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# Assessment of bioaerosols, $PM_{2.5}$, and $PM_{10}$ in liver transplantation operating rooms in Tehran, Iran: Implications for air quality

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**Abstract**

**Background:** Research on the relationship between bioaerosols and particulate matter (PM) concentrations is necessary, especially in hospitals where airborne bioaerosols can facilitate disease transmission. This study aimed to investigate the relationship between PM ($PM_{2.5}$ and $PM_{10}$) and bioaerosols, as well as the factors influencing them (temperature and humidity), in the air of liver transplant operating rooms (LTOR) at Imam Khomeini Hospital in Tehran.

**Methods:** Bioaerosol samples (32 samples) were collected using the passive sampling method, employing open-door plates containing culture medium, during June and July of 2019. PM samples were obtained concurrently with bioaerosol samples using the GM8803 air quality detector, during four one-hour periods. Simple linear regression analysis was performed to determine the relationship between bioaerosol and PM concentrations.

**Results:** It was revealed that the average concentrations of $PM_{2.5}$ and $PM_{10}$ were 17.8 (SD = 2.2) and 27.0 (SD = 2.6) μg/m$^3$, respectively. Additionally, the average concentrations of bacterial and fungal bioaerosols were 2132 (SD = 837) and 550 (SD = 189.4) CFU/m$^2$.h, respectively. Linear regression analysis demonstrated a strong correlation between bacterial bioaerosols and PM concentrations, whereas the relationship with fungal bioaerosols was relatively weaker.

**Conclusion:** The findings of this study indicate that the indoor air in LTOR exhibits a higher level of microbial contamination than the recommended guidelines for high-risk environments. To improve the air quality in LTOR, it is recommended to implement periodic microbial monitoring, ensure the proper functioning of ventilation systems, and pay attention to their maintenance and operation.

**Keywords:** Air pollution, Indoor, Particulate matter (PM), Operating room (OR), Passive sampling

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## Introduction

In the realm of healthcare, it is imperative to address the challenges associated with hospitalization, and one critical concern is exposure to airborne bioaerosols. Hospitals and medical facilities are tasked with effectively managing and reducing the presence of bioaerosols to create a safer environment for both patients and staff (1).

Bioaerosols comprise a wide range of airborne particles, including viruses, pollen grains, algae, plant debris, microbiological insect fragments, as well as human and animal skin cells, along with bacterial and fungal spores. The aerodynamic size of bioaerosols spans from 0.02 to 100 μm, with particles smaller than 10 μm presenting notable health risks, especially within hospital settings. These bioaerosols significantly contribute to indoor air pollution, constituting approximately 5 to 34% of the overall pollution load (2,3).

Atmospheric PM consists of a diverse assortment of particles originating from both chemical and biological sources, which can significantly impact the health of both

humans and animals. Exposure to PM has been correlated with a range of health issues, including asthma, lung cancer, and cardiopulmonary diseases. Understanding the intricate relationship between bioaerosols and PM concentrations, particularly within hospital environments, is a complex issue that necessitates comprehensive investigation (4,5).

Multiple studies have extensively investigated the occurrence of airborne microorganisms in indoor and outdoor settings, particularly in operating rooms (ORs). Various factors, including door movements, postures of medical personnel, tool exchanges, and ventilation systems, play a significant role in the presence of airborne PM and microbiological contamination within ORs. The positioning of medical staff during surgical procedures, for example, can influence the dynamics of airflow, potentially resulting in heightened microbial contamination within the surgical zone (6-8).

Ventilation systems in hospitals play a critical role in the transmission of illnesses, as research has shown a connection between these systems and the dissemination of airborne bioaerosols. Opening doors within ORs can disrupt positive room pressure, creating an opportunity for contaminants from surrounding areas to enter. This dynamic has significant implications for patient safety, particularly during surgical procedures, which can become more complex and pose a higher risk of infection in environments where air quality is compromised (9-15).

Pathogenic bacteria and fungi, notably *Staphylococcus aureus* and *Candida albicans*, pose a significant risk in hospital-acquired infections, particularly within surgical environments. It is crucial to understand the sources of these microorganisms to implement effective infection control measures. Patients, visitors, ventilation systems, and room air all contribute to the dissemination of airborne bacteria and fungi, emphasizing the necessity of comprehensive preventive strategies. Hospital-acquired infections, often resulting from surgical site infections caused by airborne bioaerosols, lead to increased mortality rates, complications, longer hospital stays, and elevated healthcare expenses. Moreover, poor indoor air quality extends beyond infections, manifesting as acute respiratory complications, fatigue, headaches, and an elevated cancer risk (14,16,17).

To implement effective control measures for hospital infections, identifying the source of contamination is of utmost importance. Two methods commonly used for measuring airborne bioaerosols are active and passive sampling, each offering distinct advantages. The active sampling method is recommended for quantifying the concentration of all inhalable viable particles, while the passive method, which involves using sediment plates containing a culture medium, is preferred for assessing microorganisms deposited on surfaces. The simplicity and cost-effectiveness of the passive technique make it a practical choice for routine monitoring purposes (18-22).

In the study conducted by Palulun et al (23), titled as "Identification of Airborne Aerobic Bacteria in the ICU," the researchers reported that the predominant types of airborne bacteria in the air were *Bacillus*, coagulase-negative *Staphylococcus*, and *Staphylococcus aureus*. Additionally, the study found that the concentration of microorganisms in the indoor air exceeded the standard value. Vahidmoghadam et al (24) conducted a study on the concentration of PM and the microbiological quality of indoor air in the ICU of Kashan Hospital. They found that gram-positive staphylococci were the most prevalent bacterial species, and *Aspergillus*, *Penicillium*, and *Cladosporium* were the most abundant fungal species. The maximum concentration of $PM_{10}$ in the pediatric intensive care unit (PICU) was reported as 59.19 μg/m$^3$, while the maximum concentration of $PM_{2.5}$ in the neonatal intensive care unit (NICU) was measured at 20.23 μg/m$^3$. In a study by AlRayess et al (25), characterizing airborne bacteria and PM in the ICU of Beirut, Lebanon, PM levels in several ICUs were reported above the established international guidelines for 24-hour exposure, and they reported no statistically significant relationship between the bacterial load and the concentration of PM.

Understanding the relationship between microorganisms and PMs in the OR setting is vital, and research in this area has been limited thus far. While previous research in Iran has explored bioaerosols and air quality in hospital environments, the present study specifically focused on liver transplant operating room (LTOR). This particular setting poses unique challenges due to the presence of immunocompromised patients and the need to maintain high air quality standards to minimize the risk of airborne infections.

The present research incorporates the assessment of both $PM_{2.5}$ and $PM_{10}$, which by considering both $PM_{2.5}$ and $PM_{10}$, we can assess the potential risks posed by fine and coarse particles, thereby contributing to a more accurate assessment of air quality and its implications for the health of patients and healthcare workers. Furthermore, this study aimed to provide implications for air quality improvement in LTORs. By identifying the sources and characteristics of bioaerosols and PM, we can suggest targeted measures and interventions to mitigate the presence of harmful pollutants. This emphasis on practical implications and recommendations distinguishes our research and highlights its potential for directly impacting patient safety and well-being. Through these innovative elements, our study seeks to advance the understanding of bioaerosols, $PM_{2.5}$, and $PM_{10}$ in LTOR, and contribute to the development of strategies for improving air quality and ensuring the highest standards of care for patients undergoing liver transplant procedures.

The main aim of this study was to identify and quantify the microbial loads in the air in LTOR in Imam

Khomeini Hospital in Tehran. Specific objectives include comparing bioaerosol levels with established standards, measuring PMs levels ($PM_{2.5}$ and $PM_{10}$) in LTOR, as well as investigating the relationship between bioaerosols and PMs. In addition, in this study, the effect of temperature and relative humidity (RH) on pollutants in the air was also investigated.

## Material and Methods

### *Sampling site*

This cross-sectional study was conducted in the LTOR at Imam Khomeini Hospital in Tehran. Imam Khomeini Hospital is a specialized and sub-specialized teaching hospital affiliated with Tehran University of Medical Sciences. With 1500 beds, it holds the distinction of being the largest hospital in the Middle East in terms of treatment capacity. The sampling for this study took place during surgical operations within the LTOR, which has an area of 22 $m^2$. Samples were collected from four cardinal directions (north, south, east, and west) at a distance of 1 m from the floor, at least 1 m away from walls or any obstacle. The researcher adhered to strict hand hygiene protocols and utilized personal protective equipment such as gowns, masks, hats, gloves, and shoes during the study.

### *Sampling procedure*

#### *Sampling procedure to airborne bioaerosols*

During a specific period spanning June and July 2019, air sampling of the LTOR was performed. Intraoperative sampling offers several distinct advantages that contribute to a comprehensive understanding of bioaerosol dynamics in the surgical environment. By sampling during this procedure, we can directly assess the production and dispersion of bioaerosols in real time, which is critical for assessing potential risks and developing effective mitigation strategies. In addition, intraoperative sampling enables us to examine the influence of various surgical factors such as the type of procedure, the surgical instruments used, and the ventilation systems used. The sampling took place four times, specifically during afternoon working shifts between 14:00 and 15:00. Passive sampling was employed to collect bioaerosol samples following the 1.1.1 scheme (21,26).

According to this plan, Petri dishes with a 9 cm diameter should be placed 1 m above the floor and 1 m away from the walls for 1 hour (20). Steps 1 and 2 (sampled in June at one-week intervals) and steps 3 and 4 (sampled in July at one-week intervals) under the same conditions as the preceding stages are represented by the four sampling times in LTOR. Four samples of fungi and four samples of bacteria were taken from the OR's air during each sampling phase. Thirty-two samples (16 bacterial and 16 fungal) were gathered. The appropriate culture media for both bacteria and fungi were prepared before sampling. For bacterial bioaerosols, it meant using Tryptic Soy Agar (TSA) culture medium, which contained 500 mg/L of cycloheximide ($C_{15}H_{23}NO_4$) to inhibit fungal proliferation, and for fungal bioaerosols, it meant using Sabouraud Dextrose Agar (SDA) culture medium, which contained 100 mg/L of chloramphenicol ($C_{11}H1_2C_{l2}N_2O_5$) to inhibit bacterial proliferation. Both culture media were sterilized for 15 minutes. The plates containing the culture medium were transferred to LTOR using the necessary equipment such as alcohol, zip cap, and cool box.

### *Incubation: Creating optimal conditions for airborne bioaerosols growth*

To ensure accuracy and prevent contamination, the plates were sealed with parafilm after sampling and promptly transported to the laboratory in a cool box. The TSA plates were inverted and incubated at a temperature of 35 ± 0.5 °C for 48 hours (27,28) while the SDA plates were kept in the laboratory at a temperature of 25-28 °C for 3 to 7 days (29). The colonies on the plates were then counted.

### *Procedures for the detection of airborne bioaerosol isolates*

Standard microbiological procedures were meticulously followed to conduct the necessary tests for the identification of bacterial and fungal isolates. Bacterial isolates were examined for colony morphology and identified using gram staining and relevant biochemical tests. Different methods were employed to differentially diagnose various types of fungi, including observing colony appearance and microscopic forms using a light microscope at 400 × magnification. The appearance characteristics taken into consideration for fungal diagnosis include growth rate, colony shape (flat, convex, regular, irregular), colony view, colony color (white, yellow, green, blue, cream, purple), presence of pigment, and color of the back of the colony due to pigment production. Genus identification was carried out following the classification method proposed by Ainsworth and Baron (30,31). Finally, the results were expressed in colony forming units per square meter per hour (CFU/$m^2$.h) (32,33).

### *Sampling procedure for PMs*

To assess the potential impact of PM concentration on the presence of bioaerosols, the measurement of LTOR air particles was conducted simultaneously with the sampling of bacterial and fungal bioaerosols. The sampling of PM was performed following the guidelines recommended by the World Health Organization (WHO) and the Environmental Health Agency (EPA) (34). Four one-hour sampling periods were carried out, with stages 1 and 2 taking place in June at one-week intervals, and stages 3 and 4 conducted in July at one-week intervals, all under similar conditions in the LTOR during afternoon working shifts between 14:00 and 15:00.

To measure $PM_{2.5}$ and $PM_{10}$ in the indoor air of the LTOR, the GM8803 air quality detector was utilized. This portable detector, which is equipped with a built-in digital particle content sensor, utilizes a laser emission principle. It can continuously detect particle content and provide timely responses, with a minimum particle size resolution of 0.3 μm. The measurement of particulate matter was performed at a distance of 1 m from the floor of the OR, at the breathing level of the patients, as well as 1 m from the patient's bed. The measurements were taken in the four directions (east, west, south, and north) of the OR continuously for one hour. Sixty data points were recorded per hour, and the results were reported as the average concentration over the entire hour.

### *Verification of the operating parameters*

Operational characteristics, such as (*i*) air temperature and (*ii*) RH, were measured during the microbiological and PM sampling tests to make sure the LTOR's indoor environment complied with the guidelines suggested by the Ministry of Health policies (35). A portable monitor (Lutron model PHB-318) was used to detect temperature and RH concurrently with the bioaerosol sampling process.

### *Quality assurance and quality control (QA/QC)*

Particulate measurement using the GM8803 air quality monitor required strict QA/QC measures to ensure accurate and reliable results. To maintain the accuracy of the measurement, the instrument was regularly calibrated, and its readings were matched against reference standards or calibrated instruments. Periodic verification tests using known particle concentrations or approved verification equipment were conducted to verify device performance. Proper maintenance, including cleaning the sensors and replacing consumables, ensured long-term performance. Data validation methods including filters, statistical analysis, and comparison with other monitoring devices or stations were employed to identify and correct anomalies or errors. Comprehensive documentation of QA/QC procedures, calibration records, maintenance reports, and corrective actions was maintained to ensure traceability and accountability. These QA/QC criteria have maintained the integrity of the particulate measurements obtained from the GM8803 and have enabled reliable air quality assessment and monitoring.

### *Data analysis*

This study employed descriptive statistical parameters, including mean, standard deviation, median, minimum, and maximum, to describe the concentration of PM, bacterial and fungal bioaerosols. Linear regression analysis was utilized to examine the relationship between particle concentration and the presence of bacterial and fungal bioaerosols. The correlation between microbial load and temperature as well as RH during each sampling step was evaluated using Pearson correlation. Furthermore, Microsoft Excel 2016 was utilized to generate all graphs.

## Results

The average of microbial load, RH, and temperature in the LTOR according to sampling steps are presented in Table 1.

Pearson's correlation coefficients for microbial load-RH and microbial load-temperature were -0.3 and -0.09, respectively. Table 2 presents a summary of the bacterial and fungal bioaerosol concentrations (CFU/$m^2$.h) during the sampling process. Using the passive method, a total of 217 bacterial CFU and 56 fungus CFU were collected from 32 plates. The bacterial bioaerosols identified were *Staphylococcus aureus*, *Staphylococcus epidermis*, and *Escherichia coli*. The study identified several types of fungal bioaerosols in the LTOR air, including *Aspergillus*, *Cladosporium*, *Rhodotorula*, *Monilia*, *Paecilomyces*, and *Penicillium*. Specifically, *Aspergillus niger* and *Aspergillus ochraceous* were detected among the *Aspergillus* fungi. At the sampling site, the mean concentration of bacteria was 2132 CFU/$m^2$.h (SD = 838 CFU/$m^2$.h) while the mean concentration of fungi was 550 CFU/$m^2$.h (SD = 189.5 CFU/$m^2$.h) with the highest average values observed in the third and fourth sampling stages, respectively. The frequency of bacterial bioaerosols identified in the present study was as follows: *S. aureus* (52.5%), *S. epidermidis* (39%), and *E. coli* (6.4%) (Figure 1). The present research revealed that *Aspergillus* (50%), *Penicillium* (19.6%), and *Cladosporium* (16%) species were the most commonly observed fungal genera throughout all sampling steps. Less frequently, *Paecilomyces* (5.3%), *Rhodotorula* (5.3%), and *Monilia* (3.3%) species were also detected (Figure 2). The average total concentration of $PM_{2.5}$ and $PM_{10}$ particles at the sampling site was 17.8 μg/$m^3$ (SD = 2.2 μg/$m^3$) and 27.0 μg/$m^3$ (SD = 2.6 μg/$m^3$), respectively (Table 3 and Figure 3). Statistical linear regression analysis was performed to examine the correlation between bioaerosol concentration (bacterial and fungal) and PM ($PM_{2.5}$ and $PM_{10}$). As depicted in Figure S1 (a, b, c, and d), the linear regression analysis demonstrated a strong correlation between bacterial bioaerosols and $PM_{2.5}$ concentrations ($R^2 = 0.7$, $P < 0.05$) and $PM_{10}$ concentration ($R^2 = 0.6$, $P < 0.05$). However, the linear regression analysis

**Table 1.** Microbial load (CFU/$m^2$.h), temperature, and relative humidity levels in the LTOR during the sampling process

| Sampling steps | Microbial load | Temperature (°C) | Relative humidity (%) |
|---|---|---|---|
| 1 | 1651 | 23 | 27 |
| 2 | 2555 | 23 | 26 |
| 3 | 3813 | 22 | 25 |
| 4 | 2712 | 23 | 25 |
| Total | 2683 | 22.75 | 25.75 |

**Table 2.** The levels of bacterial and fungal bioaerosols (CFU/m².h) in the LTOR air during the sampling process

| Sampling steps | Fungi (n=16 (4×4)) | | | Bacteria (n=16 (4×4)) | | |
|---|---|---|---|---|---|---|
| | Min-Max | Mean±SD | Median | Min-Max | Mean±SD | Median |
| 1 | 157 – 629 | 393±188 | 393 | 943 – 1729 | 1258±330 | 1179 |
| 2 | 314 – 629 | 511±141 | 550 | 1415 – 2830 | 2044±581 | 1965 |
| 3 | 471 – 786 | 589±141 | 550 | 2673 – 3931 | 3224±566 | 3145 |
| 4 | 629 – 786 | 707±78 | 707 | 1729 – 2359 | 2005±267 | 1965 |
| Total | 157 – 786 | 550±189.5 | 629 | 943 – 3931 | 1.7±838 | 1965.5 |

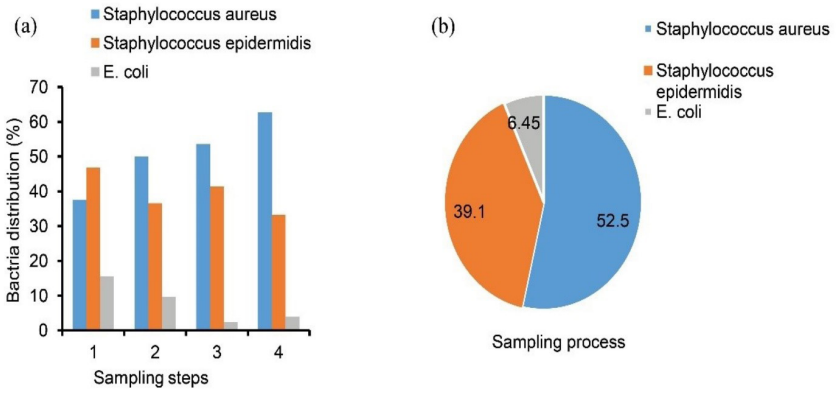


**Figure 1.** Bacteria bioaerosols distribution (%) in the LTOR air during the sampling steps (a) and sampling process (b)

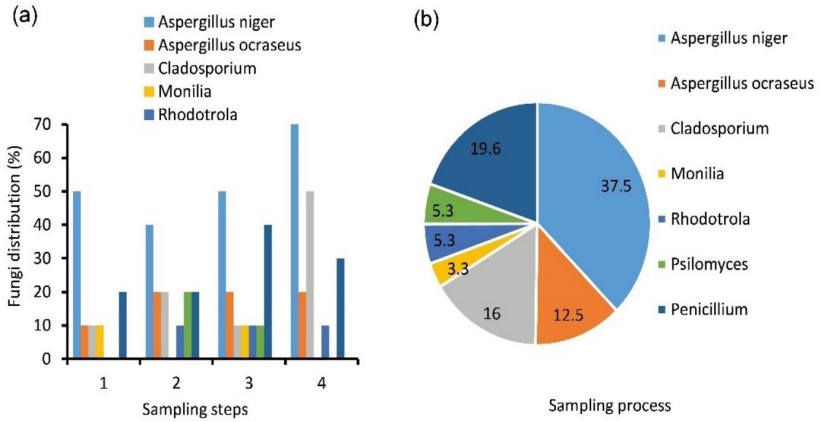


**Figure 2.** Fungal bioaerosols distribution (%) in the LTOR air during the sampling steps (a) and sampling process (b)

between fungal bioaerosols and $PM_{2.5}$ concentrations ($R^2=0.3$, $P<0.05$) and $PM_{10}$ concentration ($R^2=0.2$, $P<0.05$) showed a weaker relationship.

The air microbial pollution levels (CFU/m².h) in the LTOR air during the sampling process compared to the existing standard are shown in Figure 4.

## Discussion

### *Temperature and RH*

The measured data indicates that the ambient temperature and RH fall within the recommended values specified in the Ministry of Health Standard's infection control policies and procedures (Table 1) (35). The Centers for Disease Control and Prevention (CDC) and the Healthcare Infection Control Practices Advisory Committee (HICPAC) recommend temperature ranges of 21–24 °C and 23–27 °C for most hospital wards during winter and summer, respectively (36). According to data from Canadian hospitals, the surgical site infection rate increased to 10.7% with an RH of 60–85% but was between 3.3 and 5.6% when OR were prescribed an RH below 60% (37). Throughout the sampling process, temperatures and RH remained mostly stable and had no discernible effects on the microbial loads that were found. Higher values of these factors generally encourage the growth of microorganisms (38).

AlRayess et al (25) in their study on the profile of airborne bacteria and PM in the ICU, reported that due to the constant temperature and relative humidity parameters during sampling in the patient room, a significant

**Table 3.** The levels of PMs ($PM_{2.5}$ and $PM_{10}$) (μg/m³) in the LTOR air during the sampling process

| Sampling Steps | $PM_{2.5}$ | | | $PM_{10}$ | | |
|---|---|---|---|---|---|---|
| | Min–Max | Mean±SD | Median | Min–Max | Mean±SD | Median |
| 1 | 15 – 18 | 16.05±0.9 | 16 | 23 – 27 | 25.15±0.9 | 25 |
| 2 | 15 – 21 | 17.46±1.6 | 17 | 23 – 28 | 26.11±1.0 | 26 |
| 3 | 19 – 24 | 20.48±1.4 | 20 | 29 – 34 | 31.2±1.4 | 31.5 |
| 4 | 15 – 21 | 17.15±1.7 | 17 | 24 – 27 | 25.65±0.7 | 26 |
| Total | 15 – 24 | 17.81±2.2 | 17 | 23 – 34 | 27.02±2.6 | 26 |

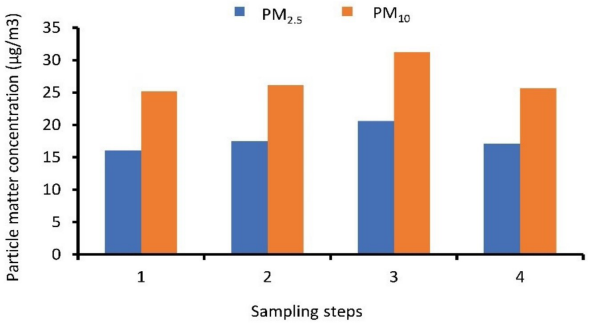


**Figure 3.** The concentrations of the PM ($PM_{2.5}$ and $PM_{10}$) in the LTOR air during the sampling process

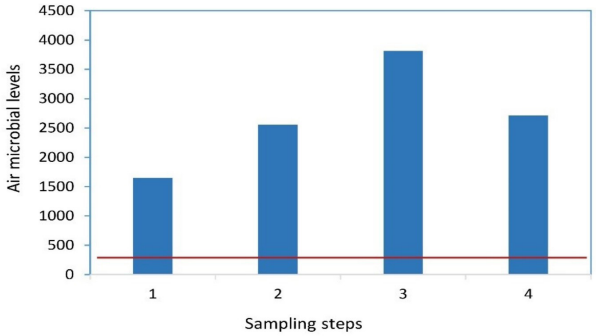


**Figure 4.** Air microbial pollution levels (CFU/m².h) in the LTOR air during the sampling process compared to the existing standard (maximum levels of the index of microbial air contamination: MAL of IMA) in OR (red line): 91 CFU/m².h)

correlation between the total burden microbial and parameters mentioned above were not found. According to the study by Hansen et al (39), there was a substantial correlation between temperature and humidity and the concentrations of molds that could grow at 22 °C. Hwang et al (38) discovered that temperature and total airborne microorganisms were significantly correlated, although RH was not. The New York City Department of Health and Mental Hygiene states that to prevent the growth of fungi, interior spaces should have RH levels below 65%. Additionally, keeping humidity levels low is another tip from the department to avoid moisture condensation on windows and other surfaces (40).

### *Frequency and types of species*

Prior studies have demonstrated that surgical site infections are caused by bacteria growing in surgical wounds (41,42). The present study detected the presence of *Staphylococcus aureus*, *S. epidermis*, and *E. coli* as bacterial bioaerosols. *S. aureus* was the most prevalent bacterial genus observed. *S. aureus* (52.5%) and *S. epidermidis* (39%) were the most common bacterial bioaerosols detected in the present study (Figure 1). The findings of this study are consistent with those of other studies (43). In this study, 93% of the bacterial bioaerosols were identified as gram-positive. Previous studies have reported the proportion of gram-positive bacteria to be 88% (44), 90%–92% (45), 89% (46), 100% (32,47), 92%–100%, and 77.6%–80.8% (48,49). Gram-positive bacteria are more prevalent than gram-negative bacteria in both macro and micro environments. This is due to their high resistance to unfavorable environmental conditions and their presence in the natural flora of various organs in animals and humans (43,45-47,49-51). In our study, *S. epidermis* was found to be the second most prevalent bacteria after *S. aureus*. Coagulase-negative staphylococci, such as *S. epidermidis*, *S. saprophyticus,* and *Staphylococcus haemolyticus*, are important causes of infection in high-risk groups. Staphylococcal infections are primarily transmitted through direct contact, with OR staff being considered the main carriers. Individuals with underlying diseases and weakened immune systems are more susceptible to staphylococcal infections (52). Another study reported that *E. coli*, *Pseudomonas*, *Klebsiella*, and *S. aureus* were the most frequently detected bacterial bioaerosols (53).

Our research revealed that *Aspergillus* (50%), *Penicillium* (19.6%), and *Cladosporium* (16%) species were the most commonly observed fungal genera throughout all sampling steps (Figure 2). These findings confirm the presence of fungal bioaerosols in the LTOR, which has been reported in previous studies (31,33,44,54,55). Numerous studies have linked Aspergillus fungal bioaerosol to hospital-acquired infections in this field of research (31,56,57). The presence of fungi in LTOR confirms the presence of fungal spores. Airborne transmission of *Aspergillus* is a significant factor in the spread of nosocomial infections to vulnerable individuals (58). According to the study by Mahdavi Omran and Sheidfar (59), the most common fungal bioaerosol identified in hospital air samples is *Penicillium*. Hashemi et al (60) reported that *Penicillium* is the predominant fungal bioaerosol found in hospital

air samples. According to the study conducted by Panagopoulou et al. (61), the genus *Aspergillus* was found to have the highest number of fungal bioaerosols detected in the hospital. The abundance of *Aspergillus, Penicillium*, and *Cladosporium* fungi in the LTOR can be attributed to several factors. These fungi are known for their ability to produce small and lightweight spores, facilitating their easy transfer and long-term survival in various weather conditions. Additionally, they can obtain necessary resources from different sources.

*Aspergillus* species, in particular, can cause *Aspergillosis*, skin and ear infections, and are commonly transmitted through inhalation, posing a risk to susceptible individuals. The fungi identified in this study hold significance as either pathogenic or opportunistic pathogens in the field of medicine. Considering that the hospital in question serves as both an educational and treatment center, one of the reasons for the high concentration of bacterial and fungal bioaerosols in its LTOR is likely the large number of surgeries performed. The frequency of airborne bioaerosols observed in studies varies due to factors such as the type of ventilation system, regular monitoring, disinfectants used, proximity to the street, number of visitors, sampling season, and adherence to health protocols by staff and patients.

According to the WHO's Environmental Monitoring of Clean Rooms Standard and the Malaysian Ministry of Health's Policies and Procedures on Infection Control Standard, the only microorganisms present in the cleanroom overall are bacteria and fungus (62,63). To the best of the author's knowledge, no study mentioned the existence of microorganisms, such as viruses, in an OR.

In every sampling step, the concentration of bacterial bioaerosols was greater than that of fungal bioaerosols. Consistent with our findings, another study found that at all sampling locations, the concentration of bacterial bioaerosol was often higher than that of fungal bioaerosol (26). In a study conducted by Sarıca et al (64) in Turkey, it was also found that the concentrations of bacteria were higher than those of fungi. The availability of more indoor bacterial sources and more environmental conditions that favor bacterial development are the causes of this outcome (31). Table 4 shows the comparison results of bacteria and fungi in the air reported in the present study with those reported in different studies.

### *Comparison of air microbial pollution levels with available standards*

As Iranian official documents do not offer any national guidelines or standardized limits for the index of microbial air contamination, we rely on the Swiss Hospital Association standards and other applicable standards as reference points for determining the maximum levels of the index of microbial air contamination (referred to as MAL of IMA) in operating theaters (red line, > 91 CFU/m$^2$.h) (Table 5). The average concentration of the total bioaerosol in the sampling of the first to the fourth stage was higher than the mentioned standards (Figure 4). This indicates that the investigated OR is highly contaminated with bacterial and fungal bioaerosols in the air, posing a risk of bioaerosol contamination for patients requiring liver transplant surgery and the OR staff. The study conducted by Choobineh et al (69) found that the concentration of bioaerosols in the OR exceeded the recommended standard, which is consistent with the findings of this research. According to the research conducted by Dedashti et al (70), the OR was identified as the most heavily contaminated area in terms of bacterial presence. Good microbiological air quality was found in a study using a similar methodology, where the average microbial load in various hospital areas was lower than the standard, which is inconsistent with the results of the

**Table 4.** Comparison of bacteria and fungi observed in this study with those in different studies

| Study (y) | Title | Airborne Bacteria and Fungi | Reference |
|---|---|---|---|
| This study | Bioaerosols and $PM_{2.5}$ and $PM_{10}$ Assessment in Liver Transplantation Operating Rooms in Tehran, Iran: Implications for Air Quality Improvement | *Staphylococcus aureus, Staphylococcus epidermis, Escherichia coli, Aspergillus, Cladosporium, Rhodotorula, Monilia, Paecilomyces, Penicillium* | - |
| Palulun et al (2024) | Identification of airborne aerobic bacteria in the intensive care room using MALDI-TOF MS | *Bacillus, coagulase-negative Staphylococcus and Staphylococcus aureus* | (23) |
| Vahidmoghadam et al (2023) | Determining the Concentration of Particulate Matters and Microbiological Quality of Indoor Air in Intensive Care Units of Kashan Hospital, Iran | *Staphylococcus, Diphtheroid, Bacillus, Aspergillus, Penicillium, Cladosporium* | (24) |
| Chen et al (2024) | Pathogenic bacteria and fungi in bioaerosols from specialized hospitals in Shandong province, East China | *Vibrio metschnikovii, Staphylococcus epidermidis, Staphylococcus haemolyticus, Fusarium pseudensiforme, Aspergillus ruber,* | (65) |
| Obaid (2024) | Assessment of Air Quality Containing Fungi in Al-Nu'man Teaching Hospital | *Penicillium, Aspergillus, Alternaria,* Yeast species, *Rhizopus, Fusarium* | (66) |
| **Ye et al (2024)** | Distribution characteristics and analysis of fungal aerosol concentration and particle size in air-conditioned wards in Wuhan | *Aspergillus, Penicillium, Cladosporia, Alternaria, Trichoderma, Rhizopus* | (67) |
| Montazer et al (2020) | Microbiological analysis of bacterial and fungal bioaerosols from burn hospital of Yazd (Iran) in 2019 | *Citrobacter freundi, Klebsiella pneumoniae, Escherichia coli, Staphylococcus epidermidis, Staphylococcus aureus, Staphylococcus saprophyticus, Penicillium, Alternaria, Aspergillus niger, Aspergillus flavus* | (68) |

present study (71).

### *The concentration of PMs ($PM_{2.5}$ and $PM_{10}$)*

The findings demonstrated that throughout every sampling step, the concentration of $PM_{10}$ was higher than that of $PM_{2.5}$. Compared to smaller particles, particles in this size range can be deposited in one hour more readily due to their higher deposition velocity. It was anticipated that larger particles (greater sedimentation rate) would have a higher concentration based on the passive sampling method. Adherence to certain health standards in the OR, such as implementing traffic restrictions and closing the entrance to other departments, may indicate a lower concentration of particles.

Several factors, such as the ventilation system, number of patients and employees, and adherence to hygiene practices by employees, can influence the concentration of suspended particles in the OR (73). The study by Rezaei et al (74) showed that in 80% of cases, the hospital room's $PM_{10}$ particle concentration surpassed the WHO threshold, while the concentration of $PM_{2.5}$ particles exceeded the EPA level in 42% of cases and the WHO standard in 64% of cases. Another study by Basiri et al (75) reported an average concentration of 29 μg/m$^3$ for $PM_{10}$, 24.2 μg/m$^3$ for $PM_{2.5}$, and 20.9 μg/m$^3$ for $PM_{10}$. Table 6 shows the comparison of PM concentration in the present study with that in different studies.

### *The relationship between PM and bioaerosols*

It is crucial to consider the impact of PM on the concentration of bioaerosols since the sampling duration for PM was consistent with that of bioaerosols sampling.

**Table 5.** Air total microbial count (according to Fisher) in different hospital environments (CFU on Petri dishes 9 cm in diameter, with blood-agar, left open to air according to the scheme 1/1/1) (21,72)

| Place | Total microbial count (CFU/m².h) | | |
|---|---|---|---|
| | Optimal | Acceptable | Not acceptable |
| Medical wards | 0–450 | 451–750 | >751 |
| Surgery | 0–250 | 251–450 | >451 |
| Pharmacy | 0–100 | 101–180 | >181 |
| Aseptic room | 0–50 | 51–90 | >91 |
| Operating theatre (at rest) | 0–4 | 5–8 | >9 |
| Operating theatre (in activity) | 0–60 | 61–90 | >91 |

As shown in Figure S1 (a, b, c, and d) (see Supplementary file 1), the linear regression analysis demonstrated a strong correlation between bacterial bioaerosols and the concentration of both $PM_{2.5}$ and $PM_{10}$. However, the linear regression analysis between fungal bioaerosols and the concentration of $PM_{2.5}$ and $PM_{10}$ indicated a weaker relationship. This suggests that other factors in the OR, such as the number of staff, coughs and sneezes of patients, etc., may influence fungal bioaerosols. The results indicated that PM may have a greater contribution to bacterial bioaerosols compared to fungal bioaerosols, as bacterial bioaerosols exhibited higher regression coefficients and steeper regression line slopes for both $PM_{2.5}$ and $PM_{10}$.

This is consistent with the findings of Mirhoseini et al (80), who reported a significant relationship between 1–5 μm particles and the density of bacterial bioaerosols in the surgical and intensive care unit (ICU) departments. However, some studies have reported conflicting results. For instance, Yousefzadeh et al (81) found no significant relationship between the number of particles and the number of bacteria, while Adhikari et al (82) showed a relationship between PM concentration and the concentration of fungal bioaerosols. Palladino et al (83) detailed examination of the air quality led them to theorize that $PM_{10}$ contains bacterial bioaerosols. To date, the precise PM size that correlates with microbiological numbers has not been agreed upon (84). The study of Nikpey et al (85), in connection with the assessment of indoor air quality in different departments of a hospital in Qazvin, showed that there is no significant relationship between $PM_{2.5}$ and microbial pollution.

### *The strengths and limitations of the study and future studies' directions*

The present study contributes to the understanding of the effects of airborne pathogens on the health of patients and staff, aligning with the key principles of care centers. We have conducted thorough microbial monitoring, providing valuable insights into OR quality standards. The study emphasizes the importance of periodic microbial monitoring and the need to ensure the proper functioning of ventilation systems for maintaining optimal air quality in the OR.

**Table 6.** Comparison of the PM concentration (μg/m$^3$) measured in this study with those reported in different studies

| Study | Hospital Unit | $PM_{2.5}$ | | $PM_{10}$ | | Reference |
|---|---|---|---|---|---|---|
| | | Min-Max | Mean | Min-Max | Mean | |
| This study | OR | 15–24 | 17.81 | 23–34 | 27.02 | - |
| AlRayess et al (2022) | ICU | 10–54 | 30 | 10–65 | 33 | (25) |
| Slezakova et al (2012) | Radiology ward | 10.5–41.9 | 23.4 | 13–58.8 | 30.8 | (76) |
| Baurès et al (2018) | Seven hospital locations | 0–45.4 | 0.6 | - | 12 | (77) |
| Powell et al (2015) | - | 15–122 | 51.5 | 28–186 | 61.3 | (78) |
| Jung et al (2015) | Nurse station | - | 10.3 | - | 18.3 | (79) |

The study was based on a specific timeframe and location, which may limit the generalizability of the findings to other settings. The sample size used in the study, while sufficient for initial analysis, could be expanded in future studies to obtain more reliable correlation percentages and further reduce the margin of error. As with any research, there may be inherent limitations in the methodology employed, and we have acknowledged these potential constraints.

For future studies, we suggest the following directions; (i) Conducting further research to evaluate the impact of appropriate and continuous disinfection measures on improving air quality in the OR. (ii) Exploring the implementation of positive pressure inside the room to prevent the entry of exhaust air into the OR. (iii) Adopting larger sample sizes in future studies to enhance the reliability and precision of correlation percentages.

## Conclusion

The present study aimed to assess airborne bioaerosols and particulate matter ($PM_{2.5}$ and $PM_{10}$) in a specific LTOR. The passive method employed for measuring airborne bioaerosols proved to be effective in controlling LTOR pollution as it detected pathogens present on surgical instruments. Among the bacterial bioaerosols, *S. aureus* was found to be the most abundant bioaerosol. As for fungal bioaerosols, the most common genera isolated from the LTOR air were *Aspergillus*, *Cladosporium*, and *Penicillium*. The concentration and density of airborne bioaerosols in the LTOR exhibited a similar pattern to previous studies conducted by researchers in different hospitals worldwide. Our findings suggest that particles have a greater impact on bacterial bioaerosols compared to fungal bioaerosols. Additionally, temperature and RH did not significantly affect the detected microbial loads during the sampling procedure. While some studies indicate that ORs have the lowest pollution levels among hospital departments, our results showed that although the concentration of $PM_{2.5}$ and $PM_{10}$ remained within national air quality standards, the concentration of bacterial and fungal bioaerosols during all sampling stages exceeded the relevant standards. Evaluating the effects of airborne pathogens on the health of patients and staff is crucial in healthcare facilities, and our study provides valuable insights into the quality standards of ORs.

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## Competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Ethical issues

Not applicable.

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## Supplementary Files

Supplementary file 1 contains Figure S1.

## References


1. Lai KM, Nasir ZA, Taylor J. Bioaerosols and hospital infections. In: Colbeck I, Lazaridis M, eds. Aerosol Science: Technology and Applications. John Wiley & Sons; 2013. p. 271-89.
2. Ghasemian A, Khodaparast S, Savaheli Moghadam F, Nojoomi F, Rajabi Vardanjani H. Types and levels of bioaerosols in healthcare and community indoor settings in Iran. Avicenna J Clin Microbiol Infect. 2017;4(1):41036-. doi: 10.17795/ajcmi-41036.
3. Perrino C, Marcovecchio F. A new method for assessing the contribution of Primary Biological Atmospheric Particles to the mass concentration of the atmospheric aerosol. Environ Int. 2016;87:108-15. doi: 10.1016/j.envint.2015.11.015.
4. Eduard W, Heederik D, Duchaine C, Green BJ. Bioaerosol exposure assessment in the workplace: the past, present and recent advances. J Environ Monit. 2012;14(2):334-9. doi: 10.1039/c2em10717a.
5. Perrone MG, Gualtieri M, Ferrero L, Lo Porto C, Udisti R, Bolzacchini E, et al. Seasonal variations in chemical composition and in vitro biological effects of fine PM from Milan. Chemosphere. 2010;78(11):1368-77. doi: 10.1016/j.chemosphere.2009.12.071.
6. Abera A, Tilahun M, Tekele SG, Belete MA. Prevalence, antimicrobial susceptibility patterns, and risk factors associated with enterococci among pediatric patients at Dessie Referral Hospital, Northeastern Ethiopia. Biomed Res Int. 2021;2021:5549847. doi: 10.1155/2021/5549847.
7. Lin T, Zargar OA, Lin KY, Juiña O, Sabusap DL, Hu SC, et al. An experimental study of the flow characteristics and velocity fields in an operating room with laminar airflow ventilation. J Build Eng. 2020;29:101184. doi: 10.1016/j.jobe.2020.101184.
8. Stauning MA, Bediako-Bowan A, Bjerrum S, Andersen LP, Andreu-Sánchez S, Labi AK, et al. Genetic relationship between bacteria isolated from intraoperative air samples and surgical site infections at a major teaching hospital in Ghana. J Hosp Infect. 2020;104(3):309-20. doi: 10.1016/j.jhin.2019.11.007.
9. Allegranzi B, Zayed B, Bischoff P, Kubilay NZ, de Jonge S, de Vries F, et al. New WHO recommendations on intraoperative and postoperative measures for surgical site infection prevention: an evidence-based global perspective. Lancet Infect Dis. 2016;16(12):e288-303. doi: 10.1016/s1473-3099(16)30402-9.
10. Dexter F, Ledolter J, Epstein RH, Loftus RW. Importance of operating room case scheduling on analyses of observed reductions in surgical site infections from the purchase and installation of capital equipment in operating rooms. Am J Infect Control. 2020;48(5):566-72. doi: 10.1016/j.ajic.2019.08.017.
11. Elmously A, Gray KD, Michelassi F, Afaneh C, Kluger MD, Salemi A, et al. Operating room attire policy and healthcare cost: favoring evidence over action for prevention of surgical site infections. J Am Coll Surg. 2019;228(1):98-106. doi: 10.1016/j.jamcollsurg.2018.06.010.
12. Gola M, Settimo G, Capolongo S. Indoor air quality in inpatient environments: a systematic review on factors that influence chemical pollution in inpatient wards. J Healthc Eng. 2019;2019:8358306. doi: 10.1155/2019/8358306.
13. Shahi Zavieh F, Mohammadi MJ, Vosoughi M, Abazari M, Raesee E, Fazlzadeh M, et al. Assessment of types of bacterial bio-aerosols and concentrations in the indoor air of gyms. Environ Geochem Health. 2021;43(5):2165-73. doi: 10.1007/s10653-020-00774-1.
14. Sadeghian P, Wang C, Duwig C, Sadrizadeh S. Impact of surgical lamp design on the risk of surgical site infections in operating rooms with mixing and unidirectional airflow ventilation: a numerical study. J Build Eng. 2020;31:101423. doi: 10.1016/j.jobe.2020.101423.
15. Sadrizadeh S, Pantelic J, Sherman M, Clark J, Abouali O. Airborne particle dispersion to an operating room environment during sliding and hinged door opening. J Infect Public Health. 2018;11(5):631-5. doi: 10.1016/j.jiph.2018.02.007.
16. Figuerola-Tejerina A, Hernández-Aceituno A, Alemán-Vega G, Orille-García C, Ruiz-Álvarez M, Sandoval-Insausti H. Developing a faster way to identify biocontamination in the air of controlled environment rooms with HEPA filters: airborne particle counting. Sci Rep. 2020;10(1):2575. doi: 10.1038/s41598-020-59367-8.
17. Haleem Khan AA, Mohan Karuppayil S. Fungal pollution of indoor environments and its management. Saudi J Biol Sci. 2012;19(4):405-26. doi: 10.1016/j.sjbs.2012.06.002.
18. ISO B. 14698-1:2003 Cleanrooms and Associated Controlled Environments-Biocontamination Control. London: British Standards Institution; 2003.
19. Cao C, Jiang W, Wang B, Fang J, Lang J, Tian G, et al. Inhalable microorganisms in Beijing's PM2.5 and PM10 pollutants during a severe smog event. Environ Sci Technol. 2014;48(3):1499-507. doi: 10.1021/es4048472.
20. Haig CW, Mackay WG, Walker JT, Williams C. Bioaerosol sampling: sampling mechanisms, bioefficiency and field studies. J Hosp Infect. 2016;94(1):104. doi: 10.1016/j.jhin.2016.03.021.
21. Pasquarella C, Pitzurra O, Savino A. The index of microbial air contamination. J Hosp Infect. 2000;46(4):241-56. doi: 10.1053/jhin.2000.0820.
22. Wu B, Qi C, Wang L, Yang W, Zhou D, Wang M, et al. Detection of microbial aerosols in hospital wards and molecular identification and dissemination of drug resistance of *Escherichia coli*. Environ Int. 2020;137:105479. doi: 10.1016/j.envint.2020.105479.
23. Palulun P, Rasita YD, Massi MN, Sjahril R, Katu S, Pattelongi I. Identification of airborne aerobic bacteria in the intensive care room using MALDI-TOF MS. J Environ Health. 2024;16(1):68-75. doi: 10.20473/jkl.v16i1.2024.68-75.
24. Vahidmoghadam R, Mirzaei N, Mousavi G, Nazari-Alam A, Nazeri M, Gholipour S, et al. Determining the concentration of particulate matters and microbiological quality of indoor air in intensive care units of Kashan hospital, Iran. J Environ Health Sustain Dev. 2023;8(2):1975-87. doi: 10.18502/jehsd.v8i2.13045.
25. AlRayess S, Sleiman A, Alameddine I, Abou Fayad A, Matar GM, El-Fadel M. Airborne bacterial and PM characterization in intensive care units: correlations with physical control parameters. Air Qual Atmos Health. 2022;15(10):1869-80. doi: 10.1007/s11869-022-01222-y.

26. Bolookat F, Hassanvand MS, Faridi S, Hadei M, Rahmatinia M, Alimohammadi M. Assessment of bioaerosol particle characteristics at different hospital wards and operating theaters: a case study in Tehran. MethodsX. 2018;5:1588-96. doi: 10.1016/j.mex.2018.11.021.
27. Moon KW, Huh EH, Jeong HC. Seasonal evaluation of bioaerosols from indoor air of residential apartments within the metropolitan area in South Korea. Environ Monit Assess. 2014;186(4):2111-20. doi: 10.1007/s10661-013-3521-8.
28. Pasquarella C, Veronesi L, Castiglia P, Liguori G, Montagna MT, Napoli C, et al. Italian multicentre study on microbial environmental contamination in dental clinics: a pilot study. Sci Total Environ. 2010;408(19):4045-51. doi: 10.1016/j.scitotenv.2010.05.010.
29. Murray PR, Rosenthal KS, Pfaller MA. Medical Microbiology E-Book. Elsevier Health Sciences; 2020.
30. Polednik B. Aerosol and bioaerosol particles in a dental office. Environ Res. 2014;134:405-9. doi: 10.1016/j.envres.2014.06.027.
31. Cabo Verde S, Almeida SM, Matos J, Guerreiro D, Meneses M, Faria T, et al. Microbiological assessment of indoor air quality at different hospital sites. Res Microbiol. 2015;166(7):557-63. doi: 10.1016/j.resmic.2015.03.004.
32. Faridi S, Hassanvand MS, Naddafi K, Yunesian M, Nabizadeh R, Sowlat MH, et al. Indoor/outdoor relationships of bioaerosol concentrations in a retirement home and a school dormitory. Environ Sci Pollut Res Int. 2015;22(11):8190-200. doi: 10.1007/s11356-014-3944-y.
33. Napoli C, Marcotrigiano V, Montagna MT. Air sampling procedures to evaluate microbial contamination: a comparison between active and passive methods in operating theatres. BMC Public Health. 2012;12:594. doi: 10.1186/1471-2458-12-594.
34. Jafari MJ, Hajgholami MR, Omidi L, Jafari M, Tabarsi P, Salehpour S, et al. Effect of ventilation on occupational exposure to airborne biological contaminants in an isolation room. Tanaffos. 2015;14(2):141-8.
35. Ministry of Health Malaysia. Policies and Procedures on Infection Control. Malaysia. 2th ed. Ministry of Health Malaysia; 2010. Available from: https://www.moh.gov.my/moh/images/gallery/Polisi/infection_control.pdf
36. Sehulster L, Chinn RY. Guidelines for environmental infection control in health-care facilities. Recommendations of CDC and the Healthcare Infection Control Practices Advisory Committee (HICPAC). MMWR Recomm Rep. 2003;52(RR-10):1-42.
37. Bruce N, Ouellet C, Suh K, Roth V. Does high humidity in the operating room (OR) impact surgical site infection (SSI) rates? Am J Infect Control. 2007;35(5):E191. doi: 10.1016/j.ajic.2007.04.212.
38. Hwang SH, Park DU, Ha KC, Cho HW, Yoon CS. Airborne bacteria concentrations and related factors at university laboratories, hospital diagnostic laboratories and a biowaste site. J Clin Pathol. 2011;64(3):261-4. doi: 10.1136/jcp.2010.084764.
39. Hansen D, Blahout B, Benner D, Popp W. Environmental sampling of particulate matter and fungal spores during demolition of a building on a hospital area. J Hosp Infect. 2008;70(3):259-64. doi: 10.1016/j.jhin.2008.07.010.
40. NYC Department of Health and Mental Hygiene (DOHMH). Guidelines on Assessment and Remediation of Fungi in Indoor Environments. United States: DOHMH; 2008.
41. de Carvalho RL, Campos CC, de Castro Franco LM, De Mattia Rocha A, Ercole FF. Incidence and risk factors for surgical site infection in general surgeries. Rev Lat Am Enfermagem. 2017;25:e2848. doi: 10.1590/1518-8345.1502.2848.
42. Jentzsch T, Kutschke L, Zingg PO, Farshad M. Operating room architecture is not a risk factor for surgical site infections. Sci Rep. 2021;11(1):13391. doi: 10.1038/s41598-021-90574-z.
43. Ekhaise FO, Isitor EE, Idehen O, Emoghene AO. Airborne microflora in the atmosphere of an hospital environment of University of Benin Teaching Hospital (UBTH), Benin city, Nigeria. World J Agric Sci. 2010;6(2):166-70.
44. Hedayati MT, Mayahi S, Denning DW. A study on *Aspergillus* species in houses of asthmatic patients from Sari city, Iran and a brief review of the health effects of exposure to indoor *Aspergillus*. Environ Monit Assess. 2010;168(1-4):481-7. doi: 10.1007/s10661-009-1128-x.
45. Tang CS, Wan GH. Air quality monitoring of the post-operative recovery room and locations surrounding operating theaters in a medical center in Taiwan. PLoS One. 2013;8(4):e61093. doi: 10.1371/journal.pone.0061093.
46. Goudarzi G, Shirmardi M, Khodarahmi F, Hashemi-Shahraki A, Alavi N, Ahmadi Ankali K, et al. Particulate matter and bacteria characteristics of the Middle East Dust (MED) storms over Ahvaz, Iran. Aerobiologia. 2014;30(4):345-56. doi: 10.1007/s10453-014-9333-7.
47. Faridi S, Naddafi K, Kashani H, Nabizadeh R, Alimohammadi M, Momeniha F, et al. Bioaerosol exposure and circulating biomarkers in a panel of elderly subjects and healthy young adults. Sci Total Environ. 2017;593-594:380-9. doi: 10.1016/j.scitotenv.2017.03.186.
48. Azimi F, Naddafi K, Nabizadeh R, Hassanvand MS, Alimohammadi M, Afhami S, et al. Fungal air quality in hospital rooms: a case study in Tehran, Iran. J Environ Health Sci Eng. 2013;11(1):30. doi: 10.1186/2052-336x-11-30.
49. Rendon RV, Garcia BC, Vital PG. Assessment of airborne bacteria in selected occupational environments in Quezon city, Philippines. Arch Environ Occup Health. 2017;72(3):178-83. doi: 10.1080/19338244.2016.1192981.
50. Aliyu AS, Badawi AH, Umar NY, Abubakar F, Sani BB. Epidemiological study on hospital acquired infections and infection prevention and control among health care workers in Specialist Hospital Bauchi state, Nigeria. ARC J Public Health Community Med. 2020;5(3):1-13. doi: 10.20431/2456-0596.0503001.
51. Wan GH, Chung FF, Tang CS. Long-term surveillance of air quality in medical center operating rooms. Am J Infect Control. 2011;39(4):302-8. doi: 10.1016/j.ajic.2010.07.006.
52. Shiaka GP, Yakubu SE. Comparative analysis of airborne microbial concentrations in the indoor environment of two selected clinical laboratories. IOSR J Pharm Biol Sci. 2013;8(4):13-9. doi: 10.9790/3008-0841319.
53. Davoodian P, Karmostaji A, Vaeghi Z. Study of nosocomial infection and pattern of antibiotic resistance in Shahid Mohamadi hospital of Bandarabas. Hormozgan Med J. 2001;5(3):14-7. [Persian].
54. Bogomolova E, Kirtsideli I. Airborne fungi in four stations of the St. Petersburg Underground railway system. Int Biodeterior Biodegradation. 2009;63(2):156-60. doi: 10.1016/j.ibiod.2008.05.008.

55. Kim KY, Kim HT, Kim D, Nakajima J, Higuchi T. Distribution characteristics of airborne bacteria and fungi in the feedstuff-manufacturing factories. J Hazard Mater. 2009;169(1-3):1054-60. doi: 10.1016/j.jhazmat.2009.04.059.
56. Morris G, Kokki MH, Anderson K, Richardson MD. Sampling of *Aspergillus* spores in air. J Hosp Infect. 2000;44(2):81-92. doi: 10.1053/jhin.1999.0688.
57. Sudharsanam S, Swaminathan S, Ramalingam A, Thangavel G, Annamalai R, Steinberg R, et al. Characterization of indoor bioaerosols from a hospital ward in a tropical setting. Afr Health Sci. 2012;12(2):217-25. doi: 10.4314/ahs.v12i2.22.
58. Warris A, Voss A, Verweij PE. Hospital sources of *Aspergillus*: new routes of transmission? Rev Iberoam Micol. 2001;18(4):156-62.
59. Mahdavi Omran S, Sheidfar M. A survey of the mycological flour contamination in Babol hospitals. Med J Tabriz Univ Med Sci Health Serv. 2000;48:42-5. [Persian].
60. Hashemi J, Sharhani M. A survey comparative saprophytes fungal existent indoor and equipments research center for blood and incology and clinical patients examples for trans-plant patient in Shariati hospital in Tehran. Tehran Univ Med J. 2002;62(3):175-9. [Persian].
61. Panagopoulou P, Filioti J, Petrikkos G, Giakouppi P, Anatoliotaki M, Farmaki E, et al. Environmental surveillance of filamentous fungi in three tertiary care hospitals in Greece. J Hosp Infect. 2002;52(3):185-91. doi: 10.1053/jhin.2002.1298.
62. Ministry of Health Malaysia. Policies and Procedures on Infection Control. 3rd ed. Ministry of Health Malaysia; 2019. Available from https://www.moh.gov.my/moh/press_releases/KKM%20Policies%20&%20Procedures%20on%20Infection%20Prevention%20and%20Control%202019.pdf
63. World Health Organization (WHO). Environmental Monitoring of Cleanrooms in Vaccine Manufacturing Facilities, Points to Consider for Manufacturers of Human Vaccines. Geneva, Switzerland: WHO; 2012. p. 1-37.
64. Sarıca S, Asan A, Otkun MT, Ture M. Monitoring indoor airborne fungi and bacteria in the different areas of Trakya University Hospital, Edirne, Turkey. Indoor Built Environ. 2002;11(5):285-92.
65. Chen L, Song Z, Zhou X, Yang G, Yu G. Pathogenic bacteria and fungi in bioaerosols from specialized hospitals in Shandong province, East China. Environ Pollut. 2024;341:122922. doi: 10.1016/j.envpol.2023.122922.
66. Obaid WA. Assessment of air quality containing fungi in Al-Nu'man Teaching Hospital. Journal of Biotechnology Research Center. 2024;18(1):11-8. doi: 10.24126/jobrc.2024.18.1.706.
67. Ye F, Shi D, Meng F, Liu L, Lin M, Shi G. Distribution Characteristics and Analysis of Fungal Aerosol Concentration and Particle Size in Air-Conditioned Wards in Wuhan. Available from: https://ssrn.com/abstract=4812341.
68. Montazeri A, Zandi H, Teymouri F, Soltanianzadeh Z, Jambarsang S, Mokhtari M. Microbiological analysis of bacterial and fungal bioaerosols from burn hospital of Yazd (Iran) in 2019. J Environ Health Sci Eng. 2020;18(2):1121-30. doi: 10.1007/s40201-020-00531-7.
69. Choobineh A, Rostami R, Tabatabai R. Type and density of the air Byvayrvsl training to selected hospitals of Shiraz University of Medical Sciences in 2008. Labour's Health Journal. 2009;6(2):69-76.
70. Dehdashti A, Sahranavard N, Rostami R, Barkhordari A, Banaee Rizi Z. Review types and concentration of bioaerosols in air Damghan city hospitals. Occupational Medicine Quarterly Journal. 2012;4(3):41-51. [Persian].
71. Mousavi MS, Hadei M, Majlesi M, Hopke PK, Yarahmadi M, Emam B, et al. Investigating the effect of several factors on concentrations of bioaerosols in a well-ventilated hospital environment. Environ Monit Assess. 2019;191(7):407. doi: 10.1007/s10661-019-7559-0.
72. Fischer G, Fodré S, Nehéz M. Das ergebnis der untersuchungen zur feststellung von gesamtkeimzahl-grenzwerten in der luft von operationssraeumen. Z Ges Hyg Grenzgeb. 1972;18(10):729-33.
73. Shokri S, Nikpey A, Safari Varyani A. Evaluation of hospital wards indoor air quality: the particles concentration. J Air Pollut Health. 2016;1(3):205-14.
74. Rezaei S, Naddafi K, Jabbari H, Yonesian M, Jamshidi A, Sadat A, et al. Relationship between the particulate matter concentrations in the indoor and ambient air of the Tehran children hospital in 2007. Iran J Health Environ. 2013;6(1):103-12. [Persian].
75. Basiri H, Godini H, Omidi-Khaniabadi Y, Sepahvand A. Study of indoor and ambient air fungal bioaerosols and its relation with particulate matters in a hospital of Khorramabad. Yafteh. 2016;17(4):25-34. [Persian].
76. Slezakova K, Alvim-Ferraz Mda C, Pereira Mdo C. Elemental characterization of indoor breathable particles at a Portuguese urban hospital. J Toxicol Environ Health A. 2012;75(13-15):909-19. doi: 10.1080/15287394.2012.690707.
77. Baurès E, Blanchard O, Mercier F, Surget E, Le Cann P, Rivier A, et al. Indoor air quality in two French hospitals: measurement of chemical and microbiological contaminants. Sci Total Environ. 2018;642:168-79. doi: 10.1016/j.scitotenv.2018.06.047.
78. Powell H, Krall JR, Wang Y, Bell ML, Peng RD. Ambient coarse particulate matter and hospital admissions in the Medicare Cohort Air Pollution Study, 1999-2010. Environ Health Perspect. 2015;123(11):1152-8. doi: 10.1289/ehp.1408720.
79. Jung CC, Wu PC, Tseng CH, Su HJ. Indoor air quality varies with ventilation types and working areas in hospitals. Build Environ. 2015;85:190-5. doi: 10.1016/j.buildenv.2014.11.026.
80. Mirhoseini SH, Nikaeen M, Khanahmd H, Hatamzadeh M, Hassanzadeh A. Monitoring of airborne bacteria and aerosols in different wards of hospitals - particle counting usefulness in investigation of airborne bacteria. Ann Agric Environ Med. 2015;22(4):670-3. doi: 10.5604/12321966.1185772.
81. Yousefzadeh A, Maleki A, Dehestani Athar S, Darvishi E, Ahmadi M, Mohammadi E, et al. Evaluation of bio-aerosols type, density, and modeling of dispersion in inside and outside of different wards of educational hospital. Environ Sci Pollut Res Int. 2022;29(10):14143-57. doi: 10.1007/s11356-021-16733-x.
82. Adhikari A, Reponen T, Grinshpun SA, Martuzevicius D, LeMasters G. Correlation of ambient inhalable bioaerosols with particulate matter and ozone: a two-year study. Environ Pollut. 2006;140(1):16-28. doi: 10.1016/j.envpol.2005.07.004.
83. Palladino G, Morozzi P, Biagi E, Brattich E, Turroni S, Rampelli S, et al. Particulate matter emission sources and

meteorological parameters combine to shape the airborne bacteria communities in the Ligurian coast, Italy. Sci Rep. 2021;11(1):175. doi: 10.1038/s41598-020-80642-1.

84. Tan H, Wong KY, Nyakuma BB, Kamar HM, Chong WT, Wong SL, et al. Systematic study on the relationship between particulate matter and microbial counts in hospital operating rooms. Environ Sci Pollut Res Int. 2022;29(5):6710-21. doi: 10.1007/s11356-021-16171-9.
85. Nikpey A, Choubdar M, Dastamouz A, Rahmani M. Evaluation of indoor air quality in different hospital wards by bioaerosol sampling and particle counting in 2016. J Occup Hyg Eng. 2018;5(1):53-60. [Persian].