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ORIGINAL PAPER

Abundance and health risk of bioaerosols in the coastal areas of Qingdao, China

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Abstract Bioaerosols can be spread through coughing, sneezing, respiratory droplets and aerosol particles, and public awareness of the health risks of bioaerosols has increased. Based on bioaerosol culturable microbe concentration data collected from March–December in 2015, 2018 and 2019, the health risks of bioaerosols were assessed by air quality level, month, population, and particle size using an average daily dose rate model. The concentration of culturable microorganisms is related to the air quality index (AQI). Under AQI values ranging from 51–100, the concentration of culturable microorganisms was the highest, while the concentration of culturable microorganisms was the lowest for AQI values ranging from 101–150. The health risk in June and July 2015 was the highest, the change trends in 2018 and 2019 were similar, the health risk was the highest in October, and the health risk of bioaerosols along the inhalation route was 10^3–10^4 times that along the exposure route. The health risk of bioaerosols was generally higher in summer and autumn than in spring and winter over the three-year period. The health risk for different categories of individuals indicated the same trend over the 3-year period, with the health risk for adults exceeding that for children and the health risk for men exceeding that for women. The health risk of bioaerosols was high under particle sizes ranging from 1.10–4.70 μm. The study results could provide data support for the analysis of bioaerosol-related health risks and offer a reference for the prevention and control of urban microbial diseases.

Keywords Bioaerosol · Health risk assessment · Average daily dose rate model · Coastal area · Culturable microbial concentration · Air quality index

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1 Introduction

The term "Bioaerosol" refers to aerosol particles which are attached to living substances such as microorganisms or biomacromolecules (Bowers

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et al., 2011; Zhang et al., 2022a), including bacteria, fungi, viruses, fern spores, pollen and parasitic ovum (Li et al., 2022; Haleem et al. 2012), which have been reported to be transmitted from person to person mainly through droplets and aerosol particles produced during talking, coughing and sneezing, thus drawing increasing public attention to the health risks of bioaerosols (Qi et al., 2018; Rodriguez-Gomez et al., 2020). Microorganisms in the air come from a wide range of sources, including soil, water, animals and plants, as well as human activities, and 80% of microorganisms in the atmosphere can attach to airborne particles, which form an important part of the ecosystem (Fröhlich-Nowoisky et al., 2016; Górny., 2020).

Prolonged exposure to high levels of bioaerosol can lead to a wide range of respiratory infections and systemic inflammations(Faridi et al., 2017). In particular, living microorganisms can deposit in the deeper respiratory tract and reproduce under suitable conditions. Therefore, there is no fixed threshold for the risk of bioaerosol exposure, and health risks to humans are also difficult to define. Health risk assessment is a process of assessing the impact of hazardous substances on a specific population or ecosystem under specific conditions, which focuses on the relationship between exposure to hazards and health risks in a specific population. There are four main methods of assessment all over the world (Zamfir et al., 2019; Hsieh et al., 2014; Liang et al., 2014; Madhwal et al., 2020): animal experiments based on common dose–response models, deposition kinetics based on different particle sizes of microorganisms, metrological responses based on big data comparisons, and estimation of average daily exposure dose rate models based on microbial concentrations.

The most commonly used model in bioaerosol-related health risk assessment is the average daily dose (ADD) model proposed by the US Environmental Protection Agency in (1999), which is still adopted by most researchers today (Wang et al., 2018; Yang et al., 2019a; Zhang et al., 2022a, 2022b). In this study, we collected bioaerosol culturable microbial samples in Qingdao from March–December in 2015, 2018 and 2019 and assessed the health risk of bioaerosols using the ADD model based on culturable microbial concentrations obtained through culture method analysis. The study results can provide data support for the analysis of bioaerosol-related health risks, provide a reference for the prevention and control of urban microbial diseases, and have practical significance for the selection of safety protection measures for respiratory diseases in different populations.

2 Materials and methods

2.1 Sampling locations

The sampling site is located at the top of the teaching building of the Laoshan Campus of Ocean University of China (36°09′N, 120°29′E), as shown in Fig. 1. The sampling site is located at a vertical height of approximately 9 m from the ground, at an altitude of approximately 90 m, and at a straight-line distance of approximately 7 km from the coastline, with no obvious pollution sources such as nearby industrial areas.

2.2 Sampling methods

An FA-1 Andersen six-stage air microbiological sampler was used. The sampler provides six stages with 400 holes per stage, with each stage exhibiting a progressively smaller hole size and different deposition locations for aerosol particles upon entry in the sampler (Table 1) (Andersen, 1958). A total of 120 sets of bioaerosol samples were collected from March to December in 2015, 2018 and 2019, with parallel repeats per sampling. Sampling was performed every Thursday, but sampling occurred earlier or later as appropriate in the case of specific weather conditions affecting the sampling process, such as rain or snow. The sampling time was 8:00–9:00 am, and the sampling flow rate was 28.30 L min^{-1}.

Plate culture and colony counting were used to detect microbial concentration. Before each sampling, the 90-mm Petri dish was sterilized at 121 °C for 30 min, and then, 30 mL of culture medium liquid was added. Sabouraud medium was used for fungi, and nutrient agar medium was used for bacteria. The sampling and culture conditions are summarized in Table 2. After culture, bacterial and fungal communities were observed, counted (in terms of colony forming units (CFU)), purified, and identified.

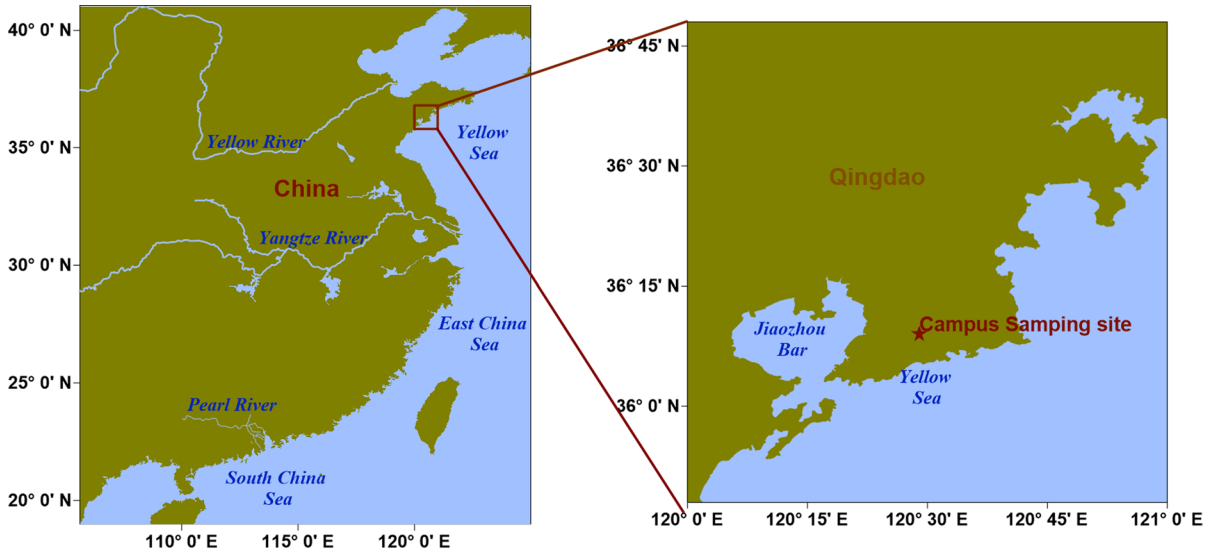

Fig. 1 Sampling location

Table 1 Distribution of the particle size and deposition location of the Andersen sampler (Andersen, 1958)

Number of levels	Aperture		Catch range	Location of deposition
	mm	m s^{-1}	μm	
F1	1.18	1.02	>7.00	Nose and mouth
F2	0.91	1.53	4.70–7.00	Throat
F3	0.71	3.65	3.30–4.70	Trachea and primary bronchus
F4	0.53	4.85	2.10–3.30	Secondary bronchus
F5	0.34	12.62	1.10–2.10	Terminal bronchus
F6	0.25	23.14	0.65–1.10	Alveoli

2.3 Data analysis

After a certain number of microbial particles was captured at each stage of the Andersen sampler, the probability of microbial particles impacting the same location through the same culture increases, resulting in an overlap of colonies in the collected culturable microbial samples, which affects the counting accuracy and must be corrected (Macher et al. 1989), as expressed in Eq. (1).

$$P_r = N \cdot \left(\frac{1}{N} + \frac{1}{N-1} + \frac{1}{N-2} + \cdots + \frac{1}{N-r-1}\right) \quad (1)$$

In the above equation:

r: number of colonies per stage before correction;

P_r*:* number of colonies per stage after correction; and.

N: number of sampler wells per stage.

The concentration of culturable microorganisms is expressed in CFU·m^{-3} and can be calculated with Eq. (2).

$$c_i = \frac{\mathrm{Pr} \cdot 1000}{Q \cdot t} \quad (2)$$

Table 2 Microbial culture conditions

Strain	Sampling time	Training base	Cultivation conditions
Fungi	4 min	Glucose: 4.00 g L^{-1}, peptone: 10.00 g L^{-1}, agar: 20.00 g L^{-1}, streptomycin sulfate: 40.00 μg mL^{-1}	Constant-temperature incubation (25℃) for 72–120 h
Bacteria	8 min	Beef paste: 5.00 g L^{-1}, peptone: 10.00 g L^{-1}, sodium chloride: 5.00 g L^{-1}, agar: 20.00 g L^{-1}, actinomycin: 100.00 μg mL^{-1}	Constant-temperature incubation (37℃) for 48–72 h

$$P_i = \frac{c_i}{c} \cdot 100\% \quad (3)$$

In the above equation:

P_r: corrected colony count;

Q: sampler flow rate (L min^{-1});

c_i: microbial concentration per stage (CFU · m^{-3});

c: total microbial concentration (CFU·m^{-3});

t: sampling time (min); and

P_i: percentage microbial concentration per stage.

Bioaerosols pose a noncarcinogenic health risk to humans (Yang et al., 2022). The Andersen six-stage microbial sampler adopted in this paper simulates the different parts of the human respiratory tract for each particle size through different pore size tiers. The noncarcinogenic health risks of bioaerosols were assessed using the ADD model proposed by the US Environmental Protection Agency in 1999. In health risk assessment, the average daily human exposure dose via the respiratory tract was calculated with Eq. (4), and the average daily human exposure dose via dermal contact was calculated with Eq. (5).

$$\mathrm{ADD_{inhalation}} = \frac{\mathrm{C \cdot IR \cdot EF \cdot ED_{inhalation}}}{\mathrm{BW \cdot AT}} \quad (4)$$

$$\mathrm{ADD_{skin}} = \frac{\mathrm{C \cdot SA \cdot ABS \cdot AF \cdot EF \cdot ED_{skin}}}{\mathrm{BW \cdot AT}} \quad (5)$$

In the above equation:

ADD: average daily exposure dose (CFU ·(kg · $d)^{-1}$);

C: microbial concentration (CFU · m^{-3});

ED: respiratory and dermal exposure years (yr);

IR: respiratory frequency ($m^3 \cdot d^{-1}$);

EF: exposure frequency ($d \cdot yr^{-1}$);

SA: contact skin surface area (m^2);

ABS: skin absorption factor ($m \cdot h^{-1}$);

AF: skin adhesion factor;

BW: body weight (kg); and.

AT: life expectancy (d).

Relevant model parameters were obtained from the Manual of Exposure Parameters for the Chinese Population (Shandong city) published by the Ministry of Environmental Protection of the People's Republic of China (PRC) (Ministry of Environmental Protection of the PRC., 2013), and specific values are listed in Table 3.

The hazard quotient (HQ) was calculated with Eq. (6), and the hazard index (HI) was obtained with Eq. (7).

$$\mathrm{HQ} = \mathrm{ADDRfD^{-1}} \quad (6)$$

$$\mathrm{HI} = \sum \mathrm{HQ_i} \quad (7)$$

HQ denotes the hazard quotient for the different particle sizes along the inhalation or exposure route, and HI denotes the sum of the HQ values for the different particle sizes along the different exposure routes. For HI < 1, the noncarcinogenic risk of microorganisms can be ignored, and for HI > 1, the noncarcinogenic risk of microorganisms cannot be ignored. A value of 500 CFU·m^{-3} was selected as the reference dose (RfD) (CFU·$(kg \cdot d)^{-1}$), which indicates the

Table 3 Health risk assessment parameters (2016a, 2016b; Ministry of Environmental Protection of the PRC., 2013)

		IR $m^3 \cdot d^{-1}$	EF $min \cdot d^{-1}$	BW kg	SA m^2	ABS $m \cdot h^{-1}$	AF	ED year	AT year
Children (0–5)	Average	6.59	59.10	13.09	0.58	0.01	0.20	6.00	12.00
	Boys	6.94	63.67	13.48	0.59				
	Girls	6.21	54.33	12.64	0.56				
Children (6–17)	Average	13.10	67.50	45.58	1.41				
	Boys	14.93	71.25	49.45	1.48				
	Girls	11.73	64.50	42.68	1.36				
Adults (18–60)	Average	16.60	196.50	62.50	1.65		0.07	25.00	76.46
	Men	18.30	209.50	67.85	1.70				74.05
	Women	15.00	183.00	57.90	1.55				79.06
Adults (> 60)	Average	16.60	181.50	59.70	1.55				76.46
	Men	18.30	198.50	63.50	1.65				74.05
	Women	15.00	169.50	56.40	1.55				79.06

maximum acceptable daily dose of a given substance, in bioaerosol-related health risk evaluation based on the recommendations of the Bioaerosol Committee of the US Governmental Conference of Industrial Hygiene Experts (Yang et al., 2019b).

2.4 Meteorological data sources

Meteorological data during the sampling period were obtained from the Qingdao Meteorological Bureau (http://sd.cma.gov.cn/gslb/qdsqxj/), and particulate matter(PM) and gaseous pollutant concentration data were obtained from the Qingdao Environmental Protection Bureau (http://mbee.qingdao.gov.cn/n28356059/index.html), with specific values provided in Table S1.

3 Results and analysis

3.1 Distribution of the culturable microbial concentration

3.1.1 Distribution of the culturable microbial concentration by month

The concentration of culturable fungi in bioaerosols was much higher than that of culturable bacteria. The total culturable microbial concentration ranged from 355.49–1960.76 CFU m^{-3}, with 83% and 17% of culturable fungi and bacteria, respectively (Fig. 2). The temperature and humidity in Qingdao are ideal for the survival and reproduction of airborne fungi, and the high vegetation cover largely explains the high concentration of airborne fungi because leaf primordia can be used as a natural medium for airborne fungal growth. (Yang et al., 2019b). The highest concentrations of culturable microorganisms in bioaerosols were found during the summer and autumn seasons. The microbial concentration changes in 2018 and 2019 started with a gradual increase in March, and a maximum was reached in September and October, after which the microbial concentration rapidly decreased (Fig. 2). The concentration of culturable microorganisms in bioaerosols during the 2015 sampling period was high in June and July and was notably affected by rain. Postrainy weather conditions were observed on 26 June and 20 July 2015, and related studies have found that intermittent rainfall can accelerate the release of fungal spores, thereby increasing airborne fungal concentrations (Gottwald et al., 1997). Kang et al. considered that the precipitation process in summer could provide a good attachment and humidity environment for the growth of aerosols, thus leading to the increase in aerosol concentration (Kang et al., 2015). Other studies have also demonstrated that the level of culturable microorganisms increases after rainfall (García-Aljaro et al., 2017).

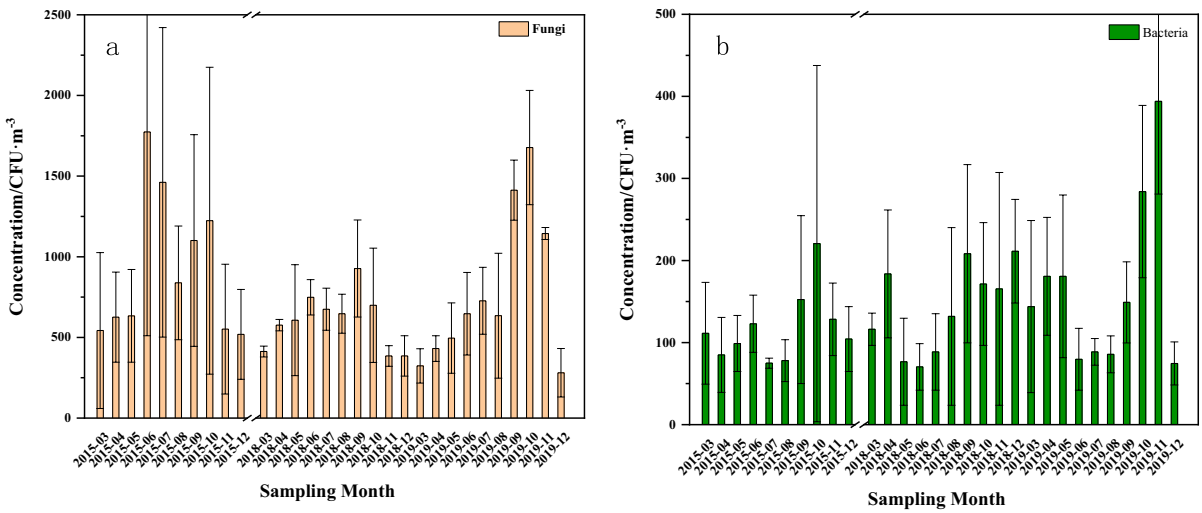

Fig. 2 Distribution of the average monthly concentration of culturable microorganisms **a** fungi; **b** bacteria

The release, dispersal and transport of microorganisms in bioaerosols are influenced by numerous internal and external factors. Temperature, relative humidity and wind speed, as well as gaseous pollutants such as SO_2 and O_3, are important environmental variables for microbial survival (Fan et al., 2019), and atmospheric microorganisms mainly originate from saprophytic environments such as vegetation and parasitic environments such as human and animal emissions. The average temperature in June 2015, September 2019 and October 2019 was 20 °C, 21 °C and 18 °C, respectively, and the average relative humidity was 75.00%, 67.00% and 63.00%, respectively, all very suitable for microbial growth, so the microbial concentration was high. Moreover, the increase in both leaf and straw burning levels in autumn could directly lead to an increase in the concentration of culturable microorganisms in bioaerosols. The temperature in December 2019 reached only 4 °C, and at low temperatures, the growth and reproduction of certain microorganisms could occur at a standstill, while the cells of some thermophilic microorganisms could occur in a frozen state. (They could even be killed.) Hence, the microbial concentrations were the lowest.

3.1.2 Distribution of the culturable microbial concentration under the different air qualities

The concentration of culturable microorganisms in bioaerosols is related to the air quality, which can be classified into three categories (Yousefian et al., 2020), namely good (< 50), moderate (51–100) and unhealthy for sensitive groups (UFSGs) (101–150). We found that the concentration of particulate matter increased with increasing pollution level, but the concentration of culturable microorganisms exhibited a trend of first increasing and then decreasing (Fig. 3), while researchers have obtained similar trends in the concentration of culturable microorganisms under different air qualities in Xi'an (Xie et al., 2018). The maximum concentration of culturable microorganisms occurred at AQI values ranging from 51–100, when the air contained some suspended particulate matter and the adsorbed SO_2 and NO was partially converted into sulfate and nitrate needed for microbial growth and reproduction, contributing to the increase in the culturable microbial concentration (Haddrell et al. 2017). The concentration of air pollutants such as PM will increase with the decline of air quality, while toxic and harmful substances are more likely to adhere to particles and reach a certain threshold (Gao et al., 2015; Sun et al., 2006), resulting in the death and concentration reduction of culturable microorganisms.

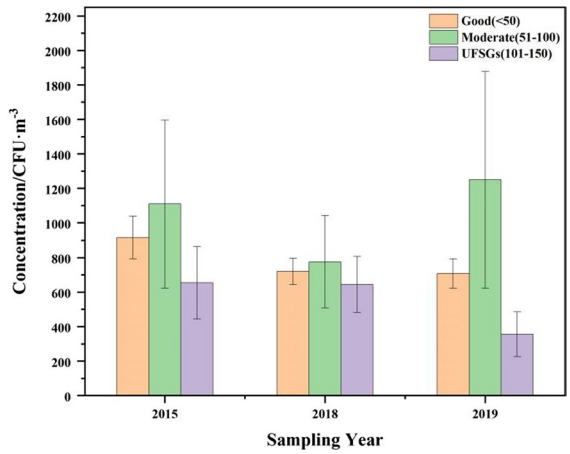

Fig. 3 Variations in the concentration of culturable microbes under the different air quality levels

Interestingly, we conducted correlation analysis according to different AQI value classifications and concentrations of culturable microorganisms, and the results showed that when the AQI value was higher than 60, the concentration of culturable microorganisms was negatively correlated with the AQI value, while the AQI value was lower than 70, the AQI value was significantly positively correlated with the concentration of culturable microorganisms. The specific correlation parameters are shown in Table S2 and Table S3. This conclusion was consistent with the previous conclusion that the concentration of culturable microorganisms first increases and then decreases with the increase of AQI value.

3.2 Health risk assessment

3.2.1 Monthly variation in the health risk

The total hazard index value HI_{total} was less than 1 in all sampling months (Fig. 4), and the noncarcinogenic risk of bioaerosols in Qingdao could be neglected. The highest average health risk was attained in 2015 with an HI_{total} value of 8.74×10^{-2}. The second highest average health risk was observed in 2019 with an HI_{total} value of 6.35×10^{-2}, while the lowest average

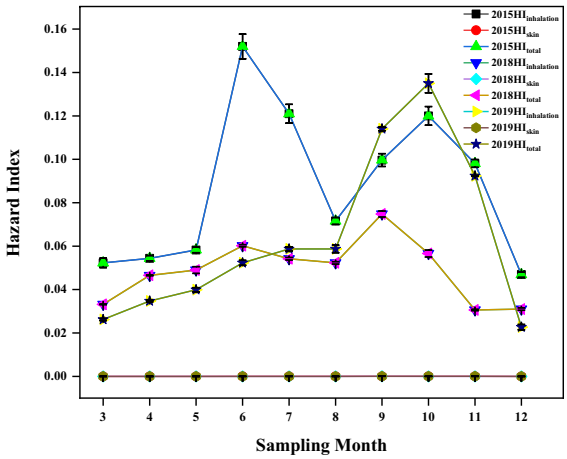

Fig. 4 Bioaerosol health risk assessment from March to December in 2015, 2018 and 2019

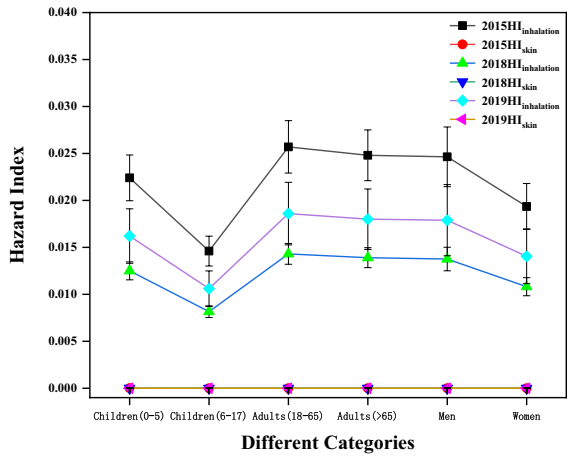

Fig. 5 Average bioaerosol-related health risk index of the different populations in 2015, 2018 and 2019

health risk was attained in 2018 with an HI_{total} value of 4.88×10^{-2}. The highest health risk occurred in June 2015, with an HI_{total} value of 1.52×10^{-2}, and the lowest health risk occurred in December, with an HI_{total} value of 4.69×10^{-2}. The highest health risk was determined in September 2018, with an HI_{total} value of 7.48×10^{-2}, and the lowest health risk was observed in November, with an HI_{total} value of 3.06×10^{-2}. The highest health risk occurred in October 2019, with an HI_{total} value of 1.35×10^{-1}, and the lowest health risk was observed in December, with an HI_{total} value of 2.27×10^{-2}. The change in the health risk exhibited the same overall trend throughout the natural year, consistent with the change in the monthly mean concentration of culturable microorganisms, except for June and July 2015 when the total HI value was high. In summary, the health risk of bioaerosols was higher in summer and autumn than in spring and winter. In 2015, the monthly mean health risk index along the inhalation route $HI_{inhalation}$ was 8.74×10^{-2}, and that along the exposure route HI_{skin} was 1.05×10^{-5}. In 2018, $HI_{inhalation}$ reached 4.88×10^{-2}, and HI_{skin} reached 5.86×10^{-6}. Moreover, in 2019, $HI_{inhalation}$ and HI_{skin} were 6.35×10^{-2} and 7.62×10^{-6}, respectively. The health risk resulting from the inhalation route was 10^3–10^4 times higher than that resulting from the exposure route, and the overall health risk resulting from the inhalation route was much higher than that resulting from the exposure route. Hence, the overall health risk of bioaerosols mainly depended on the inhalation route.

3.2.2 Health risk assessment of the different populations

Bioaerosols pose varying health risks for different types of populations (Fig. 5). The same trend was observed over the 3-year period, with adults attaining higher health risks than children, and men suffering higher health risks than women. The correlation between the health risk value and the proportion of confirmed cases in the different population groups was determined: The high health risk for adults aged 18–60 years mainly occurred because this age group works or goes to school often and experiences more complex contacts, resulting in a higher health risk than that for minors and elderly individuals; the higher health risk for males occurred because males are more likely to be exposed to infectious agents as a result of their work and business activities outside of the home, resulting in high health risks associated with bioaerosols for the male population (Chakravarty et al., 2020; Peel et al., 2011). Overall, the total health risk (HI) of bioaerosols for the different populations in all three years was less than 1. However, considering that bioaerosols in PM could still pose a noncarcinogenic risk, the different categories of individuals in Qingdao should be aware of the health risks associated with PM. Most current studies on the human aerosol exposure risk have been conducted via the simulation technique (Madhwal et al., 2020; Reitsma et al., 2021; Yang et al., 2019a, 2019b). In the present study, we confirmed the relationship between aerosol

pollution and human diseases through simulations, while the role of aerosols in disease transmission can be determined in future via animal experiments and other means.

3.2.3 Health risk assessment by particle size

The health risks associated with bioaerosols of different particle sizes vary (Fig. 6), and there are differences in the health effects of bioaerosols of each particle size on different parts of the human respiratory system. The pathogenicity of airborne microorganisms is related not only to their species but also to their particle size (Jiang et al., 2022). The peak particle size of the determined bacterial particle size distributions varied in the different sampling months, and factors such as the sampling time, location and weather could affect the particle size distribution of bacteria (Wu et al., 2021). The deposition of bioaerosol particles of different sizes in different parts of the body results in different health risk levels. Throughout the three-year period, in terms of the human health effects of bioaerosols with different particle sizes, bioaerosols with particle sizes ranging from 1.10–4.70 μm posed a high health risk to humans. Microorganisms within the 1.1–2.1 μm particle size range could become attached to particles, and particles could protect microorganisms from external conditions such as ultraviolet light (Raisi et al., 2013). Fungal spores can mostly be found suspended in air and are much more abundant than bacteria, while bacteria, with a smaller particle size, are mainly attached to PM (Dybwad et al., 2014; Zhang et al., 2022b). Particles with particle sizes ranging from 1.10–4.70 μm are mainly deposited in the trachea and bronchi of the respiratory tract (Li et al., 2015), and bioaerosols therefore affect the trachea and bronchi more notably than other parts of the respiratory tract, and the bronchi are very vulnerable to pathogens, especially in July 2015, when the health risk reached a maximum value of 5.43×10^{-2}.

4 Conclusions

In this study, a total of 120 groups of culturable microbial samples were collected in Qingdao over 3 years, and the health risks of bioaerosols under different air quality levels, months, populations and particle sizes were analyzed with the ADD rate model proposed by the US Environmental Protection Agency, and the following conclusions could be obtained:

(1) In 2015, the highest concentration of culturable microorganisms was found in June and July, followed by the second highest health risk in October, and the same trend was observed between 2018 and 2019, with the highest concentration of culturable microorganisms detected in October. The culturable microorganism concentration is related to the AQI value, with the highest concentration of culturable microorganisms at an AQI ranging from 51–100.

(2) The total health risk HI_{total} values for bioaerosols from March–December in 2015, 2018 and 2019 were all less than 1. The health risks of bioaerosols in the different months conformed with the monthly changes in the microbial concentration, with the highest health risks in June and July 2015. In 2018 and 2019, the highest health risk HI_{total} values were observed in October, and the

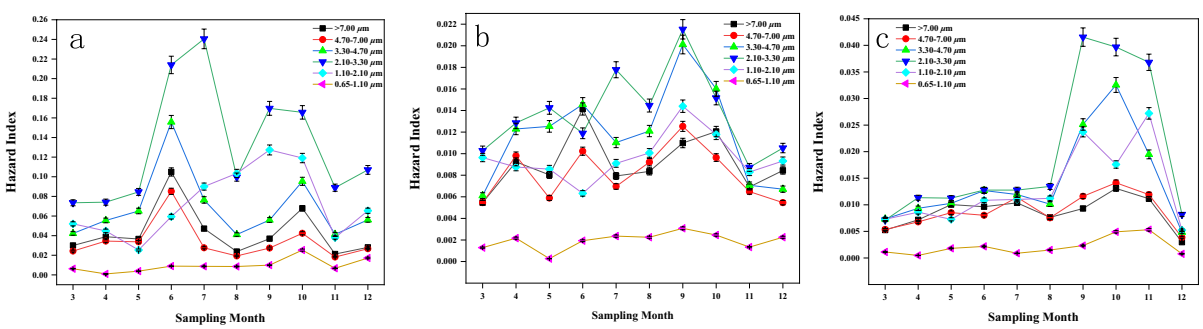

Fig. 6 Health risk index for bioaerosols with different particle sizes in the different years: **a** 2015, **b** 2018, and **c** 2019

health risks of bioaerosols resulting from the inhalation route were 10^3–10^4 times higher than those resulting from the exposure route.

(3) The health risk of bioaerosols in summer and autumn was higher than that in spring and winter. Adults attained higher health risks than children, and men suffered higher health risks than women. The impact of bioaerosols on the trachea and bronchi was greater than that on other parts of the respiratory tract, and the bronchi were highly vulnerable to pathogens.

Author contributions All authors contributed to the study conception and design. Material preparation and sampling collection were performed by Ting Zhang, Chen Han and Yao Wang. Meteorological data collection was performed by Shaohua Sun and Yongzhong Song. The first draft of the manuscript was written by Lingchong Yan. Dahai Zhang, Jianhua Qi and Xianguo Li revised the original manuscript. All authors commented on previous versions of the manuscript and approved the final manuscript.

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Data availability The data that support the findings of this study are available from the corresponding author upon reasonable request.

Declarations

Conflict of interest The authors declare no competing interests.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee.

Consent to participate Informed consent was obtained from all individual participants included in the study.

Consent for publication The manuscript is approved by all authors for publication.

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