ADMS simulation and influencing factors of bioaerosol diffusion from BRT under different aeration modes in six wastewater treatment plants

Yanjie Wang[a,c], Liying Yang[a], Oliver Wild[b], Song Zhang[a], Kai Yang[a], Wenwen Wang[c], Lin Li[c]*

a College of Public Health, Zhengzhou University, Zhengzhou, Henan, 450001, PR China

b Lancaster Environment Centre, Lancaster University, United Kingdom, UK.

c State Key Laboratory of Environmental Aquatic Chemistry, Research Center for Eco-Environmental Sciences, Chinese Academy of Sciences, Beijing 100085, P. R. China.

Contact information of authors:

1. Yanjie Wang, Email: wangyanjie_2008@126.com;
2. Liying Yang, Email: yangliying1005xz@163.com;
3. Oliver Wild, Email: o.wild@lancaster.ac.uk;
4. Song Zhang, Email: zhangsong19980430@163.com;
5. Kai Yang, Email: yangk0806@163.com;
6. Wenwen Wang, Email: wwwang_st@rcees.ac.cn;
7. Lin Li, Email: leel@rcees.ac.cn;

Corresponding author: Lin Li, Tel./fax: +86 0371 67781795, Email: leel@rcees.ac.cn

Abstract

Bioaerosols produced by municipal wastewater treatment plants (MWTP) can spread in air, thereby polluting the atmosphere and causing safety hazards to workers and surrounding residents. In this study, the biological reaction tanks (BRTs) of six MWTPs undergoing typical processes in North China, Yangtze River Delta, and the Greater Bay Area were selected to set up sampling points and investigate the production characteristics of bioaerosols. The Atmospheric Dispersion Modelling System method was used to simulate the diffusion of bioaerosols in the MWTPs. The concentrations of bacteria and, specifically, intestinal bacteria in the bioaerosols ranged from 389 CFU/m^3 to 1,536 CFU/m^3 and 30 CFU/m^3 to 152 CFU/m^3, respectively, and the proportion of the intestinal bacteria was 8.85%. The concentration of soluble chemicals (SCs) in the bioaerosols was 18.36 μg/m^3–82.19 μg/m^3, and the main SCs found were Mg^{2+}, Ca^{2+}, and SO_4^{2-}. The proportion of intestinal bacteria (75.79%) produced via surface aeration by a BRT attached to large-sized bioaerosol particles was higher than that of a BRT undergoing the bottom aeration process (37.28%). The main microorganisms found in the bioaerosols included Moraxellaceae, Escherichia–Shigella, Psychrobacter, and Cyanobacteria. The generation of bioaerosols exhibited regional characteristics. The wastewater treatment scale, wastewater quality, and aeration mode were the main factors influencing bioaerosol production. Model simulation showed that, after 1 h, the diffusion distance of bioaerosol was 292 m–515 m, and the affected area was 42,895 m^2–91,708 m^2. The diffusion distance and range of the bioaerosols were significantly correlated with the concentration at the bioaerosol source and the aeration mode adopted by the BRTs. Wind speed and direction were two environmental factors that affected the diffusion of bioaerosols. With an increase in the diffusion distance, the concentration of microorganisms, intestinal bacteria, ions, and fine particles in the bioaerosols decreased significantly, resulting in a corresponding reduction in the exposure risk. This study provides new insights to help predict bioaerosol risks at MWTPs and identify safe areas around MWTPs. The study also provides a basis for selecting safe MWTP sites and reducing bioaerosol pollution risks.

Key words: Wastewater treatment, Bioaerosol, Generation characteristics, Trajectory prediction, Diffusion factors

1. Introduction

Wastewater treatment is considered a major source of bioaerosols. Most urban wastewater treatments use activated sludge processes that use microorganisms to degrade organic matter in wastewater into harmless substances. However, oxygen is required to maintain the growth of the microorganisms and the reaction. Therefore, biochemical treatment processes usually adopt aeration, and microorganisms may enter the air during the process of bioaerosol generation. When bioaerosols contain intestinal bacteria, viruses, or fungi and their spores that cause allergic reactions, they can cause diseases in the human respiratory tract (Brenner et al. 1988) . The exposure risk and pathogenesis of bioaerosol requires further investigation.

Bioaerosols containing bacteria and fungi have been detected in the air around biological reaction tanks (BRTs) during oxidation ditch, Sequencing Batch Reactor Activated Sludge Process(SBR), and Anaerobic-Anoxic-Oxic (A^2/O) processes(Li et al. 2011, Xu et al. 2018). In a previous study, the concentration of particles in bioaerosol produced as a result of wastewater treatment reached 1,233/m^3–6,533/m^3, and the microbial concentration reached more than 1,690 CFU/m^3 (Li et al. 2016). The particle size of bioaerosols in municipal wastewater treatment plants (MWTPs) is generally 2.1μm–3.3 μm (Laitinen et al. 1994). Bioaerosols produced by MWTPs may contain various pathogenic microorganisms, such as viruses (rotavirus, norovirus, adenovirus, hepatitis virus, and coliphages) and intestinal bacteria (Micrococcus, Bacteroides, Chryseobacterium, Pseudomonas, and Acinetobacter). They can cause infections following inhalation, swallowing, and skin contact (Gotkowska-Plachta et al. 2013, Wang et al. 2018b, Wang et al. 2018c). Previous studies on wastewater treatment of bioaerosols have mainly focused on their emission levels, particle size characteristics, and population structure (Li et al. 2011, Wang et al. 2018c).

Infective pathogens can be transported over long distances via airflow(van Doremalen et al. 2020). It was found that the concentration of microorganisms decreased significantly at 500 meters downwind of the bioaerosol source, and each MWTP presented its own bioaerosol diffusion characteristics. BRTs are sewage treatment facilities built outdoors. The bioaerosol generated by them will spread around the MWTP with the wind, bringing potential risks to the downwind area. Bioaerosol transport and dispersion models that describe the spatial and temporal distributions of

aerosol concentrations are commonly generated using computational fluid dynamics (CFD), which usually represent the turbulent transport of momentum and energy using eddy diffusivities in the corresponding governing equations (Hayashi et al. 2002, Yang et al. 2014). The viruses in public places, bioaerosols in hospital diagnosis and treatment sites and particulate matter emissions from aquaculture farms were calculated and simulated by this method to predict the infection caused by the spread of particulate pollutants and prevent the exposure risk brought about by them(Archer et al. 2019, Zhang et al. 2020). Analytical solutions obtained using the Gaussian-like Atmospheric Dispersion Modelling System (ADMS) based on the CFD principle can provide a quick prediction of the probability of taking in bioaerosol. In this study, the ADMS was used to study bioaerosol diffusion at the MWTP scale.

Because it is helpful to reduce bioaerosols and their exposure risk, studies on bioaerosol diffusion characteristics have attracted increasing attention. In this study, six MWTPs undergoing oxidation ditch, SBR, and A^2/O processes were investigated in three regions of China. The chemical composition of bioaerosol was analyzed by ion chromatography. High throughput sequencing was used to analyze microbial population in bioaerosols. The ADMS was used to simulate the diffusion of bioaerosols from the MWTPs to explore the diffusion law of bioaerosols. Analyzing the influencing factors of bioaerosol diffusion and evaluating the bioaerosol transport risk may contribute to risk predictions of bioaerosols from MWTPs, which, in turn, can provide a basis for safely selecting MWTP sites and reducing bioaerosol pollution risks.

2 Materials and methods

2.1 Municipal MWTP descriptions

Sampling sites for bioaerosol capture were set up 1.5 m above the aeration tanks of six MWTPs. The six MWTPs, NC-O and NC-A in North China, YZD-O and YZD-A in the Yangtze River Delta, GBA-O and GBA-A at the Greater Bay Area. Treatment processes are listed in Table S1. Samples were collected during the morning from 8 to 10 a.m. without disturbing the normal operation of the aerators. Meteorological parameters, e.g., temperature (Tem), relative humidity (RH), wind speed (WS) and solar radiation (SR), at the given sampling site were also recorded using portable equipment (Table S1). The air temperature and RH were determined using a Dewpoint Thermohygrometer (WD-35612, OAKTON, Germany). The WD and WS were recorded using a portable

anemometer (HD2303, Delta OHM, Padova, Italy) while the SR was measured using a portable irradiance meter (HD2302.0, Delta OHM, Italy).

2.2 Bioaerosol capture

A six-stage viable Andersen Impactor (228–9530 K, SKC Gulf Coast, Inc., Houston, TX, USA) with 400 holes was used to capture bioaerosols at each sampling site as described in a previous study (Wang et al. 2018c). A culture medium was kept under each stage of the sampler. The sampling time was typically 3 min at a speed of 28.3 L/min and the total impaction volume was 84.9 L (Table S1). Dishes were removed from the sampler when the required volume of air had been drawn. Bacteria and intestinal bacteria were cultured by LB and MacConkey Agar Medium (Hopebio, China), respectively. In this study, air samples were collected every hour (8 a.m., 9 a.m., 10 a.m.), and two parallel samples were collected each time. A total of 6 samples were taken at each sampling site and the data was the average of the 6 samples. After each sample was collected, the sampler was sterilised with a solution of 75% ethanol. Results were calculated as the geometric mean of the replicates, expressed as colony-forming units per cubic meter of air (CFU/m^3). A TH-150 medium flow sampler (Medium Flow sampler, Wuhan, China) was applied to collect the total suspended particulates (TSPs) in the atmosphere. The glass fiber membrane with diameter of 90 mm was utilized as the deposition medium. The particle rejection coefficient of medium was 99.90%. The glass fiber membranes were dried in a desiccator for 48 hours in advance.

2.3 Analysis method

2.3.1 Chemicals analysis

Concentrations of SO_4^{2-}, NO_3^-, Cl^-, PO_4^{3-}, and NO_2^- anions in each sample were determined with an ion chromatogram analyser (ICS-3000, Dionex, Sunnyvale, CA, USA). Concentrations of NH_4^+, Ca^{2+}, K^+, Mg^{2+}, and Na^+ cations in each sample were determined with an ion chromatogram analyser (IC plus 883, ion chromatography system, Metrohm, Herisau, Switzerland). The chromatography apparatus included a column oven, conductivity detector, manual injector, and chromatography workstation (Metrohm); AS19 Column and Metrosep C 4150/4.0 separation column; eluent: 20 mM

NaOH (anions), 2.0 mM HNO_3 (cations); column temperature: 30°C; flow-rate: 1.0 mL/min; inject volume: 10 L. The detection limit of the method was less than 0.05 mg/L for anions and cations.

2.3.2 Microbial population

Illumina MiSeq high-throughput sequencing was applied to analyse the bacteria population in the bioaerosols. DNA was extracted, purified. and sequenced as described in previous studies(Wang et al. 2018c). After raw FASTQ files were demultiplexed and quality filtered, operational taxonomic units (OTUs) were clustered with a 97% similarity cut-off and chimeric sequences were identified and removed. The taxonomy of each 16S rRNA gene sequence was analysed using a confidence threshold of 70% (Amato et al. 2013, Jiang et al. 2015). Each sample was normalized at the same sequence depth (34,536 reads). The alpha and beta diversity and similarity of the bacterial community's structure were analysed(Table S2).

2.3.3 Simulation on bioaerosol

Atmospheric Dispersion Modelling System (ADMS) was an advanced dispersion model used to obtain the air quality impact of existing and proposed industrial installations which was originally developed for regulatory authorities in the UK. It has been widely applied to simulate the diffusion of gases or particles in various situations (Hood et al. 2020, Jaffe et al. 2020). ADMS was utilized in the simulation of bioaerosol diffusion in this study. Considering the change of microbial activity in bioaerosols, this study adjusted and improved the model parameters, such as the type selection of particles representing aerosols, dry and wet sedimentation, boundary layer processes, chemical reactions and topography. The adjusted model is compared with the observed results and fitted with the actual measured values. In order to simulate the diffusion characteristics of bioaerosols accurately, we measured and obtained ground structures and terrain conditions in more detail. The aerosols were simulated from the six MWTPs studied in this study. The simulated area size was within 1000000 m^2, Meteorological data are obtained from the meteorological Bureau (http://www.weather.com.cn/) and underlying surface data are obtained from field monitoring obtain meteorological parameters, and input the obtained data including temperature, humidity, illumination, wind direction and wind speed; Underlying surface data such as terrain building height into building modules.

2.4 Exposure Risk assessment

The average daily doses of a respiratory inhalation model were calculated using Eqs. (1) (Liu et al. 2018, Vilavert et al. 2012).

$$\mathrm{ADDinhalation} = \frac{\mathrm{c}\times\mathrm{InR}\times\mathrm{EF}\times\mathrm{ET}}{\mathrm{BoW}\times\mathrm{ALE}} \quad (1)$$

Where $ADD_{inhalation}$ is the average daily dose to the respiratory system (CFU/d/kg), c is the average bioaerosol concentration (CFU/m^3), InR is the inhalation rate (m^3/d), EF is the exposure frequency (d/yr), ET is the respiratory inhalation exposure time (yr), ALE is their average life expectancy (d), and BoW is the body weight of the exposed person (kg). The dose rate was estimated from the average bioaerosol concentrations. As the people exposed in this study were mainly adults, the objects of the greatest risk evaluation were adults. Most of the exposure parameters of Chinese people proposed by Wang et al. (2009) were employed in this study (Wang et al. 2009). Some parameters such as average body weight (BoW) and average time (ALE), were retrieved from the Exposure Factors Handbook of the Chinese Population (Adult) based on the behavioural characteristics of environmental exposure in China in 2017.

Functional Annotation of Prokaryotic Taxa（FAPROTAX）was a functional annotation database of culturable microorganisms(Liang et al. 2019). Substitute the 16S based OTU classification in the high-throughput sequencing of microorganisms into the python script linked to the OTU classification and the FAPROTAX database to obtain the prediction results of microbial community functions(Parfrey et al. 2016). FAPROTAX was utilized to forecast bioaerosols hazard in this study.

3. Results

3.1 Aerosol emission

3.1.1 Aerosol emission level

Figure 1 demonstrates the bacteria and intestinal bacteria in bioaerosol escaped in the BRT section of the six MWTPs. The concentrations of bioaerosols presented upwind were also measured with 275 CFU/m^3 for NC-O, 177 CFU/m^3 for NC-A, 75 CFU/m^3 for YZD-O, 42 CFU/m^3 for YZD-A, 99 CFU/m^3 for GBA-O, and 51 CFU/m^3 for GBA-A, respectively. The emission level of bioaerosols found at all BRTs were higher than those presented in the air upwind (Table S1). The concentration of airborne bacteria at

the MWTPs ranged from 389 CFU/m^3 to 1,536 CFU/m^3, with the concentrations in the NC-O, NC-A, YZD-O, YZD-A, GBA-O, and GBA-A being 1,536 CFU/m^3, 1,009 CFU/m^3, 1,184 CFU/m^3, 554 CFU/m^3, 1,056 CFU/m^3, and 389 CFU/m^3, respectively. MWTPs using the surface aeration method had a higher concentration of bacteria (1,258 CFU/m^3). Intestinal bacteria were found at each MWTP, with concentrations ranging from 30 CFU/m^3 to 1,52 CFU/m^3, accounting for 8.04% of the total bacterial population. MWTPs in the NC area generated more bacteria and, specifically, intestinal bacteria (with averages of 1,273 CFU/m^3 and 140 CFU/m^3, respectively) than did those in the other areas (which had averages of 796 CFU/m^3 and 47 CFU/m^3, respectively). Figure 2 shows the soluble chemicals (SCs) detected in bioaerosols at the six MWTPs. SCs were detected at each MWTP, and the amount of ten kinds of SCs ranged from 18.36μg/m^3–82.19 μg/m^3. Bioaerosols at the MWTPs of the NC and GBA areas had higher SC contents (with an average of 73.74 μg/m^3) than did those in the YZD area (with an average of 37.89 μg/m^3). Mg^{2+}, Ca^{2+}, and SO_4^2 were the major SCs in the bioaerosols at each MWTP, accounting for 29.84% of the total SC content.

3.1.2 Aerosol particle size distribution

The size distributions of the bioaerosol particles to which the airborne and intestinal bacteria attached at each MWTPs are shown in Figure 3. The aerodynamic cut-size diameters of the Six Stage Viable Andersen Cascade Impactor are: over 7.0 μm in stage 1, 4.7-7.0 μm in stage 2, 3.3-4.7 μm in stage 3, 2.1-3.3 μm in stage 4, 1.1-2.1 μm in stage 5, 0.65-1.1 μm in stage 6. The microbial aerosol particles collected in this study were classified as fine particles which picked from the stage 5 and the stage 6, and coarse particles that sampled from the stage 1 to the stage 4. The inhalation of particles smaller than 10 μm has detrimental health effects, such as causing asthma and cardiovascular disease (Urbán et al. 2015, Vestlund et al. 2014). At the sampling sites NC-O, YZD-O, and GBA-O, 69.73% to 80.56% of the intestinal bacteria were attached to coarse particles with an average concentration of 64 CFU/m^3, while the average concentration of the fine particles was 23 CFU/m^3. At the sampling sites NC-A, YZD-A, and GBA-A, 62.72% of the intestinal bacteria on average were attached to fine particles. The concentrations of fine and coarse particles were 4 CFU/m^3-24 CFU/m^3 and 10 CFU/m^3-68 CFU/m^3, respectively.

3.1.3 Microbial population in bioaerosols

This study analyzed the microbial populations at each MWTP. Table S3 shows the composition and proportion of the microorganisms at each MWTP. The main microorganism species in the bioaerosols varied among MWTPs. Moraxellaceae (11.17%), Pseudomonas (7.11%), and Chroococcidiopsis (6.64%) dominated the bacterial population in NC-A. Pseudomonas, Escherichia–Shigella, and Psychrobacter accounted for 84.63% of the bacterial population in NC-O. Cyanobacteria was the dominant microorganism in YZD-A, accounting for 80.46% of the overall bacterial population, while in YZD-O, Cyanobacteria (28.65%), Candidatus (8.76%), and Saccharibacteria (4.55%) were the most abundant. The dominant species in GBA-A were Sphingomona (9.74%), Sphingomonadales (6.97%), and Mitochondria (6.29%), while Peptostreptococcaceae (13.79%), Mycobacterium (4.75%), and Sphingobacteriaceae (4.47%) were the dominant bacteria in GBA-O. Intestinal bacteria, including Moraxellaceae, Pseudomonas, Acinetobacter, Arcobacter, Neisseriaceae, Escherichia–Shigella, Mycobacterium, Saccharibacteria, Romboutsia, Serratia, and Flexibacter, were detected in bioaerosols at the MWTPs. Pseudomonas is a common pathogenic bacterium that can cause endocarditis, osteomyelitis, pneumonia, urinary tract infections, gastrointestinal infections, meningitis, and sepsis(Rice et al. 2012). Diseases caused by Pseudomonas are becoming more common(Gomez-Gamboa et al. 2021). Acinetobacter mainly causes respiratory and urinary tract infections, sepsis, and secondary meningitis (Gao et al. 2014, Gou et al. 2016). Serratia can cause urinary tract, respiratory tract, and wound infections(Kowalski et al. 2017). Escherichia–shigella is a common pathogenic bacterium that can cause extra-intestinal infections, acute diarrhea, bacillary dysentery, and other diseases (Kotloff et al. 2018, Rodriguez-Angeles 2002). Therefore, in addition to strengthening the personal protection of workers, such as by wearing masks and goggles, BRTs, especially the oxidation ditch, need to be equipped with protective covers and other facilities to prevent the escape of bioaerosols.

3.1.4 Bioaerosol generation

There were differences in the number, particle size distribution, and composition of total bacteria and Enterobacteriaceae in aerosols released from each sewage treatment plant, and the concentrations of SCs in the aerosols differed. Similar results were reported in previous studies(Wang et al. 2018a, Wang et al. 2018d). Reasons for this difference might be differences in regional characteristics and aeration modes. The water quality and environmental conditions of sewage treatment plants differ regionally.

Aerosol generation is related to water quality and environmental conditions. Therefore, the concentration of microorganisms generated in NC, which has a high concentration of microorganisms in its sewage, was high. The diffusion of bioaerosols produced by wastewater treatment plants undergoing surface oxygenation and bottom oxygenation differed within the same area. This may be related to the different mechanisms of bioaerosol generation as a result of bottom aeration and surface aeration. The bottom aeration process filled the bottom of the water body with gas, and the gas formed bubbles at the bottom of the liquid and gradually rose to the liquid surface. During this process, sludge flocs from the bottom and middle of the liquid were carried to the surface. During the process of surface rupture, the bubbles reaching the surface transported substances in the sewage to the air, resulting in a loss of water and aerosolizing in the air, thereby forming aerosols. Surface aeration oxygenation facilities are located at liquid surfaces and often disturb water surfaces owing to the motion of rotating brushes. After flapping and raising, water surface liquid is wrapped with substances in sewage and is transported into the air, and the splashing of large droplets, water loss, and atomization of small droplets leads to bioaerosols generation near the specific brush. Data on aerosol production characteristics in this study also confirmed that both surface and bottom aeration produces bioaerosols. The higher the amount of bioaerosols produced, the higher the concentration in the air at 200 m-DW. Compared with surface aeration, bottom aeration produces less bioaerosol. The concentration in the air emitted by bottom aeration was also lower at 200 m-DW.

3.2 Bioaerosol diffusion

The ADMS was used to calculate the diffusion of bioaerosols, microorganisms, particles, and water-soluble ions. The FLOWSTAR model was used to calculate flow and turbulence fields over the terrain by introducing meteorological parameters. Wind direction and wind speed are the dominant factor influencing bioaerosol diffusion direction and range (Wei et al. 2020). The rose chart of the wind direction in North China, the Greater Bay Area, and the Yangtze River Delta are shown in Figure 5. The dominant wind directions in North China were south (44.66%) and north (22.74%). East (20.55%), northeast (16.71%), and north (15.07%) winds were dominant in Yangtze River Delta. North (25.21%), southeast (21.10%), and northeast (20.01%) winds were dominant in the Greater Bay Area.

3.2.1 Aerosol diffusion direction

The concentrations of bioaerosols produced at the MWTPs were used to simulate the diffusion of bacteria in air using the ADMS model (Figure 6 and Table S3). The simulation results showed that bioaerosols from the MWTPs diffused evenly. After 1 h of diffusion, high aerosol concentrations were found close to the generation source, and their diffusion was distributed in clumps. MWTPs in the same region had similar diffusion directions that closely correlated to the wind direction. The diffusion trends in NC-A and NC-O had a southwest-northwest (139°) elliptic distribution. The diffusion trends in GBA-A and GBA-O exhibited southeast to northwest (164°) elliptic distributions. Diffusion in YZD-A exhibited an elliptic distribution with a southwest-northeast (243°) trend, while that in YZD-O exhibited a west-east trend (285°) elliptic distribution.

3.2.2 Aerosol diffusion scope

The concentrations of bacteria in the aerosol diffusion direction at 100 m (100 m-DW; the boundary of the MWTPs) and 200 m (200 m-DW; residential areas) varied among the MWTPs. The bioaerosol concentrations at 100 m-DW and 200 m-DW in NC-A and NC-O ranged from 63 CFU/m^3–75 CFU/m^3 and 25 CFU/m^3–35 CFU/m^3, respectively. The spread of bioaerosol from YZD-A and YZD-O at 100 m-DW and 200 m-DW ranged from 41 CFU/m^3–66 CFU/m^3 and 18 CFU/m^3–50 CFU/m^3, respectively. The spread of bioaerosol in GBA-A and GBA-O also ranged from 20 CFU/m^3–72 CFU/m^3 and 50 CFU/m^3–55 CFU/m^3 at 100 m-DW and 200 m-DW, respectively. The concentration of intestinal bacteria at 100 m-DW and 200 m-DW at each MWTP ranged from 1 CFU/m^3–8 CFU/m^3 and 1 CFU/m^3–3 CFU/m^3, respectively. Bacteria attached to fine particles also decreased with increasing distance, with concentrations ranging from 4 CFU/m^3–22 CFU/m^3 and 3 CFU/m^3–11 CFU/m^3 at 100 m-DW and 200 m-DW, respectively. The total concentration of SCs decreased to 3.62 μg/m^3 and 2.02 μg/m^3 on average at 100 m-DW and 200 m-DW, respectively.

The bioaerosol diffusion distance (292–515 m) and affected area (42,895–91,708 m^2) differed among the MWTPs. Aerosols in NC-O spread 423 m in the northwest direction and affected an area of 61,385 m^2. The microorganisms in NC-A spread 301 m northwest, with an affected diffusion area of approximately 42,895 m^2. Microorganisms in YZD-O spread 307 m northwest, affecting an area of 47,333 m^2. The diffusion

distance of microorganisms in YZD-A was 554 m, and the diffusion area was 80,614 m^2. Microorganisms in GBA-O spread 292 m northwest, with an affected diffusion area of approximately 48,073 m^2. Microorganisms in GBA-A spread 515 m northwest, with an affected diffusion area of approximately 91,708 m^2. The concentration at the diffusion edge was as low as in the range of 2 CFU/m^3–20 CFU/m^3.

3.2.3 Factors influencing diffusion

Influence of source strength on the diffusion range

Diffusion analysis of aerosols generated at the MWTPs indicated that there were differences in the aerosol diffusion results during the BRT stage. Mechanical disturbances were major causes of aerosol generation. The high microbial content and intense sewage aeration led to a large amount of aerosol being generated in the BRT section, as supported by previous studies(Wang et al. 2019, Yang et al. 2019). Therefore, the bioaerosol concentrations in NC-O were higher at 200 m-DW than at the MWTPs in the other areas. The level of bioaerosol generated at the source influenced bioaerosol diffusion; the higher the level at the source, the more microorganisms dispersed in the downward direction.

The species and concentrations of bacteria varied with place, depending on the process selected and meteorological parameters (e.g., temperature, RH, and WS). Canonical correlation analysis (CCA) was conducted to describe the influence of these factors on bacterial populations in bioaerosols (Figure S1) (Li et al. 2017). The results demonstrated that environmental factors influence bacterial emissions. Among them, RH, temperature, SR, and WS were the major factors influencing the level of airborne bacteria in the air, which is consistent with results obtained in previous reports(Yan et al. 2019a). The RH, temperature, and SR were positively correlated with airborne intestinal bacteria release and dispersal, whereas WS was negatively correlated with airborne intestinal bacteria release and dispersal (Table 3). High temperatures and RH favor microbiological growth, whereas strong winds help disperse bioaerosol particles. Owing to the suitable temperature, appropriate RH, and low WS, higher bioaerosol concentrations were maintained in the air. Similar results were obtained from CCA of strain-influencing factors. Some microbes were sensitive to meteorological conditions. CCA results also showed that environmental factors have different effects on different types of microorganisms. The RH had a strong positive effect on Pseudomonas,

Psychrobacter, and Serratia that were positively affected by temperature, and most of the bacteria were negatively correlated with WS and SR.

Changes in microbial concentrations in bioaerosols are related to WS. The results showed that a high WS is conducive to aerosol diffusion, whereas light intensity has the opposite effect. A high RH is favorable for bacterial activity and diversity during diffusion. WS and RH are major factors influencing aerosol diffusion and are important parameters in estimating aerosol diffusion. The range of influence of bioaerosol diffusion is related to WS; the higher the WS, the greater the range of influence of aerosols. Pseudomonas, Escherichia–Shigella, Psychrobacter, and Serratia were strongly affected by environmental factors. Intestinal bacteria accounted for approximately 8.85% of the total bacterial population and may pose risks to human health following breathing or skin contact.

Influence of meteorological parameters on diffusion

Temperature, RH, SR, WS, and other environmental conditions influenced aerosol diffusion. Sampling of the six MWTPs occurred during summer at an air temperature of 31.9℃–37.2 ℃. The optimal growth temperature for most bacteria in bioaerosols is 10℃–30 ℃ (Rai et al. 2021). The RH ranged from 31.5%–57.5% at the MWTPs. The concentration of bioaerosols in the air at the MWTP was high when the RH was high. The CCA results also showed that a high RH is favorable for bacterial survival in air. Most microorganisms require water for survival; therefore, the RH is positively correlated with the abundance of microorganisms. A higher RH in air is conducive to the survival of microorganisms. (Yan et al. 2019b). The SR was in the range of 216.16 W/m^2–585.6 W/m^2, and the CCA result revealed a negative correlation with microorganism abundance. Most bacteria are sensitive to light, and UV light may inactivate and kill microorganisms, thereby negatively affecting the diffusion of bioaerosols (Fahimipour et al. 2018). WS had an important relationship with the diffusion speed and distance of the bioaerosols. The wind direction affected the diffusion direction. Therefore, temperature, RH, SR, and WS were affecting bioaerosol diffusion. When simulating diffusion, it was suggested to consider the influence of pathogenic bacteria, particle size, and ions to make the evaluation results more accurate and conducive to risk prevention.

3.3.1 Risk assessment

As bioaerosols contain intestinal bacteria, they can be used to estimate the exposure risk of people downwind of aerosols produced by sewage plants. The results of an exposure dose calculation and a risk assessment are shown in Figure 7. The ranges of $HQ_{inhalation}$ inside the MWTPs were 3.59×10^{-2}–1.42×10^{-1}. The concentration of bacteria was higher in the air undergoing surface aeration (1.16×10^{-1}) than in that undergoing submerged aeration (6.01×10^{-2}). The hazard quotient (HQ) and HI decreased with increasing distance downwind. The $HQ_{inhalation}$ of the bacteria at 100 m-DW and 200 m-DW at each MWTP ranged from 1.85×10^{-3}–6.93×10^{-3} and 9.24×10^{-4}–4.62×10^{-3} respectively, which were 5.88% and 3.28% on average in the BRT section, respectively. The $HQ_{inhalation}$ of the bacteria in air undergoing surface aeration at 100 m-DW and 200 m-DW were 6.56×10^{-3} and 4.16×10^{-3}, respectively, which were higher than those in air undergoing submerged aeration (3.82×10^{3} and 1.63×10^{-3}, respectively). Bioaerosol diffusion probably exerted higher risks at 100 m-DW and 200 m-DW in GBA-A than at the other MWTPs due to its higher $HQ_{inhalation}$ (1.66×10^{-3} and 3.79×10^{-3}, respectively). The $HQ_{inhalation}$ of intestinal bacteria at 100 m-DW and 200 m-DW at each MWTP ranged from 9.24×10^{-5}–7.39×10^{-4} and 9.24×10^{-4}–4.62×10^{-3}, respectively. The individual risks were low at the sites investigated, and they remained below the acceptable levels stipulated by the USEPA (1 for noncarcinogenic pollutants). As the risk to the skin was several orders of magnitude lower than that following inhalation and was negligible, respiratory inhalation was regarded as the main pathway for microbial bioaerosol entry into the human body. The HI reached its highest level in NC-O. The order of exposure HQ following inhalation was: children>adult males>adult females. It is worth noting that the exposure hazard for children is generally higher than that for adults owing to their lower weight.

Figure 2S demonstrated the prediction results of FAPROTAX microbial community function at BRT of all MWTPs. A total of 7 dominant predicted functional groups were detected, including chemoheterotrophy, aerobic chemoheterotrophy, nitrate reduction, fermentation, human associated functional group and pathogenic functional groups (animal parasites or symbionts, human pathogens). Among them, human associated functional group, human pathogens and animal parasites or symbionts, were higher at NC-A, indicating that the microorganism at NC-A has potential risk to human health.

3.3.2 Factors influencing exposure risk

The results of the evaluation obtained using the aerosol generation source and total bacterial value diffused to the outside indicated that the exposure risk of bioaerosols was negligible. The MWTPs in the NC area had higher internal and downwind risks. Meanwhile, because of the high WS, the NC area had a wider bioaerosol exposure risk range. It is worth noting that, compared with bottom aeration, the risk of bioaerosols produced by surface aeration was greater. With an increase in the distance from the aerosol generation source, the concentration of bioaerosols in the air gradually decreased, and the exposure risk decreased accordingly.

Impact of microorganism abundance on bioaerosol risk

The microorganism concentrations were used to calculate the bioaerosol risk in this study. However, this risk of bioaerosol in reality might be higher than evaluated. Microorganisms present in the air may multiply during transportation. (Fahimipour et al. 2018). In addition, air contains Mg^{2+}, Ca^{2+}, SO_4^{2-}, and other chemicals, which may create a microenvironment suitable for the survival of airborne microorganisms. With the progression of diffusion, microorganisms in air generally lose their activity or die. However, aerosols produced in water may behave differently from those in other environments, such as soil and vegetation. Aeration generates a droplet membrane that breaks the water surface to generate aerosols. Therefore, most aerosols produced in water contain microorganisms in the droplet membrane. The droplets help the microorganisms to attach to the aerosol particles, reduce the direct irradiation of some ultraviolet rays, protect the viability of some microorganisms, and protect microorganisms from external damage (e.g., wind-induced damage) or death. Droplet membranes also increase the humidity in the environment of microorganisms and allow bacteria to survive. Meanwhile, water and soluble chemicals in the droplet membrane can provide nutrients for microorganisms.

The current risk assessment calculation method does not consider intestinal bacteria, small particles, SCs, or other factors. Table S3 presents the species and genera of intestinal bacteria. After diffusion, intestinal bacteria, SCs, and fine particles were still present. The detected concentrations of intestinal bacteria were 2 CFU/m^3–8 CFU/m^3 and 1 CFU/m^3–3 CFU/m^3 when intestinal bacteria spread to 100 DW and 200 DW, respectively; therefore, intestinal bacteria pose a potential risk. Intestinal bacteria,

including Pseudomonas, Acinetobacter, and Serratia, were detected in the aerosols. They may proliferate and maintain their activity during diffusion, thereby increasing the risk associated with aerosols. Second, although the number of intestinal bacteria was low, the current calculation method does not consider pathogenicity. If bacteria are pathogenic, although the risk of exposure is low, the risk of long-term exposure for on-site workers cannot be ignored.

Fine particles of 4 CFU/m^3–24 CFU/m^3 and 3 CFU/m^3–11 CFU/m^3 were also detected at 100-DW and 200-DW, respectively. The exposure risk was mainly related to the concentration of bacteria. The number of bacteria decreased gradually from the proximal end to the distal end of diffusion, and the amount of coarse and fine particles also decreased. At the proximal end of diffusion, the aeration mode had a greater impact. Surface aeration produces coarser particles that mainly affect the upper respiratory tract. Because of the sedimentation of coarse particles, the distal end (100 m-DW and 200 m-DW) of diffusion is dominated by fine particles, which may enter deeper parts of the human body.

Soluble ions at concentration of 1.36μg/m^3–5.60 μg/m^3 and 0.60μg/m^3–3.89 μg/m^3 were detected at 100 m-DW and 200 m-DW, respectively. Soluble chemicals diffused far into the air to provide nutrients for microorganisms and help them maintain vitality. Therefore, the bioaerosols produced by sewage treatment plants may contain more active intestinal bacteria.

In the current risk assessment calculation method, intestinal bacteria, fine particles, SCs, and other factors are not considered. Therefore, on-site protection should be strengthened. Suggestions for protecting workers include wearing masks and protective clothing.

4. Conclusions

Concentrations, particle size distribution, population, and soluble ion number of the total bacteria and intestinal bacteria of bioaerosol in the six MWTPs present variation because of regional differences and aeration modes. MWTPs in North China generated more bioaerosol compared with other areas, while MWTPs with surface aeration process produced higher amounts of bioaerosol than submerge aeration process. Wind

direction affected the diffusion direction. The diffusion distance and range were related to the WS and source strength of the bioaerosol. The dispersion of bioaerosols with different particle sizes varied. Surface aeration resulted in more aerosol diffusion. With an increase in the distance from the MWTPs, the exposure risk gradually decreased. The number of microorganisms, pathogenic bacteria, ions and fine particles affect the risk of diffusion. Since there may be more than one source of bioaerosol production, this part of the study can be used to distinguish whether bioaerosols in residential exposure areas come from MWTPs, thus speculate the impact of bioaerosol generated by MWTPs on surrounding area. This study simulated the bioaerosol diffusion generated from MWTP, helping MWTPs' sites selection and reducing bioaerosols' possible risks.

[Acknowledgements]

We would like to thank Editage (www.editage.cn) for English language editing.

[Funding sources]

This project is supported by the special fund of State Key Joint Laboratory of Environment Simulation and Pollution Control (21K01ESPCR).

[References]

Amato, K.R., Yeoman, C.J., Kent, A., Righini, N., Carbonero, F., Estrada, A., Gaskins, H.R., Stumpf, R.M., Yildirim, S. and Torralba, M. (2013) Habitat degradation impacts black howler monkey (Alouatta pigra) gastrointestinal microbiomes. Isme Journal 7(7), 1344-1353.

Archer, S.D.J., Lee, K.C., Caruso, T., Maki, T., Lee, C.K., Cary, S.C., Cowan, D.A., Maestre, F.T. and Pointing, S.B. (2019) Airborne microbial transport limitation to isolated Antarctic soil habitats. Nature Microbiology 4(7), 925-932.

Brenner, K.P., Scarpino, P.V. and Clark, C.S. (1988) Animal viruses, coliphages, and bacteria in aerosols and wastewater at a spray irrigation site. Applied & Environmental Microbiology 54(2), 409-415.

Fahimipour, A.K., Hartmann, E.M., Siemens, A., Kline, J., Levin, D.A., Wilson, H., Betancourt-Román, C.M., Brown, G.Z., Fretz, M., Northcutt, D., Siemens, K.N., Huttenhower, C., Green, J.L. and Van Den Wymelenberg, K. (2018) Daylight exposure modulates bacterial communities associated with household dust. Microbiome 6(1), 175.

Gao, J., Zhao, X., Bao, Y., Ma, R., Zhou, Y., Li, X., Chai, T. and Cai, Y. (2014) Antibiotic resistance and OXA-type carbapenemases-encoding genes in airborne Acinetobacter baumannii isolated from burn wards. Burns 40(2), 295-299.

Gomez-Gamboa, L., Perozo-Mena, A., Bermudez-Gonzalez, J., Villavicencio, C., Villasmil, J., Ginestre, M.M. and Velasquez, J. (2021) Detection of carbapenemase-producing bacteria in a public healthcare

center from Venezuela. Journal of Infection in Developing Countries 15(1), 163-167.
Gotkowska-Plachta, A., Filipkowska, Z., Korzeniewska, E., Janczukowicz, W., Dixon, B., Golas, I. and Szwalgin, D. (2013) Airborne Microorganisms Emitted from Wastewater Treatment Plant Treating Domestic Wastewater and Meat Processing Industry Wastes. Clean-Soil Air Water 41(5), 429-436.
Gou, H., Lu, J., Li, S., Tong, Y., Xie, C. and Zheng, X. (2016) Assessment of microbial communities in PM1 and PM10 of Urumqi during winter. Environmental Pollution 214, 202-210.
Hayashi, T., Ishizu, Y., Kato, S. and Murakami, S. (2002) CFD analysis on characteristics of contaminated indoor air ventilation and its application in the evaluation of the effects of contaminant inhalation by a human occupant. Building & Environment 37(3), 219-230.
Hood, C., Stocker, J., Seaton, M., Johnson, K. and Carruthers, D. (2020) Comprehensive evaluation of an advanced street canyon air pollution model. Journal of the Air & Waste Management Association.
Jaffe, D.A., O'Neill, S.M., Larkin, N.K., Holder, A.L. and Rappold, A.G. (2020) Wildfire and prescribed burning impacts on air quality in the United States. Journal of the Air & Waste Management Association 70(7).
Jiang, W., Liang, P., Wang, B., Fang, J., Lang, J., Tian, G., Jiang, J. and Zhu, T.F. (2015) Optimized DNA extraction and metagenomic sequencing of airborne microbial communities. Nature Protocols 10(5), 768.
Kotloff, K.L., Riddle, M.S., Platts-Mills, J.A., Pavlinac, P. and Zaidi, A.K.M. (2018) Shigellosis. Lancet 391(10122), 801-812.
Kowalski, M., Wolany, J., Pastuszka, J.S., Płaza, G., Wlazło, A., Ulfig, K. and Malina, A. (2017) Characteristics of airborne bacteria and fungi in some Polish wastewater treatment plants. International Journal of Environmental Science and Technology 14(10), 1-12.
Laitinen, S., Kangas, J., Kotimaa, M., Liesivuori, J., Martikainen, P.J., Nevalainen, A., Sarantila, R. and Husman, K. (1994) Workers' exposure to airborne bacteria and endotoxins at industrial wastewater treatment plants. American Industrial Hygiene Association Journal 55(11), 1055-1060.
Li, J., Zhou, L., Zhang, X., Xu, C., Dong, L. and Yao, M. (2016) Bioaerosol emissions and detection of airborne antibiotic resistance genes from a wastewater treatment plant. Atmospheric Environment 124, 404-412.
Li, L., Gao, M. and Liu, J. (2011) Distribution characterization of microbial aerosols emitted from a wastewater treatment plant using the Orbal oxidation ditch process. Process Biochemistry 46(4), 910-915.
Li, Z., Lu, L., Guo, J., Yang, J., Zhang, J., He, B. and Xu, L. (2017) Responses of spatial-temporal dynamics of bacterioplankton community to large-scale reservoir operation: a case study in the Three Gorges Reservoir, China. Scientific Reports 7, 12.
Liang, S., Deng, J., Jiang, Y., Wu, S. and Zhu, W. (2019) Functional Distribution of Bacterial Community under Different Land Use Patterns Based on FaProTax Function Prediction. Polish Journal of Environmental Studies 29(2).
Liu, H., Zhang, Z., Wen, N. and Wang, C. (2018) Determination and risk assessment of airborne endotoxin concentrations in a university campus. Journal of Aerosol Science 115, 146-157.
Parfrey, Wegener, L., Louca, Stilianos, Doebeli and Michael (2016) Decoupling function and taxonomy in the global ocean microbiome. Science.
Rai, S., Singh, D.K. and Kumar, A. (2021) Microbial, environmental and anthropogenic factors influencing the indoor microbiome of the built environment. J Basic Microbiol 61(4), 267-292.
Rice, S.A., van den Akker, B., Pomati, F. and Roser, D. (2012) A risk assessment of Pseudomonas aeruginosa in swimming pools: a review. Journal of Water and Health 10(2), 181-196.

Rodriguez-Angeles, G. (2002) Diagnosis and main characteristics of Escherichia coli pathogenic groups. Salud Publica De Mexico 44(5), 464-475.
Urbán, E., Horváth, Z., Sóki, J. and Lázár, G. (2015) First Hungarian case of an infection caused by multidrug-resistant Bacteroides fragilis strain. Anaerobe 31, 55-58.
van Doremalen, N., Bushmaker, T., Morris, D.H., Holbrook, M.G., Gamble, A., Williamson, B.N., Tamin, A., Harcourt, J.L., Thornburg, N.J., Gerber, S.I., Lloyd-Smith, J.O., de Wit, E. and Munster, V.J. (2020) Aerosol and Surface Stability of SARS-CoV-2 as Compared with SARS-CoV-1. New England Journal of Medicine 382(16), 1564-1567.
Vestlund, A.T., Al-Ashaab, R., Tyrrel, S.F., Longhurst, P.J., Pollard, S.J.T. and Drew, G.H. (2014) Morphological classification of bioaerosols from composting using scanning electron microscopy. Waste Management 34(7), 1101-1108.
Vilavert, L., Nadal, M., Figueras, M.J. and Domingo, J.L. (2012) Volatile organic compounds and bioaerosols in the vicinity of a municipal waste organic fraction treatment plant. Human health risks. Environmental Science and Pollution Research 19(1), 96-104.
Wang, Y., Huachun, L., Lin, L., Kaixiong, Y., Jiuhui, Q. and Junxin, L. (2018a) Chemicals and microbes in bioaerosols from reaction tanks of six wastewater treatment plants: survival factors, generation sources, and mechanisms. Scientific Reports 8(1), 9362.
Wang, Y., Lan, H., Li, L., Yang, K., Qu, J. and Liu, J. (2018b) Chemicals and microbes in bioaerosols from reaction tanks of six wastewater treatment plants: survival factors, generation sources, and mechanisms. Scientific Reports 8(1), 9362.
Wang, Y., Li, L., Han, Y., Liu, J. and Yang, K. (2018c) Intestinal bacteria in bioaerosols and factors affecting their survival in two oxidation ditch process municipal wastewater treatment plants located in different regions. Ecotoxicol Environ Saf 154, 162-170.
Wang, Y., Li, L., Han, Y., Liu, J. and Yang, K. (2018d) Intestinal bacteria in bioaerosols and factors affecting their survival in two oxidation ditch process municipal wastewater treatment plants located in different regions. Ecotoxicology and Environmental Safety 154, 162-170.
Wang, Y., Li, L., Xue, S., Han, Y. and Yang, K. (2019) Characteristics and formation mechanism of intestinal bacteria particles emitted from aerated wastewater treatment tanks. Water Research 163, 114862.
Wang, Z.S., Duan, X.L., Ping, L., Jing, N., Nan, H. and Zhang, J.L. (2009) Human exposure factors of Chinese people in environmental health risk assessment. Research of Environmental Sciences 22(10), 1164-1170.
Wei, M., Li, M., Xu, C., Xu, P. and Liu, H. (2020) Pollution characteristics of bioaerosols in PM2.5 during the winter heating season in a coastal city of northern China. Environmental Science and Pollution Research (9).
Xu, G., Han, Y., Li, L. and Liu, J. (2018) Characterization and source analysis of indoor/outdoor culturable airborne bacteria in a municipal wastewater treatment plant. Journal of Environmental Sciences 74, 71-78.
Yan, X., Qiu, D., Zheng, S., Yang, J., Sun, H., Wei, Y., Han, J., Sun, J. and Su, X. (2019a) Distribution characteristics and noncarcinogenic risk assessment of culturable airborne bacteria and fungi during winter in Xinxiang, China. Environmental Science and Pollution Research 26(36), 36698-36709.
Yan, X., Qiu, D., Zheng, S., Yang, J., Sun, H., Wei, Y., Han, J., Sun, J. and Su, X. (2019b) Distribution characteristics and noncarcinogenic risk assessment of culturable airborne bacteria and fungi during winter in Xinxiang, China. Environ Sci Pollut Res Int 26(36), 36698-36709.

Yang, K., Li, L., Wang, Y., Xue, S., Han, Y. and Liu, J. (2019) Airborne bacteria in a wastewater treatment plant: Emission characterization, source analysis and health risk assessment. Water Research (149), 596-606.

Yang, L., Miao, Y. and He, B.J. (2014) CFD simulation research on residential indoor air quality. Science of the Total Environment 472(15), 1137-1144.

Zhang, Y., Cao, G., Feng, G., Xue, K., Pedersen, C., Mathisen, H.M., Stenstad, L. and Skogas, J.G. (2020) The impact of air change rate on the air quality of surgical microenvironment in an operating room with mixing ventilation. Journal of Building Engineering 32, 101770.