles. Sedimentation occurs during dispersion, thereby reducing the levels of airborne bacteria and airborne fungi in the atmosphere (Ding et al., 2016). As gravity deposition applied to the particles is more

important than other forces, the diffusion of large aerosol particles, especially those with a particle size larger than 1 μm , is easily affected by gravity deposition (Pillai and Ricke, 2002). As a result, large bioaerosol particles settle faster than smaller particles during the diffusion process. Owing to their low weight and small size, small particles can be easily blown over long distances by the wind, including several kilometers downwind from their sources. Compared with large bioaerosol particles, those with a smaller size may have greater impacts on the air quality around the urban sewage plant and on the health of the residents nearby.

5.3 Bioaerosol survival

When suspended in the atmosphere, the viability of most microorganisms is limited because many environmental factors such as relative humidity, temperature, oxygen content, and ultraviolet (UV) radiation can affect their survival. However, some microorganisms have specific mechanisms that allow them to be resistant to adverse environmental conditions. The microorganism affiliated with Rhodocyclales found in the investigation by Han et al. (2012) has bacteria chlorophyll and carotenoids, which may exhibit photoautotrophic growth with molecular hydrogen. In addition, the acid proof characteristics of *Burkholderia* also ensure activity over long-distance transmission. Some viruses, spores, and spore-forming bacteria have been shown to have mesoscale or even macroscale transport capacity. It was found that the concentration of viruses in aerosols decreased by seven orders of magnitude when the horizontal distance from a WWTP increased from 0 m to 300 m. In the range of 100 m to 300 m, the concentration decreased by two orders of magnitude. However, there was no clear change in the virus concentration in the air when the distance increased to 1000 m (Courault et al., 2017). For the bioaerosols produced from wastewater, insoluble components can serve as media for microorganism adhesion, and chemicals might provide nutrients for the growth of cells in aerosols. Moreover, the layer of moisture surrounding the microorganisms may provide protection against UV damage. Previous studies indicated that even though transport through the environment can be detrimental to the viability of aerosolized microbial cells, cell growth can occur in airborne particles (Dimmick et al., 1979; Fannin et al., 1985).

Table 5 Variation of bioaerosol concentrations along vertical distance from the wastewater surface

Concentration(CFU/m ³)	Vertical distance from water surface (m)							References
	0	0.1	0.5	1.0	1.5	2.0	3.0	
Airborne bacteria	–	1588	–	–	730	–	138	Yang et al. (2019a)
	–	715	–	–	238	–	–	Wang et al. (2019a)
	1416	–	2358	–	–	646	–	Li et al. (2013a)
	1755	–	4536	1047	–	–	–	Yang et al. (2019b)

6 Risk assessment

6.1 Health effects

Human exposure to bioaerosols can cause infectious diseases, respiratory diseases, cancer, and other diseases. Infectious diseases are caused by viruses, bacteria, and other microorganisms, and their transmission channels include direct contact and vector transmission. Legionella can spread in the air owing to aerosolization processes, such as the aeration of wastewater in WWTPs (Douwes et al., 2003). Respiration is a major pathway by which bioaerosol-based pathogens reach the lungs in the human body. Inhalation of microorganisms related to bioaerosols can cause a variety of respiratory diseases. Carcinogenic viruses and biological agents are two typical factors that lead to cancer (Humbal et al., 2018; Kim et al., 2018). In addition to the aforementioned diseases, other adverse health effects such as communicable diseases have been reported in previous research (Charous et al., 1994). The transmission of pathogenic microbes via bioaerosol carriers is responsible for communicable diseases via direct contact or indirect contact (Baker and Gray, 2009).

6.2 Exposure and risk assessment

Through quantitative estimates of emission rates, airborne concentrations that represent exposure levels can be converted into doses using dosimetry models to perform quantitative microbial risk assessments (QMRAs) (Upper and Hirano, 1991). The following processes are considered to quantify the risk exposure to pathogenic bioaerosols (Van Leuken et al., 2016):

- (1) Emission rates of pathogenic bioaerosols. This is a function of pathogen availability and the aerosolization rate.
- (2) Meteorological parameters including turbulence, wind velocity, and direction and gravity settlement.
- (3) Inactivation, which is described as a temporal or meteorological function.
- (4) The amount of pathogenic bioaerosols inhaled, which is determined according to factors including the breathing rate, lung volume, and particle size.
- (5) The healthy response of subjects to the inhalation dose.

A QMRA was developed to evaluate the health risks posed by bioaerosols from recycled water sprinkling, greywater reuse, and other water-related sources (Hamilton et al., 2018). QMRA results are generally in view of one or more objective pathogenic agents. A model developed by the United States Environmental Protection Agency (USEPA, 1999) is frequently utilized to evaluate the health risks posed by exposure to WWTP bioaerosols. Two main exposure pathways are considered, namely inhalation and direct skin contact. The individual exposure dose for each

route can be expressed in daily dosage and can be calculated by a formula. The dosages of microbial aerosols received via inhalation (ADD_{inh}) and direct skin contact (ADD_{skin}) are associated with the concentration of bioaerosols at the exposure point, exposure frequency and time, average bodyweight, and inhalation rate or exposed skin surface. They can be assessed using the equations described by Li et al. (2013b). Comparing the daily dosage with the chronic exposure reference dose (RfD) can evaluate the hazard of non-carcinogenesis. The hazard quotient (HQ) indicates the danger of non-carcinogenic contaminants, which is the ratio of ADD to RfD. The hazard index, which is expressed as HI, represents the total hazards of various exposure routes for each contaminant. When the HQ (or HI) is less than 1, the risks are non-carcinogenic, which can be neglected, whereas when the HQ (or HI) is greater than 1, the risks are considered potentially carcinogenic and are a concern. The USEPA (2011) provides most of the parameters used for exposure risk assessment. With this model, a microbial risk assessment was conducted for bioaerosols generated by aerators installed in ODs for wastewater treatment. Inhalation was the main exposure route for pathogenic bioaerosols in the surrounding community. The exposure HQ associated with the inhalation route was over 10^5 times greater than that of the dermal contact route for both children and adults (Li et al., 2013b).

A QMRA including a simplified atmospheric dispersion model allowed the assessment of NoV infection risk. The probability of infection within 1 year for WWTP employees with the highest bioaerosol exposure levels was $>10^{-4}$ given strong wind speeds (≥ 3 m/s). This probability decreased by 3 log when the distance to the emission source was doubled (Courault et al., 2017). Numerous investigations have evaluated the risks of exposure to WWTP bioaerosols. Relevant findings demonstrated that the HQ and HI vary by location (Fig. 3). Among on-site workers, male employees typically had higher health risks due to exposure to pathogenic bioaerosols than female employees. In communities surrounding the sources, children were far more at risk than adults. Owing to the presence of pathogens in bioaerosols, the exposure risks must be considered, although in most studies, the HQ and HI associated with exposure to such bioaerosols were below 1. Both the pathogenic concentration and particle size are related to the exposure risk of bioaerosols. Coarse particles with aerodynamic diameters greater than $2.1 \mu\text{m}$ primarily settle in the extrathoracic area, and fine particles can reach the alveolar area of human lungs. Moreover, small particles can easily spread to regions hundreds of meters or even kilometers away by wind. Therefore, aerosols with small particle sizes play an important role in city air quality and the health of residents. In recent years, increasing attention has been paid to bioaerosol exposure, particularly with respect to the relationship between health risks and the deposition fraction of microbial aerosols from

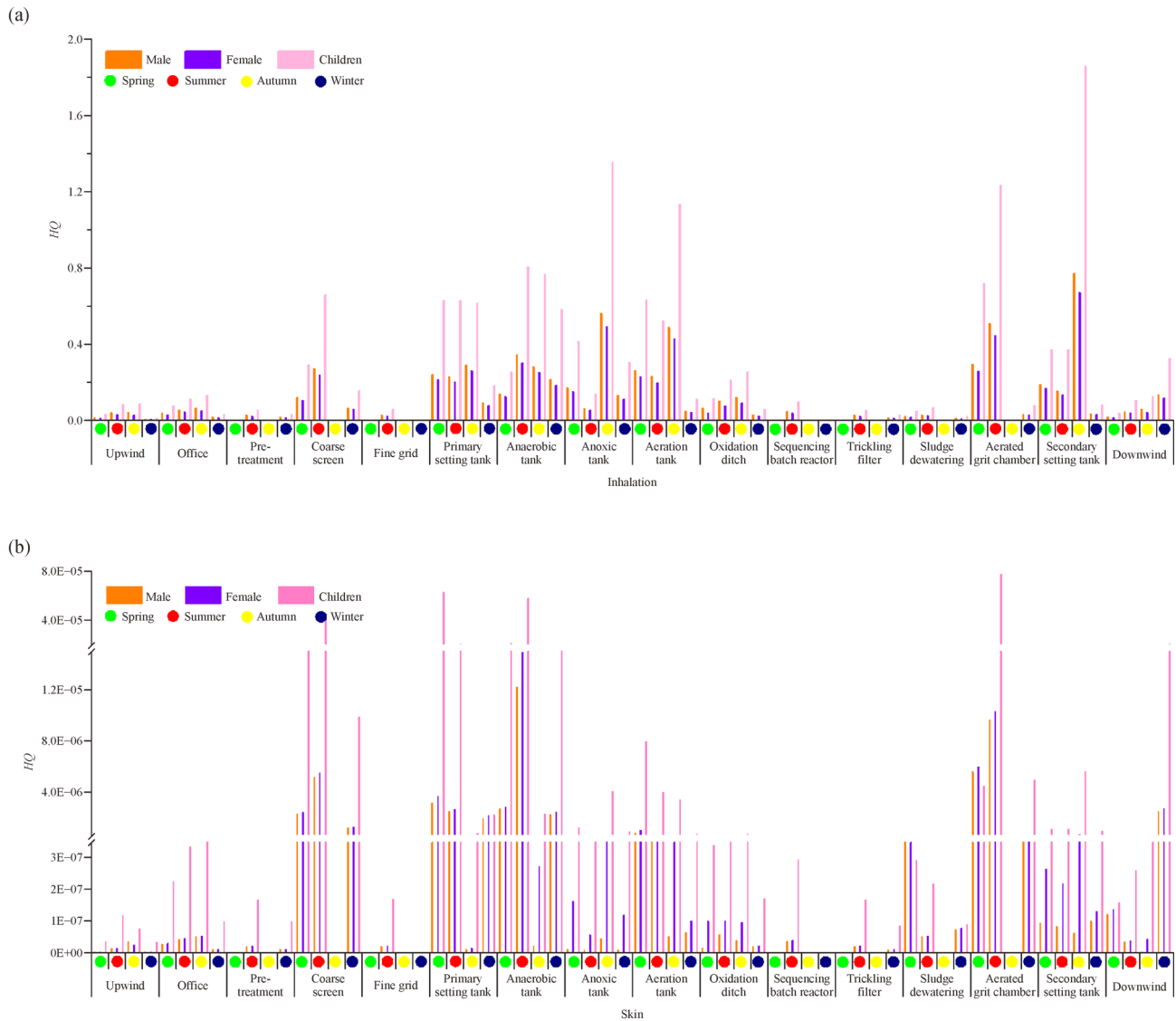


Fig. 3 Hazard quotient (HQ) of bioaerosols for adult male, female, and children within and/or around WWTPs: Inhalation (a) and skin (b) exposure route (Data Sources: Niazi et al., 2015; Ding et al., 2016; Yang et al., 2019a; Yang et al., 2019b).

WWTPs. The risk of human exposure can be assessed by the fraction of bacteria deposited in the air inhaled through the lungs. The HQ ranged from 0.0166 to 1.0600 in various studies. Exposure to $5 \mu\text{g}/\text{m}^3$ of fine particles each year will increase the danger of a heart attack by 13% (Stafoggia et al., 2014). Thus, workers in WWTPs should wear protective equipment to reduce their exposure to pathogenic microorganisms.

The HQ was calculated based on the concentration of bioaerosols presented in the air. The culture-dependent approach was traditionally utilized to assay the microbial population in air samples in most of the previous studies. However, the overall potential health risk of exposure to bioaerosols may be underestimated by using cultivation methods alone because the traditional isolation and culture technology can only obtain approximately 1% of the total

number of microorganisms in nature. In addition, factors such as the sensitivity of individuals to biological exposure and the interaction of bioaerosols with non-biological agents should also be considered in future risk assessments. A lack of source term data on emission rates, temporal characteristics of emissions, and a lack of dose-response and process-based exposures to the natural background bioaerosols are the current challenges for further exploring the exposure risk of bioaerosols released from WWTPs.

7 Prospects

By summarizing the concentrations, particle size distribution, composition, dispersion characteristics, and health

risks of bioaerosols in WWTPs, it was found that many potentially pathogenic substances in such bioaerosols are easily transmissible through the atmosphere. However, the current understanding of bioaerosols in WWTPs is not comprehensive, especially in terms of their component characteristics, harmfulness, and pollution control. Therefore, the following subjects are suggested for future research:

(1) The composition characteristics and temporal and spatial variation in bioaerosols should be identified in WWTPs. At present, potential pathogens, ARGs, endotoxins, and other toxic substances in bioaerosols have received much attention, but the research on allergens is relatively scarce. For the vast majority of people, allergens in bioaerosols may cause more severe discomfort. Therefore, it is necessary to more comprehensively and deeply identify the components that are harmful to human health in the bioaerosols and to construct a list of the components that are potentially harmful to human health in the bioaerosols of WWTPs. Simultaneously, the particle size distribution characteristics of these components have been studied in detail, including the abundance and diversity of different particle sizes, in order to provide a theoretical basis for subsequent research on the control standards and control technologies of bioaerosols.

(2) The spatial and temporal variation in bioaerosols in WWTPs should be clearly understood. At present, studies on bioaerosols are short-term, lack basic data, and are inconsistent, which makes it impossible to clearly reveal the interaction between bioaerosols in WWTPs and the surrounding atmospheric environment. Therefore, long-term, continuous, and differentiated studies on bioaerosol characteristics should be systematically conducted through real-time detection. In view of the difficulty of actual research, it is suggested to conduct studies in key areas and then gradually spread out so that a detailed database of bioaerosols from WWTPs can be formed.

(3) Development of a bioaerosol risk assessment system for WWTPs. Currently, available exposure risk assessments are based primarily on the total bacterial and chemical compositions of aerosols and do not account for the self-reproduction of pathogenic biomass once it enters the body. This may lead to an underestimation of risks by currently available assessments. Therefore, it is important to consider hazard characteristics, dose–response, and the potential probability and consequences of hazards as the risk assessment parameters to establish an accurate bioaerosol risk assessment system, which will help to more accurately understand the health risks of bioaerosols in WWTPs.

(4) Establish bioaerosol control standards and strategies for WWTPs. Presently, there are no dedicated bioaerosol control standards for WWTPs, which makes it challenging to establish bioaerosol control strategies. Regardless of the level of harm, it is essential to protect the workers in the plant, the surroundings, and even the wider population

from possible risks caused by bioaerosols. Therefore, based on a deepened understanding of the risks posed by WWTP bioaerosols, control standards and strategies should be established as soon as possible to effectively reduce the harm posed by WWTP bioaerosols to human health. For now, the most basic protective materials (gloves, masks, simple protective suits, etc.) should be fully equipped in the WWTP.

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