Study on the scavenging effect of steady-state displacement air purification system on indoor bioaerosol

Lijuan Wu[a] Zhigang Cai[a] Jihong Li[b] Jingwen Li[a] Hao Pu[c] Xianghong Liu[d] Xixin Yan[a,*]

a. Department of Respiratory Medicine, The Second Hospital of Hebei Medical University, Shijiazhuang, Hebei 050000, China
b. Department of Clinical Laboratory, The Second Hospital of Hebei Medical University, Shijiazhuang, Hebei 050000, China
c.Shanghai Bingyuan Medical Technology Co., Ltd, Shanghai, 201913, China
d.Shijiazhuang Aoxiang Pharmaceutical Engineering Co., Ltd, Shijiazhuang, Hebei 051431, China.

Corresponding author. E-mail address:xi_xin_yan@126.com

Abstract

Many kinds of microorganisms, including the new coronavirus, can spread through aerosols. There are abundant bio-aerosols in indoor medical environment, and these aerosols are likely to cause infections among medical staff and patients in hospitals. Especially in the current severe situation of the new coronavirus, the use of a steady-state displacement air purification system may have a certain effect on reducing the spread of the new coronavirus or other microorganisms. To analyze the purification effect of the steady-state displacement air purification system on the bio-aerosols in the bronchoscope room of the hospital. In this study, the bio-aerosol in the outpatient department of the Respiratory Medicine Department of the bronchoscope room were collected during different time periods from 2021.04 to 2021.05. Among them, the microorganisms contained in the bio-aerosol are identified by NGS and microorganism culture and strain identification. During the experiment, the researcher took 5 sampling points to collect aerosols, performing NGS and microbial culture counting and identification to detect microorganisms. The total purification efficiency are 88.0% (NGS) and 87.5% (microbial culture count and identification results). The results above was statistically significant. In an occupant environment in the bronchoscope room, the steady-state displacement air purification system has a good removal effect on bio-aerosols. Such purification efficiency may have a positive effect on the control of the new coronavirus epidemic and various infectious diseases.

Keywords: Bio-aerosol, Bronchoscope room, Steady-state displacement air purification system, Indoor air.

1 INTRODUCTION

Indoor bio-aerosols are effective against the known new coronaviruses (Jarvis, 2020), Mycobacterium tuberculosis, measles virus, varicella-zoster virus (Tellier et al., 2019), hand, foot and mouth disease virus (Colenutt C et al., 2016), Ebola virus and Middle East Respiratory Syndrome. The spread of established infectious pathogens such as virus has an important impact to human health (Judson et al., 2019). Indoor bio-aerosols in different medical places also have an important impact on medical staff and patients.

The test of NGS is a kind of next-generation sequencing technology, which can directly analyze the genome of microbes from environmental samples. It can provide information about the microorganisms present in environmental samples (Segata et al., 2013). Some researchers used a wet cyclone portable air sampler to collect air samples at a flow rate of 450 L min^{-1}, each with a sampling volume of 54,000 L, and used whole-genome sequencing to study the airborne microbial communities in various indoor and outdoor environments in New York City (Yooseph et al., 2013). Some researchers have also applied high-throughput sequencing technology to characterize the atmospheric sedimentary microbial communities collected in northeastern Spain for three years (Barberán et al., 2014). In the past few years, high-throughput sequencing technologies such as NGS have been used to characterize microbial communities in various environmental samples (Madsen et al., 2015). The method of microbial cultivation and colony counting is a relatively simple and low-cost method. The microorganisms in the air are collected and cultured on a semi-solid medium. The result is expressed as the number of colonies in CFU. A single colony is often formed by a single microorganism, so CFU provides information on the number of microorganisms present in the sample

(Ghosh et al., 2015). If we need to obtain information on the types of microorganisms, we can also further identify the species (Heidelberg et al., 1997).

2 METHODS

2.1 Experimental equipment and experimental materials

Steady-state replacement flow air purification system (China Aoxiang), Circulating water vacuum pump (China Lichen), 37mm filter membrane sampling clip (US SKC), Gelatin filtration Membrane (Sartorius, Germany).

2.2 Introduction to the Steady-State Displacement Flow Air Purification System

The steady-state displacement flow air purification system used in this experiment is Aoxiang® Steady-State Displacement Flow Clean and Disinfection System, which is characterized by steady-state displacement flow field and high-efficiency air filtration technology. The steady-state displacement flow air purification system uses an energy-saving and efficient air cleaning technology: steady-state displacement flow technology. In the indoor environment between the air outlet and the air inlet, a "vector flow" one-way, propelling airflow is established to remove particles and aerosols with a minimum amount of diffusion and form an air isolation barrier in the controlled area. It is a purely physical epidemic prevention technology. Reduction indoor bio-aerosol pollution may play a positive role in the prevention and control of epidemics in public places, including medical environments. Fig. 1 demonstrates the model diagram of steady-state replacement flow air purification system.

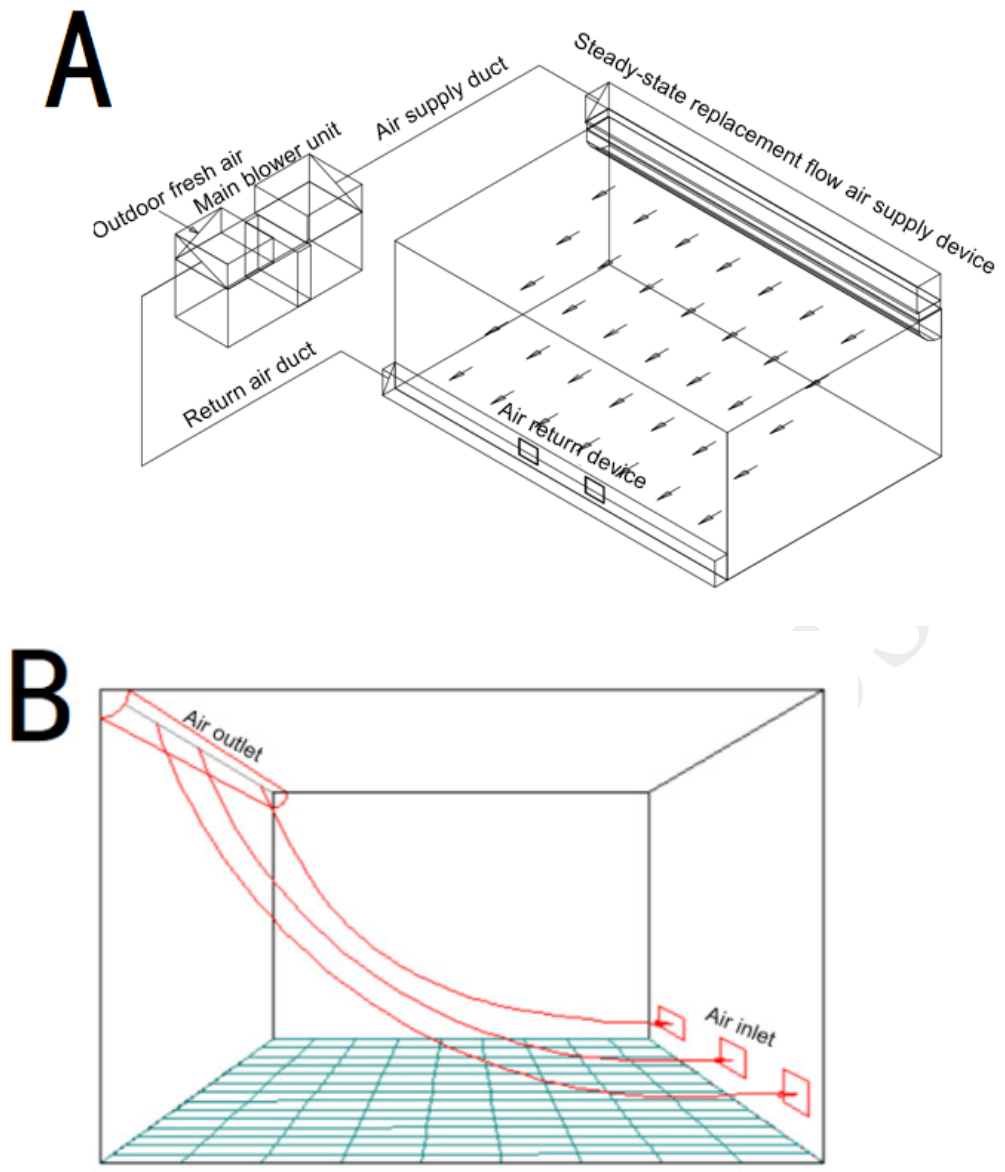

Fig. 1 Model diagram of steady-state replacement flow air purification system. Figure A: Schematic diagram of the steady-state replacement flow air supply system. Figure B: Schematic diagram of the working principle of the steady-state replacement flow system.

2.3 Grouping: This study collected bio-aerosol in the outpatient department of the bronchoscope room of the Second Hospital of Hebei Medical University during different time periods from 2021.04 to 2021.05. Among them, the microorganisms contained in the bio-aerosol are identified by NGS and microorganism culture and strain identification.

There are two groups: the unpurified group (turning off the purifier) and the purified group(turning on the purifier).

2.4 Sample collection method

Under the normal dynamic diagnosis and treatment environment of the bronchoscope room, the gelatin filter membrane is used to collect indoor bio-aerosols. The aerosols-collection began half an hour after the patient enters the bronchoscope room. The picture of sampling device is shown in Fig. 2 (B and C).

The sampling membranes of 1, 2, 3, 4, and 5 were used to sample the unpurified group for 1 hour; then turned on the purifier for 40 minutes, during which no sampling would be performed; The purifier were kept open 40 minutes later, The sampling membranes of A, B, C, D, and E were used to sample the aerosols of the purified group for 1 hour. The sampling flow chart is shown in Fig. 2 (D). The process of sampling was carried out during normal working hours, and each sampling membranes contained the total sampling of 39.5 hours. The NGS detection used is undertaken by Shanghai Bingyuan Medical Technology Co., Ltd. The method of NGS can detect 560 kinds of clinically common microorganisms including bacteria, fungi, viruses and parasites, most of which are bacteria and fungi, and 70 kinds of viruses in total are included. The method of microorganisms culture and identification are tested by the clinical laboratory of the Second Hospital of Hebei Medical University. The No. 1, 3, 4, and 5 sampling membranes were dissolved in 4ml of the NGS special specimen storage solution after the sampling process, then placed in a 37℃ water bath for 7 minutes and marked as the unpurified group. The sampled membranes A, C, D, and E were dissolved in 4ml of the NGS special

Fig. 3. Mild-to-high copy number results of NGS test: A: NGS comparison results of the unpurified group and purified group. B: Low copy number results of NGS comparison results of the unpurified group and purified group. C: The statistical results of the unpurified group and purified group.

Among them, the purification efficiencies of individual microorganisms are: Gardnerella vaginalis 62.5%, Staphylococcus capitis 100%, Micrococcus luteus 100%, Actinomyces naevitii 100%, Pantoea agglomerans 100%, purulent actinomycetes Bacteria 100%.

The researchers used SPSS statistics 26 for nonparametric tests, and the statistical results are as follows:

At the level of $\alpha = 0.05$, $P < 0.05$, the researchers can consider that there is a statistically significant difference in the copy number of NGS of different microorganisms between the unpurified group and the purified group.

3.2 Results bacterial and fungal culture and identification

3.2.1 Colony count results

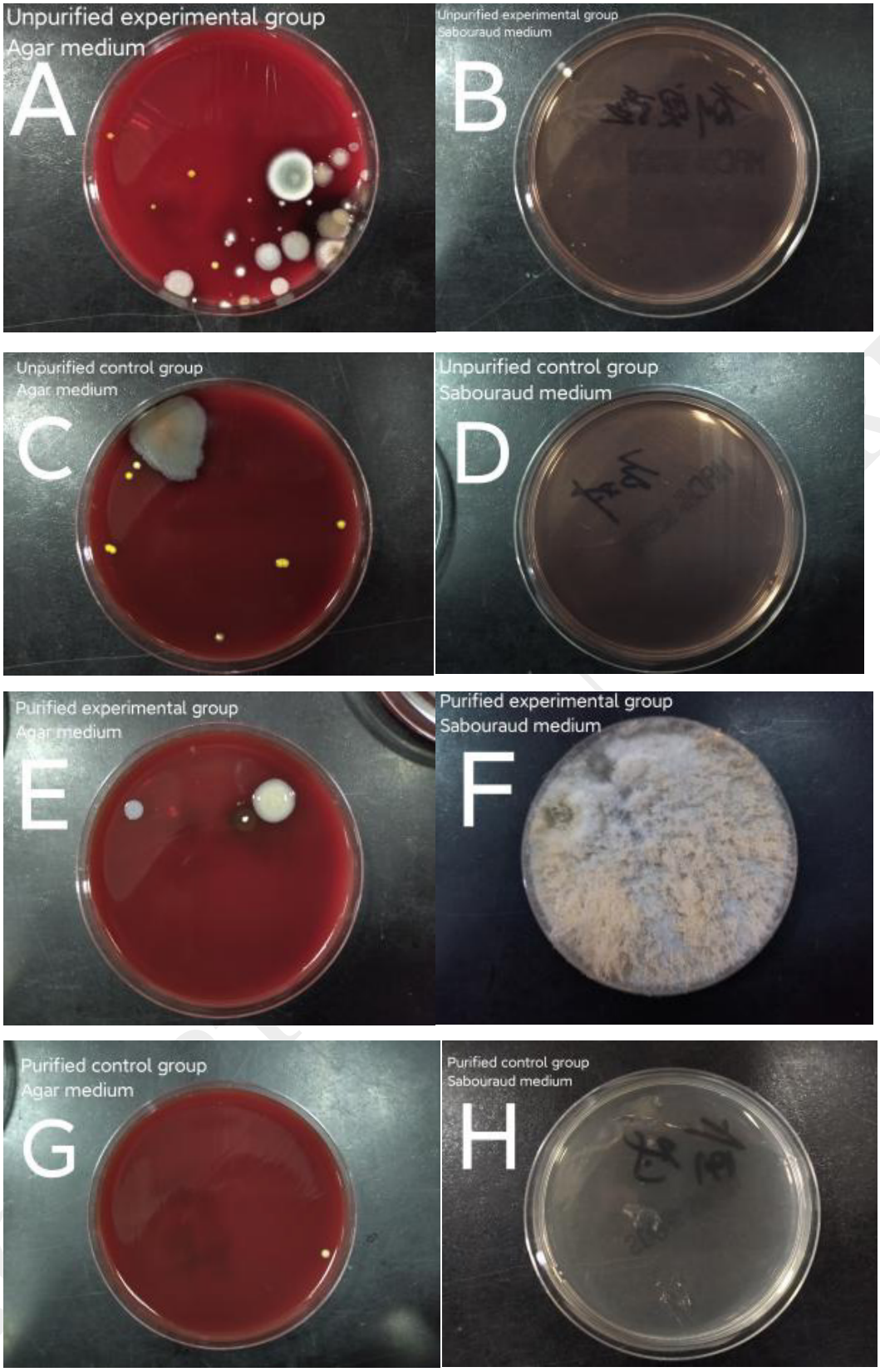

Fig. 4. Results of microbial culture and colony count.A,B: Unpurified test group. Number of colonies: 32. C,D: Unpurified control group. Number of colonies: 9. E,F: Purified test group. Number of colonies: 4. G,H: Purified control group. A,C,E,G: Agar medium. B,D,F,H: Sabouraud medium. Number of colonies: 1.

The comparison of the total colony count between the purified group and the unpurified group is as follows:

Efficiency of removing bioaerosol pollution = purification efficiency = (colony count in unpurified group - colony count in purified group) / colony count in unpurified group = 87.5%

3.2.2 Identification results of bacterial and fungal species

The researchers identified the dominant colonies in the culture medium. The result is as follows:

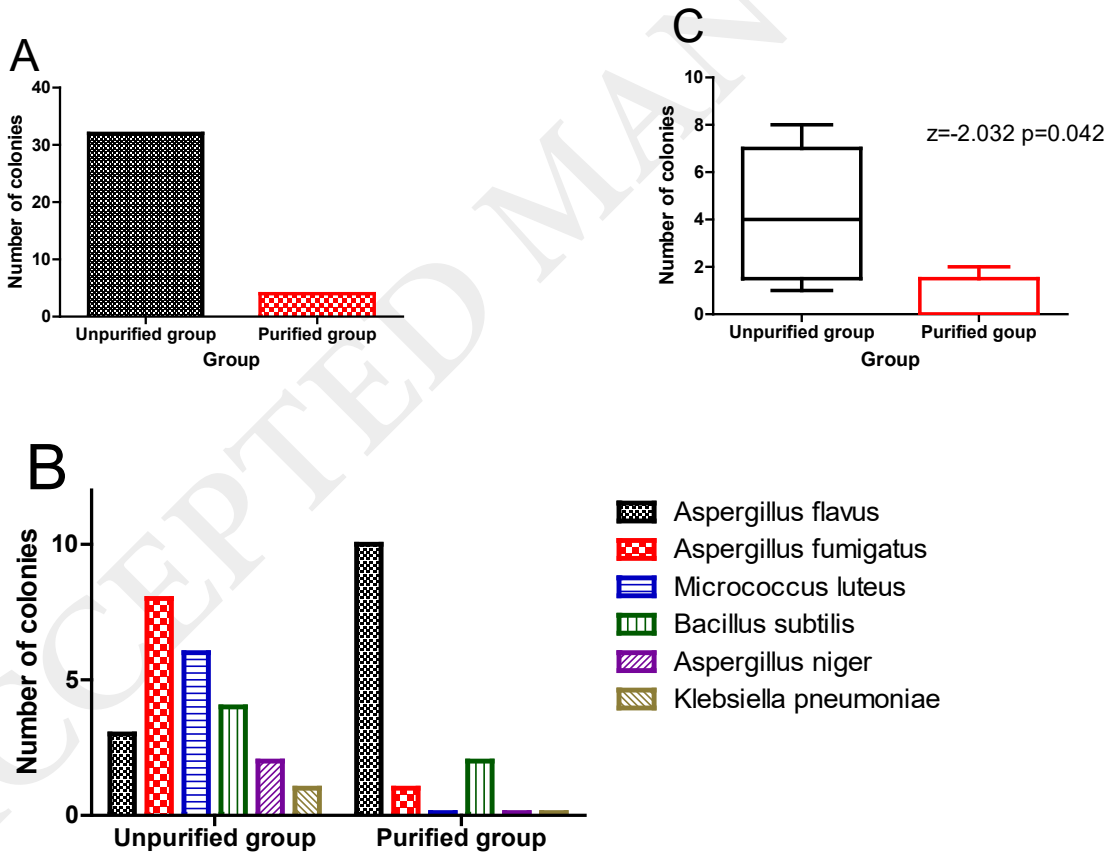

Fig. 5. Results of microbial culture and identification test: A: Results of total number of colonies in the microbial culture and identification test. B: Microbial culture and identification comparison results of the unpurified group and purified group. C: The statistical results of the unpurified group and purified group.

Among them, Aspergillus flavus is a special case, and the discussion part is analyzed separately. The purification efficiencies of the other five microorganisms were: Aspergillus fumigatus: 87.5%, Aspergillus niger: 100%, Klebsiella pneumoniae: 100%, Micrococcus luteus: 100%, Bacillus subtilis: 50%.

The researchers conducted nonparametric tests for the remaining 5 microorganisms: the researchers used SPSS statistics 26 to conduct nonparametric tests, and the statistical results were as follows:

At the level of $\alpha=0.05$, $P<0.05$, the researchers can think that the difference in the number of colonies of different types of microorganisms between the unpurified group and the purified group is statistically significant.

3.3 Case Study

The researchers retrieved information on patients undergoing bronchoscopy during the sampling period and recorded their clinical microbiological test results, such as sputum culture, blood culture, respiratory pathogen antibody detection, bronchoalveolar lavage fluid culture, bronchoalveolar lavage fluid NGS, etc. The researchers found that during the sampling period of the unpurified group, the corresponding microorganisms that were consistent with the experimental results were found as follows: Klebsiella pneumoniae, Actinomyces neisseria, Staphylococcus capitis, Gardnerella vaginalis, Pseudomonas aeruginosa spp., Stenotrophomonas maltophilia, Fusarium spp. During the sampling period of the purified group, the corresponding bacteria or fungi that were consistent with the experimental results were found as follows: Aspergillus flavus.

Related microbial test results of this experiment

Sampling days	Sampling in the unpurified group	Sampling in the decontamination group
Day 3	Klebsiella pneumoniae	
Day 4		Micrococcus luteus
Day 5	Pseudomonas aeruginosa, Actinomycetes, Gardnerella vaginalis, Fusarium	
Day 6	Klebsiella pneumoniae	Pseudomonas aeruginosa
Day 7	Staphylococcus capitis, Klebsiella pneumoniae	Actinomycetes, Aspergillus flavus, Pseudomonas aeruginosa
Day 8	Pseudomonas aeruginosa, Stenotrophomonas maltophilia	
Day 9	Pseudomonas aeruginosa, Klebsiella pneumoniae	
Day 11	Klebsiella pneumoniae	
Day 13	Stenotrophomonas maltophilia	
Day 14	Pseudomonas aeruginosa	
Day 15	Klebsiella pneumoniae	
Day 16	Pseudomonas aeruginosa	

DISCUSSION

Some researchers took samples from the filter for microbiological testing, which indirectly confirmed the purifying effect of the filter (Luongo et al., 2017). However, the microorganisms on the filter may reproduce or die and decompose, and cannot directly reflect the microorganisms in the air. Some researchers proved that air purifiers can reduce the concentration of allergens, improve the clinical performance of patients with allergic rhinitis, and reduce the need for drugs (Park et al., 2017; Park et al., 2020). Some researchers proved that air purifiers can reduce the total number of colonies in the culture

medium through the method of fungal culture (Chen et al., 2018). However, it has not been tested for bacterial species, and the purification effect of each microorganism cannot be seen directly. Some researchers used aerosol generators to artificially make certain types of microorganisms into aerosols, and verified the purifying effect of air purifiers in the ideal environment of a closed experimental chamber (Lee et al., 2019). Although all the above can intuitively reflect the purification effect, it is not an evaluation of the purification effect of the microorganisms that actually exist in the environment under the dynamic environment of the hospital's normal diagnosis and treatment. Some researchers proved that air purifiers can reduce the pathogenicity rate of a certain virus to animals by detecting the infection rate of animals to a certain virus (Dee et al., 2019). However, it cannot directly reflect the direct purification effect of the purifier on the biological aerosol.

The experiments in our research have confirmed that in a dynamic environment, after being purified by the new type of purifier, the total number of copies of microorganisms and the total number of colonies in the bronchoscope room are greatly reduced. The overall purification efficiency of NGS was 88.0% and the overall purification efficiency of Microbial culture counting and identification was 87.5%. Moreover, the single strains identified by NGS and microbial culture also declined to varying degrees, and the purification efficiency was between 50% and 100%, and the purification efficiency of most strains was 100%. It can be seen that in the dynamic environment of normal diagnosis and treatment in the bronchoscope room, the new purifier has a good purification efficiency for indoor biological aerosols. The long-term, large-capacity sampling method used in this experiment conforms to statistical laws. There have been some related studies that can confirm the purification effect of several different types of

air purifiers. The comparison results of the total copy number and total colony number of this study are similar to Zhao et al. (Yang et al., 2018), the difference is that the dynamic-bioaerosol part of this study focus on the actual collected data in the actual working environment. The results show that the purification effect of a single kind of microorganism has a certain fluctuation and is not constant. The researchers believe that the reason for the fluctuation of the purification effect of a single microorganism may be related to the different time of entering the bronchoscopy room and the different number of coughs in the different patients, which are uncontrollable factors in this study. But this is also the case, which proves that even in the dynamic environment of normal diagnosis and treatment, the steady-state displacement air purification system still has good removal efficiency for bioaerosols in the bronchoscope room.

After purification, the total amount and species of most microorganisms are greatly reduced. However, there is a special case among them: Aspergillus flavus. The investigators conducted a case search and found that the only patient who was positive for Aspergillus flavus in the clinical microbiological examination entered to the bronchoscopy room during the sampling period of the purified group . Therefore, the aerosols of Aspergillus flavus are produced in the purified group and collected by the sampling membranes. The sampling membranes sent by the researchers for microbial culture and bacterial species identification is the 2/B sampling membranes. This sampling membranes is 2.5-3.5m away from the patient and is the closest among several sampling membranes. Due to the application of the steady-state displacement flow of the purifier, the indoor air flow is slow, and the airflow cannot be detected by normal human bodies. The aerosol containing Aspergillus flavus has been eliminated by the purifier after it was

315 produced by the patient and collected by the sampling membranes. The reason for the
316 occurrence of Aspergillus flavus in the NGS results is that the sampling positions of the
317 sampling membranes of the remaining NGS No. 1, 3, 4, and 5 are far away from the
318 patients, the sampling time is short, and the aerosols generated by the patient is not
319 detected by the sampling membranes in time. Due to collection. This situation is
320 unavoidable in the dynamic environment of this experiment. The above is the reason why
321 the number of colonies of Aspergillus flavus in the purified group is more than that in the
322 unpurified group. Even so, the overall microbial clearance of the experiment was not
323 affected. In this experiment, two methods were used for microbial detection, and the
324 experimental results were surprisingly similar, which confirmed that in the dynamic
325 diagnosis and treatment environment, the purification efficiency of the new purifier for
326 indoor bioaerosol was relatively stable, ranging from 87.5% to 88%.

327 The main limitation of microbial culture and identification is that only a fraction of
328 the microorganisms present in the environment can be cultured and identified (Heidelberg
329 et al., 1997). Droffner ML, Pillai SD, et al. demonstrated that culture conditions also limit
330 the growth of viable and culturable microorganisms, such as mesophilic bacteria, Bacillus
331 subtilis, that exhibit adequate colony formation at temperatures between 5 and 55 °C
332 (Pillai et al., 2002), while thermophilic microorganisms prefer to be cultured at
333 temperatures above 50 °C. In this experiment, the G. vaginalis obtained by NGS was an
334 anaerobic bacteria, which was difficult to cultivate. Therefore, we can see that some
335 microorganisms have high copy numbers in the NGS results, but not in the microbial
336 culture and identification results. The possible reasons are as follows: (1) Not all the
337 collected microorganisms are suitable for the culture conditions at that time, and our

experiment The culture conditions used are the most commonly used conditions for microbial culture, and are more suitable for screening out those microorganisms that are suitable for surviving in the human body; (2) NGS can only detect the microorganisms above the detection limit (2 copies/ml) and within its detection range. All microorganisms, including live and dead bacteria. Therefore, some of the microorganisms detected by NGS may be dead bacteria and cannot form colonies of live bacteria on the medium. Combined with the results of NGS and microbial culture and bacterial species identification, it can be seen that the two detection methods have their own advantages and disadvantages. NGS is more sensitive, detects more types of microorganisms, and has fewer factors affecting the test results, but it cannot distinguish the dead and alive of microorganisms. That is, the microorganisms in the test results may be alive or dead. However, microbial culture and identification methods are more traditional, and the results are subject to more external interference, which is not as sensitive as NGS. However, because it is impossible for dead microorganisms to grow colonies, this method of microbial culture and identification can distinguish the dead and alive of microorganisms. Whether it is the results of colony counting or identification of bacterial species, they are all microorganisms in the living state of the reaction. Therefore, the combination of the two methods in this experiment can more objectively reflect the true reliability of the data.

The experimental purification time was set to 40 min. When the steady-state displacement air purification system is turned on for about 10 minutes, a relatively obvious particle purification effect can occur, which has been confirmed in the closed experimental chamber. However, in the normal diagnosis and treatment environment of the bronchoscope room, a more stable particle purification effect can be achieved 40

minutes after the purifier is turned on. Therefore, the purification time set in this experiment was 40 min. Compared with traditional disinfection methods, the new purifier can be applied in medical environment at any time, which can maximize the safety of medical staff and patients, reducing the probability of nosocomial infection.

At present, the new coronavirus pandemic has occurred in many countries, causing serious casualties and economic losses. Many outbreaks are closely related to bioaerosols in the indoor environment. Previous studies have agreed that infection in the endoscopic room is related to cross-infection of endoscopic instruments (Kong et al., 2021), but according to the study of bioaerosols in this experiment, the researchers found that the infection in the bronchoscopy room may also have a certain relationship with bioaerosols. However, there are a large number of bioaerosols in the hospital, which can cause drug resistance after repeated inhalation and infection by patients, making the treatment more difficult. The use of new purifiers can reduce drug-resistant bacterial infections in hospitals and reduce the rate of nosocomial infections. The steady-state displacement air purification system involved in this experiment has a removal efficiency of about 85% for microorganisms under the dynamic conditions of normal hospital diagnosis and treatment. Such removal efficiency may be useful for epidemic control and reduction of nosocomial infection of other diseases. positive effect.

The current 222nm light vaccine, which belongs to ultraviolet rays in a specific spectrum range, can kill the new coronavirus in the light-accessible position in the air without causing harm to the human body (Kitagawa et al., 2021). Traditional ultraviolet disinfection has a wide range of sterilization, but it has certain harm to the human body. Conventional disinfectant spraying can also regularly disinfect bioaerosols floating in the

air. Steady-state displacement air cleaning systems clean and replace indoor air anytime. The researchers believe that the combined use of light vaccine, traditional disinfectant spray and steady-state displacement air purification system can minimize the spread of pathogenic microorganisms such as the new coronavirus, reduce nosocomial infections, and ensure the safety of medical staffs and patients in the medical environment.

CONCLUSION

In the dynamic environment of normal diagnosis and treatment, the steady-state displacement air purification system has a good effect on removing biological aerosols in the bronchoscopy room.

Ethical approval and consent to participation (human ethics, animal ethics or plant ethics)

Not applicable.

Agree to publish

Author confirms:

The work described has not been published before;

Not considered for publication elsewhere; preprint exists: link: Lijuan Wu, XIXIN YAN. Under the dynamic environment of normal diagnosis and treatment in the bronchoscope room, the study of the removal efficiency of the new air purifier on

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whose publications have been approved by all co-authors, if any;Its publication has been approved by the competent authority of the institution in which the work is carried out.

Availability of data and materials

Not applicable.

interest competition

The authors have no conflicts of interest or financial relationships that affect this manuscript.

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Author's Contribution

Lijuan Wu: Methods, Software, Writing, Data Management, Editing

Zhigang Cai: Method

Jihong Li: Method

Jingwen Li: Method

Hao Pu: Method

Xianghong Liu: Methods, Data Management

Xixin Yan: Methods, Supervision

The manuscript was drafted by Wu Lijuan, directed and supervised by Yan Xixin. The two reviewed the manuscript and contributed the final version, which was approved by both parties.

Contributor's email

Wu Lijuan: xiaochengbuyu@163.com

Cai Zhigang: zhigang_cai@163.com

Li Jihong: 952224925@qq.com

Li Jingwen: 704920328@qq.com

Pu Hao: puhao@pathogeno.com

Liu Xianghong: liuxianghong1478@163.com

Yan Xixin: xi_xin_yan@126.com

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Highlights

1. Normal diagnosis and treatment in the bronchoscopy room.
2. The method of long-term, large-volume sampling is adopted.
3. The use of two microbial detection methods increased the accuracy and reliability.
4. The outdoor fresh air purification.
5. Steady-state replacement flow technology.

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