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Effect of equipment layout on bioaerosol temporal-spatial distribution and deposition in one BSL-3 laboratory

Zhijian Liu a,*, Wenbin Zhuang a, Xiaqi Hu b, Zhiheng Zhao b, Rui Rong a, Wenjun Ding b, Jinsong Li c, Na Li c

a Department of Power Engineering, North China Electric Power University, Baoding, Hebei, 071003, PR China
b Laboratory of Environment and Health, College of Life Sciences, University of Chinese Academy of Sciences, No.19A Yuquan Road, Beijing, 100049, China
c State Key Laboratory of Pathogen and Biosecurity, National Engineering Research Center of Biological Protective Equipment, Institute of Microbiology and Epidemiology, Academy of Military Medical Sciences, 100071, Beijing, China

A R T I C L E   I N F O

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- Deposition pollution

A B S T R A C T

Reasonable equipment layout is essential for creating a healthy and safe environment, especially in a three-level biosafety laboratory with high potential risk factors of infection. Since 2019, COVID-19, an emerging infection has swept the world and caused severe losses. Biosafety laboratories are mandatory sites for detecting high-risk viruses, so related research is urgently needed to prevent further laboratory-acquired infections of operators. This study investigated the effects of obstacles on exposure infection of staff in a biosafety laboratory with related experimental equipment. The numerical simulation results are highly verified by the measured results. The results indicate that although the equipment layout does not affect the bioaerosol removal time, nearly 17% of the pollutant particles in the actual laboratory cannot be discharged effectively compared with the ideal situation. These particles lingered in the lower space under the influence of vortex, which would increase the respiratory risk of operators. In addition, after the experiment a large part of bioaerosol particles would be captured by equipment and floor, and the deposition rate per unit area is 0.45%/m2 and 0.8%/m2, respectively. Although the results show that the equipment layout could reduce the pollution on the floor, the disinfection is still an important link, especially on the surfaces of equipment. Meanwhile, the result also indicates that the action should be light and slow when operating in BSL-3 laboratory, so as to avoid the secondary suspension pollution of bioaerosol particles on the equipment surface and floor.

1. Introduction

After experiencing a continuous high death of infectious disease in the past decades, worldwide attention to the risk transmitted by air medium has become increasingly prominent [1–5]. According to the Global Tuberculosis Report 2018 of World Health Organization (WHO), as a typical airborne infectious disease, tuberculosis (TB) was estimated to cause 10 million people infected and 1.5 million died per year [6]. In addition, one emerging infection COVID-19 has spread all over the world, resulting millions of infections and hundreds of thousands of deaths. More and more events show that bioaerosol transmission is of great significance to the infection of many infectious diseases. Therefore, in order to deal with highly pathogenic microbes and provide a relatively safe environment for their experiments, a large number of biosafety laboratories (BSLs) in the world have been invested [7]. However, due to inappropriate equipment layout and operation conditions, some places with high potential risk of infection are more likely to develop laboratory-acquired infections, such as biosafety level-3 (BSL-3) laboratory [8,9]. Hence, the concept of “biosafety” must be considered as the first priority when using BSLs to carry out the microbial experiments [10].

In recent years, many efforts by researchers were thereupon developed in terms of BSLs. A typical study to analyze relevant strategies in biosafety laboratory was performed by Zaki [11], and it summarized the management norms of air, liquid and solid waste in BSL-3 laboratory. In order to effectively assess infection-risk, Li et al. [12] investigated several risk assessment methods of BSLs in detail, determined the assessment scope and founded that risk of bioaerosol exposure was the most common risk in the experiment. Although an infection-risk assessment is very important, it is undoubtedly that CFD-based

* Corresponding author.
E-mail address: zhijianliu@ncepu.edu.cn (Z. Liu).

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method and visualization technology can provide very intuitive predictions of pathogenic bioaerosols when they dispersed in indoor environment \[13,14\]. An early study conducted by Yu et al. \[15\] started to use computational fluid dynamics (CFD) method to analyze the diffusion and suspension of virus aerosols in air and it was proved that CFD could predict the three-dimensional airflow pattern when pollutants diffuse into the room. Tang et al. \[16\] used human models and visual engineering techniques to study and observe the characteristics of human respiratory activity and enabled the movement of bacteria-carrying particles (BCPs) to be visualized and quantified on the scale of time and space. However, although biosafety laboratories have been recognized as the major place developing vaccines and testing pathogens, very few existing studies were conducted with attempt to investigate the influence of the equipment layout on infection-risk in one BSL-3 laboratory, due to the particularity and importance of its environment.

In order to quantitatively evaluate the influence of the equipment layout on the diffusion and deposition of bioaerosols, two CFD models are developed based on the experimental test of a real BSL-3 laboratory, one is actual laboratory and another is corresponding empty room. The aim of this study is to compare the bioaerosol movement processes of two models from several aspects by CFD technology. All boundary conditions and environmental parameters are based on the BSL-3 standard manual of World Health Organization \[17\], and the simulation conditions of two cases are exactly the same except for the presence or absence of equipment. The changes of bioaerosol removal rate and spatial suspension rate with time are discussed and analyzed in two cases, and the spatial-temporal distribution characteristics of high-risk regions are compared and investigated in depth. Moreover, the accumulation trends and final deposition status of contaminated surfaces in two cases are also given. The results could provide a scientific reference for optimizing the equipment layout of other similar BSL-3 laboratory, and are valuable in proposing guidelines for operation and disinfection policies for different laboratory layout.

2. Methodology

2.1. Physical model

As illustrated in Fig. 1, based on an actual BSL-3 laboratory in Beijing, a physical model of \(x \times y \times z = 8 \times 2.5 \times 4\) m is numerically constructed with two airflow inlets and two outlets located at both ends of the ceiling \[18\]. The field investigation indicates that supply air with velocity of 0.7 m/s is introduced to the room by two square diffusers (0.63 m \(\times\) 0.63 m) and discharged by two ceiling outlets (1.086 m \(\times\) 0.553 m). It also exhibits the schematics of both laboratory layouts (empty room and actual room) and monitoring points position in the CFD model. The \(X^+, X^-, Z^+, Z^-\) are introduced to simplify the names of four walls in different directions in this study, respectively. In addition, the basic facilities in the actual laboratory are equipped according to the requirements of a typical BSL-3 laboratory, including an A2 biosafety cabinet (BSC), six commonly used equipment, and two laboratory benches \[17,18\]. These devices are treated as rectangular boxes in order to simplify the numerical calculation and subsequent airflow analysis \[19\]. The laboratory layout is similar to the design manual of BSL-3 laboratory and is therefore considered reasonable. According to the field investigation of BSL-3 laboratory, the boundary conditions of inlets, outlets and other solid surfaces were defined as the velocity inlet boundary (0.7 m/s), pressure-outlet boundary and no-slip wall boundary, respectively. The description of equipment sizes and boundary conditions are shown in Table 1.

As shown in Fig. 1, the removal of bioaerosols is certainly affected by equipment. When conducting bioaerosol experiments in the laboratory, aerosols could be captured by the equipment, so increase of equipment may lead to greater risk. In order to obtain valuable results, two field experiments were conducted in May 2018 and January 2019, in which Serratia marcescens and phage \(\Phi X174\) were employed as substitutes for high-risk bioaerosols. Based on the field investigations, two numerical models of equal size were constructed. Subsequently, the relevant simulation parameters (diameter, source concentration and release rate

![Fig. 1. Schematic diagram and field measurement of increased exposure risk caused by equipment layout in one BSL-3 laboratory.](image)
of bioaerosols, etc.) were selected according to the experimental results and carefully verified. In terms of sampling points selection, the four sampling points were located near the source, the diffusion path in the main flow zone and the air outlet, respectively, so as to better analyze and avoid excessive interference with indoor airflow patterns. Four sampling points and aerosol generator locations are shown in Table 2.

The argument of this study is to investigate the influence of equipment layout on bioaerosol distribution and directional airflow operation. Therefore, in the procedure of experiments, the pollution source was placed below the inlets and the state of A2 biosafety cabinet was off-duty. It is more instructive to obtain bioaerosol-related conclusions under this adverse scenario to optimize the laboratory layout and operation strategies. In addition, the ideal situation in this study is defined as the initial airflow pattern and the diffusion distribution of bioaerosols in BSL-3 laboratory without any interference of equipment layout. Normally, there are various possibilities of equipment layout in a practical BSL-3 laboratory. Therefore, the definition of the ideal situation can provide a valuable reference standard for optimizing layout. The defined ideal situation can also be used for subsequent layout optimization of similar three-level biosafety laboratories for quantitative research.

2.2. Mathematical model and boundary conditions

All sets of grids were modelling by GAMBIT and numerical simulations were performed by ANSYS Fluent 17.0. In this study, the airflow pattern of laboratory was first assumed to be incompressible and turbulent with low speed. Therefore, the airflow-field and temperature distribution were solved by the incompressible Navier-Stokes (N-S) equation with the standard k-ε turbulence model due to its successful application in terms of airflow in enclosed environment [20,21].

In addition, in order to meet the stability requirement of numerical calculation, the second order upwind scheme is adopted and the SIMPLE algorithm is employed to calculate the airflow-field [22].

For the diffusion of bioaerosols in the laboratory, a one-way coupling Lagrangian method is employed to track spherical particles. There are several major particle driving forces to consider when calculating the motion of suspended particles [23]. Previous studies [24,25] indicated that the driving forces of small-sized particles such as pressure gradient force, Magnus force and virtual mass force are lower in magnitude than other forces and therefore not adopted in this study. Additionally, the thermophoretic force is also ignored due to the fact that environmental parameters of the laboratory are always kept isothermal. Therefore, each term of particle force in Lagrangian particle tracking model is defined in Equation (1). Significant forces including the drag force \( F_D \), the gravity \( F_G \) and the Saffman force \( F_S \) are expressed in Equations (2)–(4).

\[
\frac{d\vec{U}_p}{dt} = F_D + F_G + F_S
\]

\[
F_D = \frac{18u_d C_D Re}{\rho_p d_p^2} (u_i - u_p)
\]

\[
F_G = \frac{\pi}{6} \rho_p d_p \rho g
\]

\[
F_S = \frac{1}{2} \rho \frac{d^2}{dx/dt}(u - u_p)
\]

where \( u \) is the air molecular dynamic viscosity, \( \rho_p \) is the density of single particle, \( d_p \) is the particle diameter, \( C_D \) is the drag coefficient, \( Re \) is the particle Reynolds number, \( u_i \) and \( u_p \) are the velocity of air and particles, \( du/\partial t \) is the air velocity gradient perpendicular to the wall surface, \( u_p \) is the particle axial velocity.

In actual laboratory, the size of particles is small and the solid surfaces have high absorptivity, so it is reasonable to assume that particles are deposited immediately when hitting the solid surfaces [26,27]. In addition, when the volume fraction of particles in the environment is small, it can be approximately considered that the particles have no effect on the flow field [28]. Moreover, the diameter of bioaerosol selected in this study is less than 1 μm. According to Wei et al. [29], evaporation process in this case is rapid so that the evaporation effect is not considered in this study.

Due to various assumptions in the numerical calculation of this study, several criteria need to be strictly followed to control such uncertainty. In fact, in this study, considerable computational efforts were conducted to track a huge number of particles and repeat the process multiple times. Most importantly, the diffusion results in actual laboratory have been well verified by the field measurements.

2.3. Experimental testing and verification

Grid independence verification is the premise to ensure the accuracy of the subsequent simulation results. It has been pointed out from previous work that the grid structure and density have a serious impact on the airflow [30]. Therefore, the first step of this study is to carry out the grid independence test. In this simulation, three kinds of grids were built, and the velocity fields of typical section were compared to check their independence. The results of grid verification are shown in Fig. 2. The results show that with the increase of grid number from 2077985 to 4065482, the flow fields of the two cases are basically unchanged, so the second kind of grid is considered reasonable and selected in this study.

Besides, two experimental bioaerosols (Serratia marcescens and phage FX174) are employed to verify the concentration and the accuracy of the simulation in this study. Bioaerosol concentrations of S1, S2, S3, S4 and pollution source were measured by six-stage Andersen samplers. The results are given in Fig. 3, in which the simulated average concentration is used to better correspond to the average concentration of airborne culturable bioaerosols measured. As can be seen from Fig. 3, except of the viral experimental concentration of S2, which is different.
from the simulated value due to improper operation of the operators, other results are highly consistent. In addition, the results with a mean absolute percentage error (MAPE) of less than 10% indicate that the simulation results are basically reliable [31].

3. Results and discussion

3.1. The effect of equipment layout on removing bioaerosols in the BSL-3 laboratory

The concentration variation of bioaerosols at four sampling points in two simulations (empty room and actual room) is shown in Fig. 4. From the fitting curve, it can be seen that the spatial concentration of the sampling points in both cases decreases exponentially. However, the concentration of each sampling point fluctuated slightly up and down in the empty room because the indoor air circulation is more efficient without equipment. In addition, as time went on, it is obvious that the concentration peaks of the last three sampling points (S2, S3, S4) in empty room are earlier than that in the actual room. It indicated that the equipment layout is an important factor affecting the migration of bioaerosol particles. The indoor airflow pattern would be changed by the equipment layout, resulting in further changes in the distribution of the bioaerosol particles and the inhaling risk of the experimental personnel.

To compare the effects of space obstacles on airflow and bioaerosol diffusion in the BSL-3 laboratory, two simulations (empty room and actual room) are employed to analyze bioaerosol removal rate and suspension rate. As shown in Fig. 5(a), the trend of bioaerosol removal rate always maintain stable at about 400s, regardless of the presence of equipment obstacles in the room. This finding shows that all bioaerosols can be discharged or deposited within 400 s, which also implies the self-purification time of the laboratory. In addition, it is also noticed that the bioaerosol removing rate in the empty room is up to 46.4%, while that in the actual room accounted for only 29.5%. In other words, nearly 17% of the pollutant particles would stay in the actual room with equipment obstacles. Further analysis shows that these staying bioaerosol particles were eventually captured by building envelope or equipment, which is much higher than that of empty room without equipment. These deposited bioaerosol particles would remain bioactive for a short period of time, resulting in a significant increase in the potential exposure risk of the operators. Therefore, the surface disinfection in the laboratory is worthy to be attached attentions during the experiment process.

On the other hand, from the slope of the curve in Fig. 5(a), it can be
seen that the removal speed in the first 200 s of the empty room is higher. Therefore, a conclusion is that the particles carrying capacity of the airflow is obviously better than that of the actual room. The bioaerosol particles are discharged from the room more rapidly and orderly in empty room, which suggests the effectiveness of airflow to some extent. Compared with the research on the discharge of indoor pollutant particles by optimizing air distribution, the influence of equipment layout on bioaerosol particles was rarely studied qualitatively. Therefore, the comprehensive analysis of indoor airflow and particles distribution characteristics in BSL-3 laboratory is of great value in this scenario. The relevant rules can provide reference for the equipment layout of biosafety laboratory.

By comparing the spatial suspension rate of bioaerosols in two cases, the temporal variation rule of suspended bioaerosols can be obtained. As shown in Fig. 5(b), during the whole experiment period of 0–800 s, the number of suspended bioaerosols in the empty room is always smaller than that in the actual room. It is observed that the suspension rate of the actual room is about 14.4% higher than that of the empty room when the bioaerosol has not yet been discharged from the exhaust outlets (T = 20 s). However, as time went on, there were more and more bioaerosols deposited in the actual room (only 2.1% higher at T = 120 s) and eventually 2.3% higher than that in the empty room (T = 800 s). This finding shows that the bioaerosols trapped by equipment will first increase with time and then tend to level off. Similar conclusions are obtained in subsequent deposition analysis. The reason for the high suspension rate of actual room in the early stage (T < 20 s) was mainly due to the lack of capture and interception of obstacles. At this time, most of bioaerosols circulated in the room was carried by airflow. With the passage of time, the suspended bioaerosols in the actual room were gradually absorbed and deposited on the surface of the equipment, resulting in a reduction in suspension. Nevertheless, the regions near the experimental equipment were also high-risk regions for laboratory personnel to carry out related operations during experimental procedure (T > 50 s), so we must be vigilant.

3.2. The effect of equipment layout on high-concentration region distribution of bioaerosols in the BSL-3 laboratory

The spatial trajectories of bioaerosol particles were compared and discussed in order to visually reflect the diffusion differences caused by the layout of equipment. Fig. 6 shows the particles transport trajectory in the cases with and without equipment layout at three moments: the initial release (T = 5 s), during the diffusion process (T = 21 s) and after relatively long diffusion time (T = 30 s). Obviously, the migration and distribution of airflow and bioaerosol particles in two cases are observed to be inconsistent. Results show that in both cases, the transport of bioaerosol particles first accorded with the trajectory of airflow. Bioaerosol particles released from the pollution source were rebounding after first hitting the ground, and then most of them were captured by the ground and the wall near the pollution source side. At T = 5 s, since the experiment has just begun, the particles cloud was mainly carried by strong supply airflow and thereby the difference was not obvious. When
At T = 21s, it can be clearly observed that the DPM concentration in the empty room was significantly lower than that in the actual room. Eventually, only a small amount of high concentration particles remained in the room at T = 30s. The outcomes indicate that the risk of inhaling bioaerosols in an actual biosafety laboratory would be significantly increased due to the increase in the number of equipment. In addition, the layout of the laboratory is changed by some obstacles, resulting in changes in initial airflow patterns and a decline in the ability to carry particles. It is worth noting that Fig. 6 only shows the overall migration of bioaerosols in the first 30 s, when the particles have not been discharged from the outlets. Therefore, the concentration of the high-risk bioaerosols is represented by DPM concentration, which is closely related to the respiratory risk of the laboratory staff during experiments.

To investigate the influence mechanism of equipment obstacle on the airflow form and the carrying effectiveness on bioaerosol particles, Fig. 7 shows the vortex flow regions and particles distribution of typical cross section at x = 3 m (T = 20 s and 30 s, DPM = 50000 CFU/m³). As shown in Fig. 7(a) and (c), in the laboratory without equipment obstacles, the vortex flow regions are mainly formed in the middle of the
room, which is caused by the mutual interference of supply air from two ceiling inlets. However, Fig. 7(b) and (d) shows the opposite outcome in the actual room, in which the vortex flow regions mainly be observed in the activity range of operating personnel (near the experimental equipment). Combining the removal rate analysis of two cases, the distribution of vortex flow regions is considered as an important factor affecting the discharge of bioaerosols. Therefore, arranging experimental equipment appropriately to change the position of vortex flow regions is an effective way to reduce the risk of infection in laboratory.

To further analyze the influence of the vortex flow regions on the formation of local high-concentration regions in this lab, Fig. 8 shows a comparison of two cases at $T = 40$s. First of all, it can be seen that the high-concentration regions in the empty room are significantly less than that in the actual room. It can be further concluded that the airflow of the former is more efficient to remove bioaerosols in the early stage, leading to the full diffusion of particles in the cycle. In addition, it is observed that the high-concentration regions in the room basically coincided with the vortex flow regions in the space. As a result, it can be further inferred that the flow field of BSL-3 laboratory is disturbed by the experimental equipment, and the vortex flow regions are generated accordingly. Obviously, because the bioaerosols appears and gathers in a specific place, it inevitably increases the respiratory risk of nearby experimenters.

### 3.3. The effect of equipment layout on deposition characteristics of bioaerosols in the BSL-3 laboratory

In addition to the close relationship between the spatial high concentration area and the respiratory risk of the staffs, the deposition of bioaerosol particles on the surfaces in the lab is also directly related to their contact risk. In order to analyze the influence of equipment layout on the deposition characteristics of bioaerosols, the number of particles deposited on the inner surface and unit area deposition ratio of two cases (empty room and actual room) are given in Fig. 9. The unit area deposition rate is introduced to better reflect the surface pollution degrees under the consideration of surface area. For example, 1% means that the number of particles deposited per square meter is one hundredth of the number of particles released. The mathematical model is defined by Equation (5).

![Fig. 9. Comparison of the deposition number and unit area deposition ratio of bioaerosol particles on different surfaces in two cases.](image)

**Fig. 9.** The spatial distribution of high-concentration regions in two cases (DPM Concentration = 50000 CFU/m$^3$, $T = 40$s).
where $\theta_i$ is the unit area deposition ratio, $r_i$ is the number of deposition surface, $N_i$ is the number of deposits on surface $i$, $A$ is the released number, and $S_i$ is the area of surface $i$.

From the unit area deposition ratio in Fig. 9, it can be found that the deposition distribution of bioaerosol particles in the actual room is relatively average. In the actual room, the most serious ones are floor (0.8%), laboratory equipment (0.45%) and walls (0.3%). While in the empty room, the most serious one was floor (1.03%), which is far more than the sum of other surfaces. It is our pursuit to study how this deposition difference occurs and thereby provides guidance for the exposure risk relationship between people and equipment layout. This finding shows that for the biosafety laboratory with complex equipment, the focus of surface disinfection should be transferred to the laboratory environment, which are polluted seriously and often used by staffs. On the other hand, due to the comprehensive effect of air distribution and environmental gravity, the amount of bioaerosol particles on the floor (in both empty room and actual room) also accounted for a large part.

Therefore, in one BSL-3 laboratory, the staff should keep the movement light and slow so as to avoid the secondary suspension pollution caused by the bioaerosol particles on the floor.

To understand the cause of this phenomenon, the accumulation trends of bioaerosol deposition on each surface (floor, ceiling, walls, etc.) are compared between two cases. First of all, it can be seen from Fig. 10 that the deposition trends of all surface increase rapidly at the beginning ($T < 100s$), and then become gentle. In addition, the analysis of the two models shows that except for the floor and the $Z+$ -sidewall which is near the pollution source in the empty room, the deposition number on the other surfaces are smaller than that in the actual room. Interestingly, in the initial stage of diffusion ($T < 50s$), the deposition speed of floor in the empty room decelerated even earlier (25s for empty room and 50s for actual room). On the other hand, the deposition condition of the $Z+$ wall is completely opposite to that of other walls, and its deposition in empty room is always higher than that in the actual room. Therefore, the reason for the difference of deposition rule is mainly related to the particle transport trajectory. First, in the initial stage of the experiment ($T = 10$–$20$ s), the airflow rebounding would be prevented by the equipment after the particles cloud hit the floor. As a result, the particles cloud would be diffused near the floor, and the deposition process in the actual room will be longer. Similarly, the equipment also could prevent the airflow to contact with the $Z+$ wall, resulting in a smaller number of particles on it in the actual room.

In addition to the abnormal particle deposition on the floor and $Z+$ wall, the deposition on other surfaces of the empty room are less than that of the actual room after the experiment. Most of these "disappeared" particles are captured on the surfaces of the laboratory equipment, which means more attention should be paid to the contact risk of operators in BSLs. In addition, Fig. 10 (b) also shows the deposition accumulation trend of the laboratory tables and the biosafety cabinet (BSC) in the actual room.

To sum up, in a laboratory with complicated equipment, not only the floor but also the surface of experimental equipment need disinfection. In addition, the bioaerosols deposition on the floor of the actual room might be reduced due to the hindrance of laboratory equipment. Therefore, laboratories with less equipment need to pay more attention to the slow pace to avoid secondary suspension pollution.

4. Conclusions

In this study, a comparison between a biosafety laboratory and an empty room model is provided and discussed by means of CFD simulation technology. The outcomes reveal that the space obstacles play an important role in indoor airflow and bioaerosol diffusion. The mathematical model is well verified by experiments and the main conclusions are obtained following:

1. No matter whether there are equipment obstacles in the biosafety laboratory, the removal rate of bioaerosol is stable in about 400 s. In addition, due to the influence of experimental equipment, there are nearly 17% of the pollutant particles cannot be discharged effectively compared with the ideal situation.

2. The spatial obstacles have a significant impact on the effectiveness of indoor airflow, and the removal speed of bioaerosols in the empty room is significantly higher than that in the actual room before 200 s. Further analysis indicates that the decrease of bioaerosol removal speed is mainly due to the formation of...
spatial vortex flow regions in the room, which destroyed the initial airflow pattern.

(3) By comparing the bioaerosol suspension rates of the two cases, it can be obtained that the suspended bioaerosols in the empty room is always less than that in the actual room. However, with the deposition number of particles on the equipment increase and then tend to level off, the difference of suspension rates caused by equipment obstacles will eventually decrease from 14.4% to 2.3%. Results indicate that bioaerosol is usually captured by experimental equipment during the procedure of experiment (T > 50s), and therefore the operators should be more careful when operating the equipment.

(4) The space obstacles will lead to an increase in the proportion of vortex flow regions in the laboratory. On the one hand, the vortex changes the original airflow structure of the laboratory and reduces the particle-carrying capacity and particle-removal efficiency of the airflow. On the other hand, the vortex makes the bioaerosol particles stay near the obstacle for a constant time, thus forming a high concentration residual area of pollutants in the space and increasing the respiratory risk of the operators.

(5) In a biosafety laboratory with complicated equipment, not only the floor but also the surface of experimental equipment need disinfection. In addition, the less laboratory equipment, the more attention should be paid to gentle movement and avoid secondary suspension pollution.

In summary, the effects of equipment layout on the removal, diffusion and deposition of bioaerosols are compared and analyzed in several aspects. Results show that although the equipment layout could reduce the pollution to the floor, disinfection is still an important link, especially for the surfaces of equipment. Meanwhile, when operating in a BSL-3 laboratory, the action should be gentle. The relevant conclusions can provide valuable scientific basis for the layout optimization and operation guidelines of biosafety laboratory.

Declaration of competing interest

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.

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