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Distribution characteristics and noncarcinogenic risk assessment of culturable airborne bacteria and fungi during winter in Xinxiang, China

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Abstract

Bioaerosols are an important component of particulate matter in the atmosphere and are harmful to human health. In this study, the concentration, size distribution, and factors influencing culturable airborne bacteria and fungi in the atmosphere were investigated using a six-stage impactor device in the city of Xinxiang, China, during the winter season. The results revealed that the concentration of culturable airborne bacteria and fungi varied significantly during the sampling period: 4595 ± 3410 and 6358 \pm 5032 CFU/m³, respectively. The particle sizes of the bioaerosols were mainly within stage V (1.1–2.1 µm), and fine particulate matter accounted for $45.9\% \pm 18.9\%$ of airborne bacteria and $52.0\% \pm 18.5\%$ of airborne fungi, respectively. With the deterioration of air quality, the concentration of airborne fungi gradually increased, and that of airborne bacteria increased when the air quality index was lower than 200 and decreased when it was higher than 200. With respect to the diurnal variation pattern of bioaerosol concentration, the highest and lowest concentrations were registered at night and noon, respectively, probably because of changes in ultraviolet radiation intensity. Bioaerosol concentration positively correlated with humidity, concentration of $PM_{2.5}$, PM_{10} , SO₂, and NO₂ and negatively correlated with O₃ concentration. The risk of exposure of humans to the airborne bacteria was primarily associated with the respiratory inhalation pathway, and the risk of skin exposure was negligible. These results should improve our understanding of the threat of bioaerosols to public health.

Keywords Culturable bioaerosols · Concentration distribution · Size distribution · Influencing factors · Noncarcinogenic risk assessment . Central China

Introduction

In the past decades, due to rapid urbanization and industrial and economic development, massive consumption of energy and a rapid increase in traffic have caused a significant increase in the emission of air pollutants as well as deterioration of air quality. An increase in suspended particulate matter (PM) and haze pollution in the atmosphere has been reported worldwide, including in China, and can significantly adversely affect

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 \boxtimes Xu Yan yanxu@htu.cn ecosystems and human health (Tao et al. 2014; Zhuang et al. 2014; Latif et al. 2018).

Bioaero[sols a](#page-10-0)re a group of organic aerosols with a particle size in the range of nm10–100 mm. Specifically, they include airborne particles and large molecules that either carry living organisms or are released from living organisms (Ariyap and Amyot 2004). The biological organisms or dispersal units (dead or alive[, isola](#page-10-0)ted or aggregated), including bacteria, fungi, protozoa, algae, spores, pollen, lichen, archaea, viruses, and their solid fragments or excretions, including detritus, microbial fragments, plant debris/leaf litter, animal tissue and excrements, brochosomes, all comprise the bioaerosol (Després et al., 2012). They are often dispersed attached to other biological or [non-b](#page-10-0)iological particles, such assoil, dust, skin flakes, saliva, or water droplets (Zhai et al. 2018). Bioaerosols are an important component of PM and are [harm](#page-11-0)ful to human health. They can cause or aggravate skin allergies, respiratory tract infections, asthma, cardiovascular diseases, and chronic lung diseases

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through damaged skin, the mucosa, digestive tract, and respiratory tract (Douwes et al. 2003; Yamamoto et al. 2012; Walser et al. 2015; Frohlich-Now[oisky](#page-10-0) et al. 2016). Via m[icrobi](#page-11-0)ological and [chemi](#page-11-0)cal processes, bioaerosols [affect](#page-10-0) atmospheric chemistry and vice versa. This adverse impact may be harmful and deteriorative (Burrows et al. 2009). Additionally, bioaerosols are closely related to ecologic[al proc](#page-10-0)esses and play an important role in the natural cycling of matter (Fang et al. 2016). Research on bioaerosols in an ambient atmosphere has gr[aduall](#page-10-0)y become a hot spot in the field of environmental studies.

Most of China has frequently been affected by severe haze or smog days in recent years. Especially in winter, an anomalous steady atmosphere caused by sudden stratospheric warming has often occurred and is conducive to PM formation and cumulation and to haze pollution (Zhang et al. 2016; Shi et al. 2018). At a high concentration of atmospheric [chemi](#page-11-0)cals on ha[ze day](#page-11-0)s, potential synergistic effects between biological and chemical pollutants may further intensify the hazards to human health (WHO 2005). Therefore, it is essential to investigate the bioaerosol char[acteri](#page-11-0)stics, influencing factors, and risks of resulting exposure for public health in a period of frequent haze days.

Some researchers have focused on studying bioaerosols from the atmospheric environment in recent years (Xu et al. 2011; Xie et al. 2018b; Bragoszewska and Pastuszka 2018). [None](#page-11-0)theless, bi[oaeroso](#page-11-0)ls have significant regional ch[aracter](#page-10-0)istics owing to the broad diversity and tremendous variability of microbial composition. Bioaerosols have different sources of origin and are influenced by seasonal factors, local climatic differences, local human activities, and local wind currents (Shaffer and Lighthart 1997). The concentrations and distribution characteristics [of bio](#page-11-0)aerosols in central China have seldom been investigated. A typical medium-sized city, Xinxiang, was chosen as a model for sampling of culturable bioaerosols in this study. Xinxiang is located in central China and is a Beijing-Tianjin-Hebei Air Pollution Transmission Channel City $(2 + 26 \text{ City})$ (Ministry of Ecology and Environment of the People's Republic of China 2017), which has been affected by haze frequently in recent [years.](#page-11-0) During the winters of 2016–2018, $PM_{2.5}$ was considered the chief pollutant in the atmosphere of Xinxiang (XEPB 2016–2018).

The objective of this study was to provide a b[asis fo](#page-11-0)r [study](#page-11-0)ing the environmental and health effects of bioaerosols on haze days of winter on public health. It is also expected to serve as a reference for studies on the atmospheric environment of the Beijing-Tianjin-Hebei region and the surrounding area.

Materials and methods

Sampling sites

Xinxiang (35.3 °N, 113.9 °E, 72 m above sea level) is located in the northern Henan Province, has a population of over 6.10 million, and occupies an area of 8249 km² (http://www. xinxiang.gov.cn/sitesources/xxsrmzf/page_pc/index.html). This city is between the Yellow River in the south and Taihang Mountain in the north. It has a temperate continental monsoon climate. In winter, the predominant wind direction was northeast (HMS 2018). The topography of Xinxiang is mainly plain; this characteristic applies to 76.6[% of the total](http://www.xinxiang.gov.cn/sitesources/xxsrmzf/page_pc/index.html) [area \(Zhou](http://www.xinxiang.gov.cn/sitesources/xxsrmzf/page_pc/index.html) 1988).

The field sampling of ambient bioaerosols was carried out on the top roof of the School of Environment Building located at Henan Normal University. The distance of the site from the ground was about 30 [m. T](#page-10-0)he site is surrounded by trees and residential and school buildings. There are no large industrial pollution sources near the site.

Measurement of culturable bioaerosols

Sampling time and frequency

The distribution characteristics of the bioaerosols at different air quantities and diurnal periods were investigated. A sixstage impactor (Tisch Environmental, Inc., USA) was installed at a height of about 1.5 m above the building roof surface, and bioaerosols of different particle size ranges were collected. The size ranges of the particles captured are presented in Table 1. Bioaerosol with size smaller than 2.1 μm was defined as fine particles and that with size larger than 2.1 μm was defined as coarse particles because this impactor does not have a 2.5-μm cutoff point (Wu et al. 2017).

Samples were collected from the period of November 2017 to March 2018, and the main pollutants during this period was PM_{2.5} (78 days), PM₁₀ (43 days), NO₂ (22 days), and O₃ (5 days) (Ministry of Ecology and Environment of the People's Republic of China 2018). To detect di[urnal v](#page-11-0)ariation, samples were collected at 9:00, 14:00, and 19:00 on each sampling day. Three consecutive replicates were collected for each sampling time point. The sampling flow rate was 28.3 L/min, and sampling time was 4 min. Each sampling device was sterilized by 75% ethanol b[efore e](#page-11-0)very sampling.

Table 1 Range of particles captured

Stage	Aerodynamic diameter	Aperture
\mathbf{I}	$> 7.0 \mu m$	1.18 mm
\mathbf{I}	$4.7 - 7.0 \mu m$	0.91 mm
Ш	$3.3 - 4.7 \mu m$	0.71 mm
IV	$2.1 - 3.3 \mu m$	0.53 mm
V	$1.1 - 2.1 \mu m$	0.34 mm
VI	$0.65 - 1.1 \text{ µm}$	0.25 mm

Culture method for bacterial and fungal samples

Bacterial samples were cultured on Beef-Peptone mediums (3 g beef extract, 10 g peptone, 5 g NaCl, 16 g agar, 1 L distilled H₂O, pH 7.2–7.6) at 37 °C and incubated for 48 h. Fungal samples were cultured on Rose Bengal mediums (10 g glucose, 5 g peptone, 1 g KDP (potassium dihydrogen phosphate), 0.5 g MgSO₄ \cdot 7H₂O, 15 g agar, 0.03 g rose bengal, 1 L distilled H₂O) at 25° C and incubated for 72 h (Li et al. 2011).

Counting method

Colony-forming units (CFUs) were counted using p[ositive](#page-11-0)hole correction (Andersen, 1958). The concentration of bioaerosol was calculated via the following formula:

$$
C = \frac{P_r \times 1000}{t \times Q} \tag{1}
$$

where c is the concentration of the bioaerosol (CFU/m³), P_r is the revised colony number at each stage, t is the sampling time (min), and Q is the sampling flow rate (L/min).

Noncarcinogenic risk assessment

A noncarcinogenic risk assessment model was used to evaluate the risk of exposure to airborne bacteria for public health. According to a study by Li et al. (2013), the exposure and health risk assessment can be evaluated based on the models developed by US EPA (US Environmental Protection Agency), which included two main [expos](#page-11-0)ure pathways: inhalation and dermal contact.

Dose contacted through inhalation of bioaerosols (ADD_{inh}) can be calculated as

$$
ADD_{inh}[CFU/(kg \cdot d)] = \frac{c \times IR \times EF \times ET}{BW \times AT}
$$
 (2)

Dose absorbed through dermal contact with bioaerosols (ADDdermal) can be expressed as

$$
ADD_{\text{demal}}[CFU/kg \cdot d] = \frac{c \times SA \times SL \times ABS \times EF \times ET}{BW \times AT}
$$
 (3)

where IR is the inhalation rate, EF is the exposure frequency, ET is the exposure time, SA denotes the exposure of skin surface area, SL is the skin adherence factor, ABS represents the dermal absorption factor, BW is the average body weight, and AT denotes the averaging time to define noncarcinogenic exposure.

The risk for noncarcinogenic pollutants was expressed as the hazard quotient (HQ) and is given by the following equation:

$$
HQ = \frac{ADD}{RfD}
$$
 (4)

$$
HI = \sum HQ_i \tag{5}
$$

where hazard index (HI) represents the sum of the hazard quotients for each pathway and for each target pollutant, and RfD is the daily dose compared with the reference dose for chronic exposure. When $HO \le 1$ or $HI \le 1$, noncarcinogenic effects are not of concern, whereas when $HQ > 1$ or $HI > 1$, noncarcinogenic effects are cause for concern.

Because of the differences among geographic conditions, the parameter values of this calculation are different for each country. The parameter values that are suitable for Chinese people are presented in Table 2.

Data analysis

The concentration of particulates such as $PM_{2.5}$ and PM_{10} and meteorological parameters including temperature, relative humidity, and SO_2 , NO_2 , and O_3 concentrations were retrieved from the Air Quality Forecast and Release System in Henan (http://1.192.88.18:8088/TodayMonitor). The basic information during the period of sampling days is listed in Table 3. SPSS 24.0 software was used to calculate descriptive s[tatistical parameters and perform such tests as](http://1.192.88.18:8088/TodayMonitor) one-way analysis of variance (ANOVA), the group t test, and nonparametric Spearman's correlation analysis. A P value of less than 0.05 indica[te](#page-3-0)d a statistically significant difference at a confidence level of 95%. The correlations between culturable bioaerosols and the influencing factors were then analyzed.

Results and discussion

Concentration distribution of bioaerosols during the sampling period

The concentration distribution of culturable bioaerosols is presented in Fig. 1, as determined during the sampling period from November 2017 to March 2018. The concentrations of culturable airborne bacteria and fungi varied significantly during the sampling period with the mean value of 4595 ± 3410 CFU/m³ and [6](#page-3-0)358 \pm 5032 CFU/m³, respectively. The culturable airborne bacteria and fungi reached the highest concentrations on January 14, 2018 (12853 \pm 4520 CFU/m³) and December 27, 2017 (16534 \pm 4622 CFU/m³), respectively. The highest concentrations of fine particles of airborne bacteria (7836 ± 3530 CFU/m³) and fungi (10433 ± 2476 CFU/m³) were also detected during the same sampling period at relatively high PM_{2.5} concentrations of 131 ± 13 and 138 ± 21 µg/ m³, respectively.

Because particles in the atmosphere act as the vector for microorganisms, an increase in the PM concentration Table 2 The appropriate

people

generally leads to an increase in the concentration of bioaerosols. PM at high concentrations may contain some harmful substances such as crustal elements, pollution elements, and inorganic ions that have a negative impact on microorganisms (Sun et al. 2006; Sun et al. 2013; Gao et al. 2015b). This phenomenon probably resulted in a low concentration of airborne bacteria (2563 \pm 268 CFU/m³) on December 3, 2017, at the highest $PM_{2.5}$ concentration of 209 ± 16 g/m³. Rain and sno[w hav](#page-11-0)e a scourin[g effect](#page-10-0) on the particles in the air (Almaguer et al. 2014; Lee et al. 2016; Li et al. 2017; Xie et al. 2018b). Thus, the lowest concentrations of airborne bacteria and fungi were detected on December 17, 2017, and January 8, 2018, respectively, with snowfall before the sampling day.

Concentration and size distribution of bioaerosols at different air quality levels

The air quality index (AQI) is usually used to describe the air quality situation. According to Technical Regulation on Ambient Air Quality Index (on trial), the status of air quality was categorized into six classes: excellent (0–50), good (51– 100), slight pollution (101–150), moderate pollution (151– 200), heavy pollution (201–300), and serious pollution (> 300) (HJ 633- 2012 2012). Figures 2, 3, and 4 present the concentration and size distributions for culturable airborne bacteria and fungi at different AQIs. Because there was no

Table 3 Information of air quality during sampling days

	Number	Minimum	Maximum	Mean
Temperature $(^{\circ}C)$	34	-8.1	23.9	5.6
Relative humidity $(\%)$	34	12.0	89.0	43.6
$PM_{2.5} (\mu g/m^3)$	34	11	230	90
$PM_{10} (\mu g/m^3)$	34	29	315	144
$SO_2 (\mu g/m^3)$	34	6	56	30
$NO_2 (\mu g/m^3)$	34	17	142	64
$O_3 (\mu g/m^3)$	34	5	160	46
AOI	36	29	280	119

sampling date during serious-pollution weather in the sampling period, the A[QI hig](#page-10-0)her than [20](#page-4-0)[0](#page-5-0) was i[nt](#page-6-0)erpreted as heavy pollution.

The average culturable airborne bacteria concentrations in different AQI classes were in the range of 1237 ± 928 to 10097 ± 6380 CFU/m³. An increase in airborne bacteria concentrations with the increasing AQI was observed when AQI was lower than 200. At AQI higher than 200, the concentration of airborne bacteria decreased to 4875 ± 2745 CFU/m³.

Fig. 1 Concentration distribution of culturable airborne bacteria and fungi and $PM_{2.5}$ concentrations during sampling days (a, culturable airborne bacteria; b, culturable airborne fungi)

Fig. 2 Concentration distribution of culturable airborne bacteria and fungi at different AQIs (a, culturable airborne bacteria; b, culturable airborne fungi). **Correlation is significant at the 0.01 level (twotailed). *Correlation is significant at the 0.05 level (two-tailed)

The concentration of airborne bacteria under excellent weather conditions was significantly lower than that under other weather conditions (AQI > 50; $P < 0.05$). Generally, the concentration of airborne fungi increased with the increasing AQI, and the highest concentration of 12729 ± 6765 CFU/ m³ was observed in heavy-pollution weather. The concentration of airborne fungi in slight-pollution weather was significantly higher than that in good weather $(P < 0.05)$.

Bioaerosols were influenced by many factors in the atmosphere (physical, chemical, and biological factors) (Zhong et al. 2016). The bioaerosol characteristics often varied significantly among regions because the atmospheric conditions differed greatly among different time points and locations (Timo 1997; Wei et al. 2015; Dong et al., 2016; [Xie et a](#page-11-0)l. 2018a). In Xi'an of China, the total airborne microbial concentration was found to increase initially and then to slightly decrease with the increasing AQI. The peak appeared at [the m](#page-11-0)oderate po[llutio](#page-11-0)n level (Xie [et al.](#page-10-0) 2018b). [Wei et](#page-11-0) al. (2016) found bioaerosol concentration to be significantly higher during haze weather than during sunny weather from December 2013 to March 2014 in Beijing. Nonetheless, the community structures o[f airbor](#page-11-0)ne bacteria an[d fung](#page-11-0)i in $PM_{2.5}$ samples did not show significant differences at different AQI levels during the 2014 APEC Summit Periods in Beijing (from October 15, 2014, to November 12, 2014) (Du et al. 2018b).

The size distribution of culturable bioaerosols is presented in Figs. 3 and 4. For airborne bacteria, the concentration distributions of particle sizes at different stages were close within [the exc](#page-10-0)ellent-weather class. During sampling days with AQI greater than 50, the culturable airborne bacteria were found to be mai[nly](#page-5-0) wi[th](#page-6-0)in Stage V (1.1–2.1 μ m) (34.9% \pm 16.2%, $49.4\% \pm 8.7\%, 40.8\% \pm 15.7\%,$ and $34.6\% \pm 16.2\%$ for good weather, slight pollution, moderate pollution, and heavypollution weather, respectively). With the increase in size range, the concentration distribution gradually decreased. A similar distribution trend was observed for culturable airborne fungi. On the sampling days with AQI greater than 50, the highest concentrations were seen at Stage V $(1.1–2.1 \mu m)$ $(49.4\% \pm 20.3\%, 50.0\% \pm 13.2\%, 45.0\% \pm 7.1\%, \text{and}$ $52.7\% \pm 6.4\%$ for good weather, slight pollution, moderate pollution, and heavy-pollution weather, respectively). In the class of excellent weather, the highest concentration, $402 \pm$ 355 CFU/m³, was noted at stage III (25.4% \pm 13.0%).

Generally, our results indicate that $45.9\% \pm 18.9\%$ of culturable airborne bacteria and $52.0\% \pm 18.5\%$ of culturable airborne fungi that were detected during the sampling period had a particle size less than 2.1 μm. This result was probably due to $PM_{2.5}$, which was identified as the chief pollutant during 52% of the sampling period from November 2017 to March 2018 (PM₁₀ accounted for 29%), as described in 2.2.1. the "Sampling time and frequency" subsection. The particles served as existence vectors for the microorganisms in the atmosphere. A similar phenomenon was observed in a study by Wei et al. (2015), which revealed that the biological fraction of $PM_{2,5}$ in 11 major cities of China ranged from 55 to 91% in 2013. Different size distribution characteristics of culturable bioaerosols have also been observed in other regions. From Januar[y 14,](#page-11-0) 2013, to January 22, 2014, detected culturable airborne bacteria were mainly coarse particles, which accounted for approximately 55–80% of the total bioaerosol concentration on both haze days and nonhaze days in Beijing (Gao et al. 2015a). From October 8 to 22, 2014, in Xi'an, the culturable airborne bacteria and fungi were mainly within particle size ranges of 1.1–2.1 μ m (25.0% \pm 6.8%) and 3.3–4.7 μ m (29.4% \pm 4.1%), respectively (Li et al. 2015). In Qingdao, the bacteria[l partic](#page-10-0)les were mainly coarse particles (except in the fall season), whereas the fungal particles followed a log-normal distribution (Qi et al. 2014). Therefore, characteristics of the distribution of bioaerosol particle si[zes va](#page-11-0)ried among regions and depended on geographical and climatic factors and atmospheric and environmental characteristics. The pollutant characteristics in differe[nt regi](#page-11-0)ons caused differences in the spatial distribution of bioaerosols (Li et al. 2011; Wei et al. 2015; Zhong et al. 2016; Lu et al. 2018).

Fig. 3 Size distribution of culturable airborne bacteria at different AQIs (a, excellent weather; b, good weather; c, slight-pollution weather; d, moderate-pollution weather; e, heavy-pollution weather; f, total)

Diurnal variation of the concentration and size distribution of bioaerosols

Concentration of culturable airborne Concentration of culturable airborne Concentration of culturable airborne

bacteria (CFU/m³)

bacteria (CFU/m³)

1000

8000

6000

4000

2000

10000

8000

6000

4000

2000

 $0.65 - 1.1$ 1.1-2.1

 $0.65 - 1.1$ 1.1-2.1

bacteria (CFU/m³

The diur[nal va](#page-11-0)riation in t[he con](#page-11-0)centrati[on of](#page-11-0) culturable airborne bacteria and fungi during the study period is presented in Fig. 5. The lowest concentrations of airborne bacteria and fungi were observed at noon: 3624 ± 2887 and 4412 ± 3946 CFU/m³, respectively. The highest concentrations of airborne bacteria and fungi were registered at night: 5773 ± 4987 and 9780 ± 7233 CFU/m³, respectively. Moreover, [th](#page-7-0)e temporal variations in the culturable bioaerosols during the sampling day were not obvious (P > 0.05). This finding is probably due to the relative stability of the atmospheric environment throughout the sampling day.

The sun radiation was one of the important factors influencing the bioaerosol concentration (Dong et al. 2015). Ultraviolet radiation from the sun probably inhibited the growth of bacteria by damaging their DNA (Pakulski et al. 2007). Given that the surface solar irradiance at noon was often higher than that in the morning and evening (Tong et al. 1997; Xu et al. 2013), the reduction in solar radiation at night reduced the proportion of microbial death, resulting in accumulation of microorganisms (Hwang et al. 2010). The [temp](#page-11-0)oral distribution characteristics of bioaerosols during the study period were attributed to UV radiation. A similar phen[omeno](#page-11-0)n was obs[erved i](#page-11-0)n Beijing in a 1-year data analysis by Gao et al. (2016), which revealed the highest concentration of airborne bacteria and fungi were present at [21: 00](#page-10-0). The lowest concentration was detected at 12:00 and 15:00,

Fig. 4 Size distribution of culturable airborne fungi at different AQIs (a, excellent weather; b, good weather; c, slight-pollution weather; d, moderate-pollution weather; e, heavy-pollution weather; f, total)

respectively. The airborne bacteria concentrations at 9:00 and 17:00 were also found to be significantly higher than that at 13:00 in Han[gzhou](#page-10-0) (Fang et al. 2016).

The diurnal variations in the particle size distribution for culturable airborne bacteria and fungi were similar (Fig. 6), and no statistically significant differences were found during the three sampling diurnal periods ($P > 0.05$), which are depicted in Fig. 5. T[he par](#page-10-0)ticle size of culturable airborne bacteria and fungi was found to mainly be distributed within stage V, within the particle size range of 1.1–2.1 μm and w[it](#page-7-0)h proportions of $36.2\% \pm 16.7\%$ and $46.3\% \pm 17.4\%$, respectively. The diurnal fine particle concentrations of airborne bacteria and fun[gi](#page-7-0) were $47.2\% \pm 19.4\%$ and $46.6\% \pm 16.5\%$ in the morning, $40.7\% \pm 20.8\%$ and $51.1\% \pm 22.5\%$ at noon, and $50.0\% \pm 14.8\%$ and $58.2\% \pm 13.6\%$ at night, respectively.

The particle size is an important parameter for evaluation of the harmfulness of PM for human health (Du et al. 2018a). The deposition efficiency in the human respiratory tract varies among different sizes of particles (coarse particles are mainly deposited in the extrathoracic region, and fine particles can penetrate and deposit deeper in the tracheal, bronchial, and alveolar regions) (Gao et al. 2015a; Li et al. 2017). Moreover, ultrafine particles $(< 0.1 \mu m)$ are deposi[ted at](#page-10-0) a much higher efficiency rate and act as efficient carriers of toxic compounds into the pulmonary alveoli (Kawanaka et al. 2009).

Analysis of factors affecting t[he con](#page-10-0)centration of bioaerosols

To explore the effects of meteorological factors and particles on bi[oaero](#page-10-0)sols, the relations between the concentration of airborne bacterial or fungal aerosols and different particle sizes and temperatures, relative humidity levels, and $PM_{2.5}$, $PM₁₀$, SO_2 , NO_2 , and O_3 concentrations were analyzed by Spearman's correlation method (Fig. 7). Airborne bacteria and fungi from stage I to stage VI and total bacteria and fungi were labeled as BI-VI, FI-VI, TB, and TF, respectively.

Fig. 5 Diurnal variation of concentrations of culturable airborne bacteria and fungi (a, culturable airborne bacteria; b, culturable airborne fungi)

According to Fig. 7 , NO₂ was an important factor influencing the culturable bioaerosol concentration and had a statistically significant association with BIII-VI ($P < 0.05$), TB ($P <$ 0.01), FII-VI ($P < 0.01$ $P < 0.01$ $P < 0.01$), and TF ($P < 0.01$; Fig. 7). Being considered an acid gas, $SO₂$ also showed a positive correlation with the culturable bioaerosol concentration (except at stages BI and FI). This posi[tiv](#page-8-0)e correlation is probably due to the fact that SO_2 and NO_2 can combine with moisture in the air to form SO_4^2 and NO_3 , which are beneficial for the growth of microorganisms (Chen et al. 2008; Dong et al. 201[6\)](#page-8-0). A positive correlation between bioaerosols and acid gases has been observed in several other studies too (Grinn-Gofron et al. 2011; Xie et al. 2018b).

 $O₃$ showed a statistically significant association with BIII-V ($P < 0.05$), TB ($P < 0.01$), FIII-VI ($P < 0.05$), and TF ($P <$ 0.01). The negative correlati[on bet](#page-10-0)ween O_3 an[d airbo](#page-10-0)rne bacterial and fungal concentrations in other stages was also observed (Fig. 7). A high concentration of O_3 was toxic to the [bioae](#page-10-0)rosol, esp[ecially](#page-11-0) after reacting with atmospheric olefins and forming so-called open air factors (Cox et al. 1973; Cox 1995).

Both $PM_{2.5}$ and PM_{10} manifested a positive correlation with culturable bioaerosols of different particle sizes, and there were s[ig](#page-8-0)nificant correlations with BIV, TB, FIII-V, and TF ($P < 0.05$). Because this study was conducted in winter under relatively stable atmospheric conditions, th[e mic](#page-10-0)roor[ganis](#page-10-0)ms adhered to the particles and were difficult to disperse. Therefore, the high concentrations of culturable bioaerosols were investigated at high $PM_{2.5}$ and PM_{10} concentrations. The significant positive correlation between culturable airborne-bacteria aerosols or culturable airborne-fungi aerosols (including coarse, fine, and total aerosols) with AQI $(PM_{2.5})$ was also found in Xi'an during the autumn haze days (Li et al. 2015). On the other hand, we uncovered a significant negative correlation of the culturable airborne-bacteria concentration and culturable airborne-fungal concentration with

Fig. 6 Size distribution of culturable airborne bacteria and fungi at different time points (a and c, culturable airborne bacteria; b and d, culturable airborne fungi)

Fig. 7 Spearman's analysis of correlation between bioaerosols and influencing factors. **Correlation is significant at the 0.01 level (two-tailed). *Correlation is significant at the 0.05 level (two-tailed)

 $PM_{2.5}$ and PM_{10} in Beijing (Gao et al. 2015a; Gao et al. 2016). As carriers of air pollutants, particles adsorb greater amounts of chemical components, which may shorten the survival period of a [micro](#page-11-0)organism (Eeftens et al. 2012; Lu et al. 2018).

Most particle size stages showed a positive correlation with relative humidity, and significant correlations between BIV-V and TB and relative humidity wer[e obser](#page-10-0)ved ($P < 0.05$). Because moisture in the air may alter the integrity of the cell wall or viral capsid (Jones et al. 2004), relative humidity was found to be the main factor affecting the [cultu](#page-10-0)rable bioa[erosol](#page-11-0)s (Jo and Kang 2006; Li et al. 2011). It had different effects on different kinds of microorganisms (Macher et al. 1991; Pasanen et al. 1991; Theunissen et al. 1993). Li et al. (2017) reported high relative humidity can favor microbial growth, resulting in elevated bioaerosol [conce](#page-10-0)ntrations. The high humidity values (about 70–80%) particularly assisted the release of basidiospor[es and](#page-10-0) ascospo[res. H](#page-11-0)igh relative humidity could also trigger spore release, thereby increasing the abunda[nce o](#page-11-0)f spores and im[provi](#page-11-0)ng archaeal diversi[ty \(G](#page-11-0)abey et al. [2010](#page-11-0); Fröhlichnowoisky et al. 2014; Zhai et al. 2018). This was probably the cause of the positive correlation between bioaerosol concentration and relative humidity in this work.

Moreover, negative correlations were noted between culturable bioaerosol concentrations and temperature (Fig. 7). This finding is consistent with the results reported [by L](#page-10-0)i et al. (2017) and Lu et al. (2018) , who also [found](#page-11-0) a negative and positive correlation between bioaerosols (bacteria and fungi) and temperature and relative humidity, respectively. High temperature was disadvantageous for the microorganisms because it enhanced the release of toxic compounds and promoted their chemical reactions to occur on particulate surfac[es \(G](#page-11-0)ao et al. 2016). [The](#page-11-0) increased temperature could also speed up convective air movements, which might enhance bacterial dispersal and dilution effect, leading to a decrease of bioaerosol concentrations in the atmosphere (Smets et al. 2016; Zhong et al. 2016). Nonetheless, some investigators have reached the opposite conclusions. Bioaerosol concentration increases with incre[asing](#page-10-0) relative humidity at sufficiently high temperatures and can hardly be influenced at low temperatures (Kethley et al. 1957). Webb and Dumasia (1968) reported that the decline rate of bioaerosols is dependent on both temperat[ure an](#page-11-0)d relative hu[midity.](#page-11-0)

The formation and size distribution of the bioaerosols were complicated, and many factors were involved in this process (Chen et al. 2012). The PM concentration was considered the most significant factor on the bioaerosol size d[istribu](#page-10-0)tion, as the airborne bac[teria a](#page-11-0)nd fungi can attach to the surfaces of PM suspended in the atmosphere (Gao et al. 2015b; Dong et al. 2016). In this work, most of the particle size stages showed a positive correlation with $PM_{2.5}$ and PM_{10} . However, exceptions were found in the measurements of BI and FI, whi[ch wa](#page-10-0)s likely due to the aggregation of fine particulates that frequently occurred on hazy days (Kulmala et al. 2004). Sources of bioaerosols played a leading role in shaping the characteristic of bioaerosol particle size an[d had a](#page-10-0) larger impac[t than](#page-10-0) that caused by meteorological conditions (Zhai et al., 2018). The bioaerosol generated from the respiratory tract was found to be smaller than that generated from dust sources (Hoeksma et al., 2015). At a subway station, Dybwad et al. (2014) found a significantly larger fraction of bioaerosol [\(parti](#page-10-0)cles between 1.1 and 3.3 μ m) during the daytime than at nighttime. Anthropogenic activities (mainly passengers) were demonstrated to be major sources of airborne bacteria and predo[minan](#page-11-0)tly contributed to the bioaerosol particles of this range. Relative humidity is an important meteorological parameter influencing the [size d](#page-10-0)istribution of bioaerosol. High relati[ve hum](#page-10-0)idity will augment the probability of deposition due to the presence of suspended particles that absorb ambient moisture, leading to an increase in particle weight and size (Zhen et al., 2017). The bigger and heavier particles will have

Table 4 Individual non-carcinogenic risks corresponding to different exposure pathways

a higher settling velocity in the air. Temperature could indirectly influence the size distribution of bioaerosols by changing the parameter of cross-ventilation that decides the suspension and diffusion of microbes (Zhai et al., 2018). Moreover, factors such as organic carbon, elementary carbon, NH_4^+ , SO_4^2 ⁻, NO_3^- , metals, and polycyclic aromatic hydrocarbons have been s[hown t](#page-11-0)o affect the size distributions of the bacterial aerosol (Lai et al. 2010; Chen et al. 2012). The size distributions of airborne fungi were influenced by various factors such as microorganism species, spore age, sample culture medium, and differences in aggregation rates of sp[ores](#page-11-0) (Nasir et al. 2012).

Noncarcinogeni[c exp](#page-11-0)osure risk [asses](#page-10-0)sment

Assessment of culturable bioaerosols during the sampling period was conducted in this study, and it is important for people to understand the risk of air quality for the living environment [and](#page-11-0) [ta](#page-11-0)ke preventive measures. Table 4 summarizes the hazard quotients for inhalation (HQ_{inh}) and dermal (HQ_{dermal}) routes and the HI of bioaerosols at different time points and air quality classes during the sampling period.

In this study, the highest diurnal risk of exposure to bioaerosols was noted at night, and with the rise in AQI, the risk increased too. The ranges of HQ_{inh} for adult males, adult females, and children were 1.28×10^{-2} to 1.04×10^{-1} , $1.04 \times$ 10^{-2} to 8.49×10^{-2} , and 5.19×10^{-3} to 4.24×10^{-2} , respectively, which were several orders of magnitude higher than those of HQdermal. Therefore, the risk of exposure to bioaerosols in the atmosphere was primarily associated with the respiratory inhalation pathway. The risk of exposure of skin was negligible. For different populations, the order of HQ_{inh} values was as follows: adult males > adult females > children, whereas that of HQ_{dermal} values was children $>$ adult females $>$ adult males.

Nevertheless, the risk of exposure to bioaerosols analyzed in this study was relatively low when compared with those analyzed in other studies (Li et al. 2013). We recommended that outdoor activities be avoided when AQI is higher than

150, especially at night. Some necessary protective measures are still essential for people living in high-AQI atmospheric conditions. The noncarcinogenic risk assessment model used in this work was based on the concentration of airborne bacteria. The community structure of opportunistic pathogenic airborne bacteria is closely relate[d to](#page-11-0) human health (Fan et al. 2019), but is not considered in this evaluation process. In addition, the bioaerosols investigated in this study contained only culturable airborne bacteria, which represent only a small part of the airborne microorganisms. In further studies, other analytical methods, such as molecular tools, should be considered to comprehensively reveal the species and quantities of bioaerosols and their effects on public health.

Conclusion

In this study, we monitored the concentration of culturable airborne bacteria and fungi at different air quality levels and during different diurnal periods in winter in Xinxiang, China. The concentrations of culturable airborne bacteria and fungi were strongly linked to air quality (AQI). The particle sizes of bioaerosols were mainly within stage V $(1.1-2.1 \mu m)$, and fine PM accounted for $45.9\% \pm 18.9\%$ of airborne bacteria and $52.0\% \pm 18.5\%$ of airborne fungi, respectively. Analysis of diurnal variation of bioaerosols showed that the concentration was obviously higher at night than in the morning or at noon. Bioaerosol concentration positively correlated with humidity and concentrations of $PM_{2.5}$, PM_{10} , SO_2 , and NO_2 and negatively correlated with the concentration of O_3 . The risk of exposure to bioaerosols among humans was primarily associated with the respiratory inhalation pathway, and the risk of skin exposure was negligible.

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