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Systematic Review

Impact of Microbial Load on Operating Room Air Quality and Surgical Site Infections: A Systematic Review

Sofia Chiletzari *, Anastasia Barbouni and Konstantinos Kesanopoulos

Department of National and Community Health, School of Public Health, University of West Attica, 11521 Athens, Greece; abarbouni@uniwa.gr (A.B.); kkesanopoulos@uniwa.gr (K.K.)
* Correspondence: pch20679241@uniwa.gr

Abstract: Surgical site infections (SSIs) are one of the most common causes of hospital-acquired infections worldwide, with significant clinical and economic implications. The aim of this review was to summarize the latest body of evidence on associations between microbial air load and SSIs. The systematic review was conducted using the revised Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA, 2020) method. Pubmed and Scopus databases were searched for the period 2014–2024. English language articles were searched for their reports on the microbial burden of operating room air and its association with surgical site infections. The present review includes a total of 36 articles related to microbial air load as an aggravating factor to air quality in the operating room and its association with SSIs. A direct correlation between microbial air load and the occurrence of SSIs was established through sampling methods and genetic analysis. A lack of consensus on the effectiveness of laminar air flow (LAF) systems was underlined, while temperature-controlled air flow seemed a promising alternative. One study found that each additional person in the operating room increases the number of bacterial colonies by 4.93 CFU/m^3 while another did not find significant changes in air quality. More than 20 air changes per hour (ACH) appeared to have better results in improving the quality of the air in the operation room. Airborne microbial contamination is multifactorial, and for some of those factors, a revision of the guidelines seems necessary. Artificial Intelligence (AI) and Next-Generation Sequencing methods show great promise for improving air quality in the future. This review calls for the implementation of international guidelines regarding air contamination limits in operating rooms and standardized air sampling methods, as well as further research for the efficacy assessment of air flow systems and emerging technologies based on AI in order to reduce the burden of SSIs and improve patient outcomes.

Keywords: microbial air contamination; air quality; operating room; operating theatre; surgical site infection; SSI

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1. Introduction

According to the World Health Organization (WHO), surgical site infections (SSIs) are the second most common hospital-acquired infection (HAI) worldwide. Surgical site infections are a particularly serious problem for Public Health, as in addition to increasing the morbidity and mortality of surgical patients, it also increases the economic costs to society and healthcare systems. Moreover, given the ever-increasing resistance to antibiotics (AMR), the problem takes on a further serious dimension, so it is necessary to optimize conditions and prevent bacterial air contamination for the safety of surgical patients [1].

A study showed that patients who develop SSI have a significantly extended length of stay (LOS) in the hospital with about 16 additional days. The average cost per infection ranges from about USD 5000–13,000 and SSIs account for USD $3.5–10 billion in annual healthcare spending [2]. In another review published in 2004, in Europe alone, data were reviewed from 84 studies and the economic cost of SSIs was estimated to range between EUR 1.47–19.1 billion [3]. Furthermore, SSIs rank fourth among HAIs in terms of Disability Adjusted Life Years (DALYs) and fifth overall behind four types of HAIs and influenza, with almost 60 DALYs/100,000 population [4].

The air in the operating room can act as an important vehicle for the transport of pathogens into the surgical field. Studies have demonstrated a clear correlation between the amount and size of airborne particles and the detection of viable airborne microorganisms. In fact, these studies have also highlighted the relationship between colony-forming units (CFUs) in the operating room environment and a higher incidence of SSIs. Thus, one of the important factors for the prevention of SSIs is ultra-clean air in the operating room [5].

The patient's skin is the direct source of infection in only 2% of SSI cases, while 98% of cases are associated with airborne particles. As for the exogenous sources from which microorganisms can originate and cause SSIs, these are airborne particles, operating room personnel (hands, other skin areas or mucous membranes), as well as inanimate objects such as various surgical instruments, room equipment, lavage solutions, etc. [6].

The pathogens isolated from SSIs differ, mainly depending on the type of surgery [7]. As shown by the European Centre for Disease Prevention and Control (ECDC) studies, the most common cause of SSI in recent years is *Staphylococcus aureus*. Indeed, it is worth noting that about 50% of these cases are attributed to methicillin-resistant strains (MRSA) [8]. Coagulase-negative *staphylococci*, *Enterococcus* spp. and *Escherichia coli* are also frequently identified [7].

SSIs are a significant contributor to the total burden of antimicrobial resistance (AMR), since studies have shown that the types of bacteria isolated in this type of infection are very often resistant to available treatments. A total of 68% of the total AMR burden is caused by four resistant microorganisms acquired mainly in healthcare, namely *Escherichia coli* and *Klebsiella pneumoniae* resistant to third-line cephalosporins, *methicillin-resistant Staphylococcus aureus* (MRSA) and carbapenem-resistant *Pseudomonas aeruginosa*. It was also estimated that 63.5% of cases of infections with antibiotic-resistant bacteria were healthcare-associated, resulting in 72.4% of deaths due to antibiotic resistance, according to 2015 data from the European Union (EU) and the European Economic Area [4].

The classic method of identifying pathogens is through culture [9]. Whole-genome sequencing (WGS) is a laboratory method that has emerged as a promising tool for epidemiological research due to its ability to distinguish between endemic strains [10]. WGS technology has already found many applications in the identification of the causative agent in SSIs. A prominent example is the study by Egyir et al. [11], who identified *S. aureus* using molecular methods and antimicrobial susceptibility testing, while describing the antimicrobial resistance patterns and molecular characteristics of the pathogen isolated from SSI patients in two hospitals in Accra, Ghana. A promising way for the early detection of SSIs seems to be third-generation sequencing and, especially, nanopore sequencing (NS). In a study by Whittle et al. [12], NS detected 100% of cases with biliary microbes within 14 h of sample collection compared to the average time of 98 h (range 80–152 h) required with typical culture. NS also shows potential in improving antibiotic management by significantly reducing the duration of non-targeted broad-spectrum antimicrobial therapy, which is currently given to surgical patients. This would limit the risks associated with antibiotic use for surgical patients, but can also contribute to addressing antimicrobial resistance, which poses a serious threat against Public Health.

Several factors can help reduce SSI, such as cleaning the skin at the surgical site and wearing face masks, gloves, gowns and special hats during surgery. In addition, the use of antibacterial suture material and appropriate short-term use of antibiotics immediately before and after surgery can reduce SSI [13].

The aim of this systematic review is to summarize the latest body of evidence on associations between microbial air load and SSIs. In addition, it seeks to identify the most important factors causing bacterial load in the air within operating rooms and to present ways to improve air quality in order to prevent SSIs.

2. Methodology

The review was performed by S.C. and K.K. by searching relevant articles in the widely acclaimed databases for systematic reviews, Pubmed and Scopus, with Scopus offering extensive multidisciplinary coverage and citation analysis, and PubMed providing comprehensive access to the biomedical literature. The search was performed using the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA 2020) method. Both databases were last searched on 9 June 2024. Inclusion criteria consisted of papers in English with full text available, published during 2014–2024 to include the last decade, which were related to the microbial air load in the operating theatre and its impact on causing SSIs, factors influencing the microbial air load and methods that are used or may be used in the future to reduce the impact of air contamination in the operating room.

Papers in any language other than English, papers not available in full text and papers with no reference to the previously mentioned subjects were excluded. While assessing the available papers by reading the abstract and full text, the process of exclusion was conducted, which involved identifying papers with no relevant content regarding the assessment of the correlation between microbial air load and SSIs, the factors that can lead to air contamination and the methods used or tested in order to mitigate the microbial burden in the air of an operating room.

The Boolean operators "OR" and "AND" were utilized to include synonyms and related terms and to combine different sets, respectively. In addition, the truncation (*) technique was used to include plural endings.

For the search in the Pubmed database, the term "human" in the species category was set as a filter to extract more targeted studies. Scopus did not provide such an option.

The query used in Pubmed was "("microbial air contamination" OR "air contamination" OR "air quality") AND ("surgical site infection*")". The query used in Scopus database was "("microbial air contamination" OR "air quality" OR "operating room air contamination") AND ("surgical site infection*" OR "ssi*")".

The search identified a total of 130 articles, 43 in the Pubmed database and 87 in the Scopus database. Duplicates were removed after both datasets were extracted to csv files and all dois were compared using Microsoft Excel. Management and reviewing of references was performed using Mendeley desktop software (Version 2.128.0, Elsevier Ltd. Amsterdam, The Netherlands). In total, 36 articles met the inclusion criteria. Through this entire process, two reviewers screened each paper independently.

The 36 articles included in this systematic review were grouped for the synthesis as follows:

- Papers that study the correlation between microbial air load in the operating theatre and SSIs.
- Papers which study factors influencing microbial air load within the surgical field in the operating theatre.

- Papers regarding the effectiveness of methods that are currently used or may be used in the future to reduce the microbial air load in the operating room, with the aim of limiting the incidence of SSIs.

Data extraction was performed using Microsoft Excel (Version 2503, Microsoft Corporation, Redmond, WA, USA). The information collected included the following:

Author(s);
Publication year;
Study design;
Population and sample size;
Interventions or exposures;
Outcomes measured;
Key findings.

3. Results

The search identified a total of 130 articles, 43 in the Pubmed database and 87 in the Scopus database. Of these, 38 duplicates were removed, along with 7 more articles for which no access was available. Then, 85 articles were screened by title and abstract. Through this process, 37 articles were excluded as they were not relevant to the aim of this systematic review. Of the 48 remaining articles that were checked for their full text, 12 were rejected due to a lack of relevant content. In total, 36 articles met the inclusion criteria. The full selection and retrieval process is shown in Figure 1.

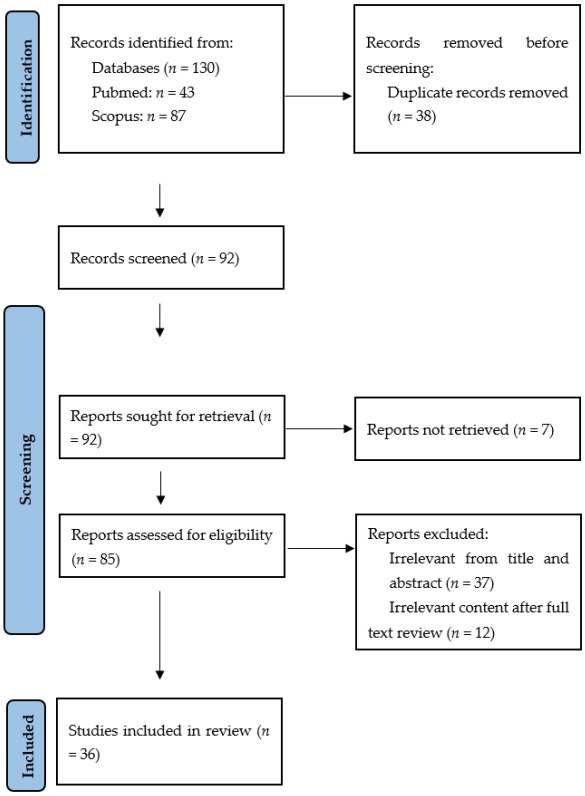


Figure 1. PRISMA flow diagram.

3.1. Air Quality and SSI Correlation

The existence of a direct correlation between microbial air load and the occurrence of surgical site infections is demonstrated by studies in which air samples were taken in operating rooms both at rest and in operation. Five relevant studies are included in this review, by Squeri et al. [14], Masia et al. [15], Gradisnik et al. [16], Montagna et al. [17]

and Birgand et al. [18]. All studies concluded that in the operating mode, the microbial air load increases significantly. This is proof that despite any preventive measures, the microbial air load in a functioning operating room is significant, thereby increasing the risk of causing SSIs.

Parvizi et al. [19] explored the potential risk of microbial contamination in the operating room air during total joint arthroplasty (TJA). The authors cited recent studies suggesting that viable airborne particles do indeed spread throughout the operating room, potentially increasing the risk of postoperative SSIs. They conclude that while maintaining a sterile operating room environment is essential, current understanding of the relationship between airborne microbial contamination and SSIs in TJA procedures is limited.

Stauning et al. [20] studied the genetic relationship between bacteria isolated from intraoperative air samples and those causing SSI. The researchers were able to highlight the genetic association between pathogens identified in intraoperative air samples and pathogens identified in the surgical wound that caused infection, thus demonstrating the direct relationship between air microbial load and SSIs.

An overview of the articles included in this section is provided in Table 1.

Table 1. Key information per article regarding "Air quality and SSI correlation".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Squeri, R., Genovese, C., Trimarchi, G. et al. [14]	Italy, 2019	Observational study	Microbiological air monitoring to evaluate differences in microbial contamination between empty and working OTs	A good ventilation system is only one requirement for clean air in operating rooms, as other factors, such as the behaviour of health professionals and environmental conditions, also affect the number of bacteria	Many important confounding factors were not analyzed, such as clothing systems, sampling points, door openings and staff behaviour
Masia, M. D., Dettori, M., Deriu, G. M. et al. [15]	Italy, 2020	Observational study	Implementation of microbial monitoring programmes to assess and control microbial contamination levels in empty and working OTs	Microbial monitoring in operating theatres can assess the health of the environment and the effectiveness of infection control measures, with significant contamination in limited areas and good surface disinfection	Machinery malfunctions may have affected the detection of Gram-negative bacteria in the air
Gradisnik, L., Bunc, G., Ravnik, J. et al. [16]	Slovenia, 2024	Observational study	Assessment of microbial air load during quiet and active periods in OTs	Microbiological air monitoring is extremely important for the safety and success of both surgical and postoperative practice	Number of locations and frequency of sampling
Montagna, M. T., Rutigliano, S., Trerotoli, P. et al. [17]	Italy, 2019	Observational study	Evaluation of air contamination levels in at-rest and in-operation orthopedic OTs, including microbial load and particulate matter	Air quality in orthopedic operating theatres in southern Italy is generally good, with no significant bacterial contamination at rest and low bacterial load even with always open doors	A limited number of confounding factors was assessed

Table 1. *Cont.*

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Birgand, G., Toupet, G., Rukly, S. et al. [18]	France, 2015	Multicentre observational study	Measurement of air contamination in operating rooms at different times during clean surgical procedures	Particle counting is a good surrogate for airborne microbiological infection in the operating room and can predict wound infection before skin closure in clean surgical procedures	- Sampling not from the exact same site - Confounders not taken into account - No identification of the strains cultured from the wound and the air
Parvizi, J., Barnes, S., Shohat, N., and Edmiston Jr, C. E. [19]	USA, 2017	Literature review	Assessment of the current practices regarding air quality in operating rooms	Air pollution in operating theatres contributes to SSIs and innovative strategies can reduce the risk of intraoperative microbial aerosols	N/A
Stauning, M. A., Bediako-Bowan, A., Bjerrum, S. et al. [20]	Ghana, 2020	Observational study	Active air sampling using a portable impactor during surgical procedures to collect intraoperative airborne bacteria	Perioperative airborne bacteria in low- and middle-income countries have a genetic link to LHD, which highlights the need for awareness of perioperative air quality in these areas	- The methodology used may have missed slow-growing and biofilm-associated organisms - Short period of study during one season

3.2. Air Flow Systems

The microbial air load in operating rooms is a multifactorial parameter. Several factors have been studied as aggravating factors with airflow systems being a controversial issue even after many relevant studies. In particular, the effectiveness of laminar airflow (LAF) systems has been the subject of extensive research over the last several decades, but researchers have not reached a common conclusion. For example, McHugh et al. [21] point out that some studies show no significant reduction in SSIs, while others suggest that LAF may even be associated with higher infection rates in some environments. Similarly, Pada and Perl [22] report that some studies suggest benefits, while others show no significant difference compared to conventional ventilation. Popp et al. [23] conclude that it is impossible to make a recommendation against the use of LAF systems in the operating room. They even report that LAF ceilings providing a 3.2×3.2 m^2 protection area proved to be superior to conventional turbulent ventilation as they are more effective in reducing bacteria and particles. These results were already noted in an earlier study in 2014 by Andersson et al. [24], who reported that LAF systems provide superior air quality during surgical procedures, with very low CFU levels near the surgical wound. Knudsen et al. [25] note that in their study, LAF-ventilated surgeries reduced the number of CFUs compared to turbulent airflow surgeries during arthroplasty procedures.

In contrast, the systematic review and meta-analysis by Ouyang et al. [26] concluded that there was no significant reduction in SSIs associated with the use of LAF systems in orthopedic surgeries compared to conventional ventilation systems. The lack of convergence of opinion on the effectiveness of LAF systems in reducing airborne microbial load is the reason why the WHO, in its most recent SSI guidelines, recommended against the use of LAF in arthroplasty procedures to reduce SSIs.

Table 2 summarizes key information from the articles included in this section.

Table 2. Key information per article regarding "Air flow systems".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
McHugh, S. M., Hill, A. D. K., and Humphreys, H. [21]	Ireland, 2015	Systematic review	To evaluate the effectiveness of laminar airflow systems in reducing surgical site infections (SSIs) and to discuss associated risks	Laminar airflow systems may not significantly reduce SSIs and could potentially increase risks in certain scenarios	N/A
Pada, S., and Perl, T. M. [22]	USA, 2015	Literature review	Analysis of common operating room practices, such as traffic patterns, door openings and airflow systems, with a focus on their effectiveness in infection control	Laminar air flow may not be necessary for prosthetic implant surgery and improving discipline in the operating room may reduce SSIs	N/A
Popp, W., Alefelder, C., Bauer, S. et al. [23]	Germany, 2019	Literature review	The use of LAF systems in operating rooms	LAF ceilings in operating rooms are beneficial for reducing SSIs, improving air quality and increasing worker safety	N/A
Andersson, A. E., Petzold, M., Bergh, I. et al. [24]	Sweden, 2014	Experimental study	The use of LAF systems in operating rooms	LAF systems in operating rooms provide high-quality air during surgery, with very low CFU levels near surgical wounds	- Sampling time - Main end point could be regarded as a surrogate end point - High risk of behavioural distortion among the participating staff
Knudsen, R. J., Knudsen, S. M. N., Nymark, T. et al. [25]	Denmark, 2021	Observational study	Use of LAF systems in operating theatres to reduce microbial air contamination	LAF ventilation systems in ORs effectively reduce airborne microbial contamination during total joint arthroplasty without significant impact from staff or door openings	- Small sample size - No identification of types of bacteria
Ouyang, X., Wang, Q., Li, X. et al. [26]	Italy, 2023	Systematic review and meta-analysis	Use of LAF ventilation systems in operating rooms to prevent SSIs	LAF systems do not lead to a significant reduction in the incidence of SSIs or in the number of bacteria in the air in orthopedic surgeries	Only English language articles were included

3.3. Door Openings

Another important factor affecting air quality in the operating rooms is the frequency of door openings. There seems to be a relative consensus in the international scientific community on the influence of this factor. Studies by Stauning et al. [27], Fernández-Rodríguez et al. [5] and Sadrizadeh et al. [28] concluded that frequent door openings in operating rooms contribute significantly to microbial air contamination, which may increase the risk of SSIs. The findings of the study by Sadrizadeh et al. [28] showed that details such as the type of door and its opening mechanism significantly affect the dispersion of

airborne particles in the operating room, with sliding doors causing less airborne microbial contamination compared to hinged doors.

Key information from the articles included in this section are presented in Table 3.

Table 3. Key information per article regarding "Door openings".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Stauning, M. T., Bediako-Bowan, A., Andersen, L. P. et al. [27]	Ghana, 2018	Observational study	Assessment of microbial air contamination in relation to door openings and the number of people present in the operating rooms	Reducing circulation flow in the operating room significantly improves air quality during orthopedic trauma surgery, supporting interventions aimed at preventing SSIs	- No internationally recognized standard for air sampling in operating rooms - The presence of an observer influenced the staff
Fernández-Rodríguez, D., Tarabichi, S., Golankiewicz, K. et al. [5]	USA, 2024	Prospective study	Use of an ultraviolet (UV) air filtration unit to reduce airborne pathogens	The air in the operating room that has an effective positive pressure ventilation system appears to contain microorganisms and identified pathogens. The use of a supplemental closed C-UVC unit with HEPA appeared to significantly reduce contamination of operating room air	- No baseline measurement of volatile organic compounds (VOCs) - Limited sampling location and small sample size - No adjustment for confounders
Sadrizadeh, S., Pantelic, J., Sherman, M. et al. [28]	USA, 2018	Experimental study	Use of sliding doors to minimize airborne particle dispersion	This study shows a significant relationship between the opening of the operating room door and room pressure, as well as the level of contaminants	- The application of CFD techniques into the engineering applications may involve limitations, such as grid-dependent solutions - Slow or uncertain convergences, and the skill level of the operator

3.4. Personnel Movement

Staff movement also affects the sterile environment in operating rooms, which is critical for the prevention of SSIs. This was pointed out by Stauning et al. [27], and it was previously demonstrated by Sadrizadeh et al. [29] that increasing the number of staff members disrupts the airflow pattern of ventilation, leading to a higher concentration of suspended bacterial particles in the surgical site. The study by Cao et al. [30] concluded that medical staff disrupt the distribution of clean air near the surgical patient, while Fu Shaw et al. [31] reported that each additional person in the operating room increases the number of bacterial colonies by 4.93 CFU/m^3. In contrast, Montagna et al. [17] reported that within their study, the number of individuals did not appear to significantly affect air quality.

An overview of not previously cited articles is presented in Table 4.

Table 4. Key information per article regarding "Personnel Movement".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Stauning, M. T., Bediako-Bowan, A., Andersen, L. P. et al. * [27]	Ghana, 2018	Observational study	Assessment of microbial air contamination in relation to door openings and the number of people present in the operating rooms	Reducing circulation flow in the operating room significantly improves air quality during orthopedic trauma surgery, supporting interventions aimed at preventing SSIs	No internationally recognized standard for air sampling in operating rooms; the presence of an observer influenced the staff
Cao, G., Storås, M. C., Aganovic, A. et al. [30]	Norway, 2018	Observational study	To investigate whether surgeons' activities and surgical facilities disturb clean air distribution in orthopedic operating rooms with laminar airflow	Surgeons' movements and certain facility designs can disturb clean air distribution, potentially increasing the risk of contamination in the surgical field	The interaction with a thermal manikin was studied at only 1 discharge velocity from the LAF system; the anemometers could not measure the direction of the airflow, only magnitude; the heating system of the thermal manikin also had some limitations on the reliability of the results
Sadrizadeh, S., Tammelin, A., Ekolind, P. et al. [29]	Sweden, 2014	Experimental study	Adjustments in the number of staff and their internal positioning within the operating room to reduce airborne contamination and SSI risk	Increased staffing in operating theatres increases the concentration of particles carrying bacteria, posing a risk of infection in the surgical field	N/A
Fu Shaw, L., Chen, I. H., Chen, C. S. et al. [31]	Taiwan, 2018	Observational study	Assessment of factors influencing microbial colonies, such as air changes per hour (ACH), room design, staff movement and environmental conditions	A well-controlled ventilation system and infection control procedures are vital to reduce microbial colonies in operating theatres, and staff and their activities play a key role	Study performed in a single medical centre, so inferences to other levels of hospitals should be drawn with caution; bacterial air sampling and data collection on holidays were excluded, only bacterial genera were identified
Montagna, M. T., Rutigliano, S., Trerotoli, P. et al. * [17]	Italy, 2019	Observational study	Evaluation of air contamination levels in at-rest and in-operation orthopedic OTs, including microbial load and particulate matter	Air quality in orthopedic operating theatres in southern Italy is generally good, with no significant bacterial contamination at rest and low bacterial load even with doors always open	A limited number of confounding factors was assessed

* The reference appears multiple times in the tables as it addresses multiple risk factors.

3.5. Air Changes per Hour

One way to maintain air quality within operating rooms at least at acceptable levels is air changes per hour (ACH). The recommendation by the WHO for this parameter is approximately 20 air changes per hour. This factor was studied by Fu Shaw et al. [31] and

Zhang et al. [32]. Both noted that more than 20 ACH seems to have better results in im-proving air quality. Key information of the studies included in this section can be found in Table 5.

Table 5. Key information per article regarding "Air Changes Per Hour".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Fu Shaw, L., Chen, I. H., Chen, C. S. et al. * [31]	Taiwan, 2018	Observational study	Assessment of factors influencing microbial colonies, such as air changes per hour (ACH), room design, staff movement and environmental conditions	A well-controlled ventilation system and infection control procedures are vital to reduce microbial colonies in operating theatres, and staff and their activities play a key role	Study performed in a single medical centre, so inferences to other levels of hospitals should be drawn with caution; bacterial air sampling and data collection on holidays were excluded, only bacterial genera were identified
Zhang, Y., Cao, G., Feng, G. et al. [32]	China, 2020	Observational study	Adjustment of air change rates (ACR) in the operating rooms to improve air quality	Increasing air change rates in operating rooms with mixing ventilation can improve air quality in the surgical microenvironment, reducing exposure risks and improving ventilation efficiency	Limited number of sensors led to a small number of measurements

* The reference appears multiple times in the tables as it addresses multiple risk factors.

3.6. Clothing Systems and Equipment

As much of the airborne microbial load in operating rooms comes from staff, studies have also been conducted on the type of clothing to be used, with the aim of preventing SSIs. Two studies investigating this factor were included in this review, by Kasina et al. [33] and Cao et al. [34]. Kasina et al. concluded that a single-use polypropylene clothing system can offer better CFU reduction than both reusable olefin and mobile laminar airflow unit-assisted cotton/polyester clothing systems. Cao et al. highlighted that the surgical team needs to wear clean air suits with the surgical hood tucked under the suit. As these studies showed, even simple practices, such as the use of appropriate clothing and the correct positioning of the surgical hood, can bring significant benefits in improving the microbial burden in the operating room air, with a direct impact on patient safety.

The air in operating theatres can also be burdened by pieces of equipment. One such case was studied by Lange [35] who focused on forced-air warming (FAW) devices, which are commonly used to maintain patient body temperature during surgery. As it has been found that FAWs pose a real risk of cross-contamination of the operating room environment, it is necessary to consider alternative ways of warming patients, as suggested by the author.

Table 6 provides a summary of the key points of the studies cited in this section.

Table 6. Key information per article regarding "Clothing systems and equipment".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Kasina, P., Tammelin, A., Blomfeldt, A. M. et al. [33]	Sweden, 2016	Observational study	Use of three different clean air suits designed to reduce bacterial load in the operating room	Use of three different clean air suits designed to reduce bacterial load in the operating room	- High rate of measurement exclusion (15%), unexplained outliers - Study conducted in standardized laboratory settings

Table 6. *Cont.*

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Cao, G., Pedersen, C., Zhang, Y. et al. [34]	Norway, 2021	Experimental study	Use of specific clothing systems (e.g., clean air suits) and optimized human activities to maintain a sterile environment	It is possible to achieve ultra-clean air requirements (10 CFU/m^3) during surgical procedures in operating rooms with mixed airflow systems, using appropriate clothing and low surgical activity	- Experimental measurements performed during different seasons - There may be unknown factors affecting air quality
Lange, V. R. [35]	USA, 2022	Retrospective study	Use of FAW devices in operating rooms to maintain perioperative normothermia	FAW devices to maintain patient body temperature in operating rooms can introduce bacteria, increasing the risk of patient infection and the risk of SSIs	N/A

3.7. Covering Goods

Wistrand et al. [36] investigated time-dependent bacterial contamination of air in covered and uncovered sterile products in the operating room. The researchers concluded that protecting sterile fields from bacterial air contamination with sterile covers enhances the durability of sterile products by up to 24 h. This could work to the benefit of patients by enhancing their safety and allowing for the advanced preparation of surgical sterile materials for acute procedures, thus improving the quality of care while reducing the corresponding cost.

The effectiveness of this practice was the subject of a systematic review and meta-analysis by Wistrand et al. [1]. The researchers concluded that covering sterile items in the operating room significantly reduces bacterial airborne contamination. Reduced bacterial contamination may result in a safer operating room environment for the patient, and the use of covers for sterile items may have the potential to further reduce SSIs.

Key information from the articles included in this section are presented in Table 7.

Table 7. Key information per article regarding "Covering goods".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Wistrand, C., Söderquist, B., and Sundqvist, A. S. [36]	Sweden, 2021	Experimental intervention study	Covering sterile fields to reduce bacterial air contamination over time	Covering sterile fields with sterile covers reduces bacterial contamination of the air, enhancing the shelf life of sterile products by up to 24 h, potentially benefiting patient safety and reducing climate impact and costs	Study performed using simulation thereby not reflecting real-life theatre work patterns
Wistrand, C., Westerdahl, E., and Sundqvist, A. S. [1]	Sweden, 2024	Systematic review and meta-analysis	Covering sterile goods to protect them from bacterial air contamination	Covering sterile goods in the operating room significantly reduces bacterial contamination of the air and prolongs the sterility of surgical goods	- The studies included used CFU as the outcome instead of measuring SSIs - Uncertainty over the sampling methods and the different methods used to collect and isolate bacteria - The meta-analysis did not evaluate the relationship between CFU and time

3.8. Assessment of Methods to Overcome Identified Challenges

3.8.1. Air Systems

A number of studies that focused on proposals with a different focus, but always with the common theme of reducing the impact of airborne microbial load in causing SSIs, are included in this review.

Mullen and Wieser [37] implemented a high-efficiency particulate air and ultraviolet air recirculation system (HUAIRS), demonstrating a remarkable reduction in airborne particulate matter and microbial contamination of the air. This improvement was associated with a significant reduction in SSI rates during the study in an orthopedic hospital.

In the only study identified as part of this systematic review that focused exclusively on pediatric procedures, Wahdan et al. [38] evaluated a new electronic air filtration unit in addition to a conventional ventilation system. The study concluded that the use of such a device, in addition to the conventional ventilation system, significantly improved air quality in operating rooms, thereby reducing the incidence of SSI.

Fernández-Rodríguez et al. [5] and Messina et al. [39] tested an air filtration device with a crystalline ultraviolet module (C-UVC). In these studies as well, an association between the use of the UV air filtration unit and the reduction in SSI was observed.

A mobile ultra-clean laminar airflow unit was evaluated by Sadrizadeh and Holmberg [40] to maintain a sterile environment and had very encouraging results. This study found that the addition of such a device reduces the counts of airborne bacteria and surface contamination to a level acceptable for infection-prone surgeries.

Another type of mobile unit, this time of laminar airflow (MLAF), was evaluated by von Vogelsang et al. [41]. It was found that the use of these devices significantly reduced the number of CFUs in the surgical field area and over the instrument table. Although the researchers' primary focus was on airborne bacteria counts, the study suggests a possible reduction in SSIs due to lower levels of contamination. As the researchers say, further re-search is needed to establish a direct correlation between the reduction in CFUs and the frequency of SSIs.

The potential of localized exhaust and air curtain systems to lower airborne particles in surgical patients was studied by Tan et al. [42]. This combination provided a notable reduction in the number of particles settling on surgical patients. The implementation of these systems could significantly improve infection control and also enhance patient safety and elevate healthcare quality and outcomes.

Alsved et al. [43] studied a counter-proposal to traditional laminar air flow (LAF) and turbulent mixed airflow (TMA) systems. Those two systems were compared with a newly developed ventilation technique, temperature-controlled airflow (TcAF), and it was found that TcAF and LAF remove bacteria more efficiently from the air than TMA, especially close to the wound and at the instrument table.

Lind et al. [44] propose an innovative solution with the installation of a ventilation unit that provides a high volume of air into the operating room with the application of low-velocity wall-mounted diffusers. This technique may provide a solution to the problem of microbial contamination of the air within the operating room due to door opening. This method has been confirmed by computational fluid dynamics (CFD) simulations and laboratory measurements.

Wang and Sadrizadeh [45] presented a numerical evaluation of a new ventilation strategy for operating rooms and compared its effectiveness with traditional turbulent mixing airflow (TMA) and vertical laminar airflow (LAF) systems. In order to simulate the airflow patterns and particle distribution in an operating room under different ventilation strategies, the researchers used computational fluid dynamics (CFD). Results showed that temperature-controlled airflow (TAF) circumvents obstacles in the airflow and also

minimizes the air circulation zones in the periphery of the operating room. The results confirmed the superiority of TAF and LAF systems to TMA in securing high-level air cleanliness. Also, TAF showed that it can be used as an alternative to LAF.

An overview of the basic characteristics of the studies cited in this section are presented in Table 8.

Table 8. Key information per article regarding "Assessment of Methods to Overcome Identified Challenges—Air systems".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Mullen, A. N., and Wieser, E. [37]	USA, 2024	Interventional study	Improvement of operating room air quality. This is suggested by the study's aim to enhance air quality to reduce infections	The implementation of high-efficiency particulate air recycling and infrared air recycling devices (HUAIRS) in a specialist orthopedic hospital is associated with a significant reduction in SSI rates and levels of intraoperative air contamination	Confounding variables not analyzed; no randomization of surgeries that either used a HUAIRS device or not; no blinded SSI assessment from HUAIRS use; single-site trial
Wahdan, M. M., El-Awady, M. Y., Abo ElMagd, N. M. et al. [38]	Egypt, 2021	Interventional study	Use of electronic air filtration systems to improve air quality and reduce the risk of SSIs	Electronic air filtration in operating rooms significantly reduces the rates of SSIs in pediatric surgery	Sampling of air and surgeries were performed in the presence of mixed ventilation in the ORs; only 70.5% of SSIs were swabbed
Fernández-Rodríguez, D., Tarabichi, S., Golankiewicz, K. et al. * [5]	USA, 2024	Prospective study	Use of an ultraviolet (UV) air filtration unit to reduce airborne pathogens	The air in the operating room that has an effective positive pressure ventilation system appears to contain microorganisms and identified pathogens. The use of a supplemental closed C-UVC unit with HEPA appeared to significantly reduce contamination of operating room air	No baseline measurement of volatile organic compounds (VOCs); limited sampling location and small sample size; no adjustment for confounders
Messina, G., Spataro, G., Catarsi, L. et al. [39]	Italy, 2020	Experimental study	Use of a mobile device designed to reduce airborne particulate matter	A mobile disinfection and air recirculation unit, equipped with a C crystalline UV reactor and HEPA filter, reduces particulate matter in the operating room, thus improving air quality and potentially reducing the possibility of SSI	Contamination measurements on the operating table were not performed in real surgery; no assessment for confounding factors
Sadrizadeh, S., and Holmberg, S. [40]	USA, 2015	Experimental study	Use of a portable ultra-clean exponential airflow unit to reduce airborne particle distribution	A mobile ultra-clean LAF unit effectively reduces airborne bacteria and surface contamination in operating rooms, making them suitable for SSI-prone surgical procedures	Limited data available in the literature are well suited for model validation; only limited experimental data including detailed and accurate information for numerical validation; must contain detailed information about flow and thermal boundary conditions, measured parameters, statistical errors, and errors related to analysis
von Vogelsang, A. C., Förander, P., Arvidsson, M. et al. [41]	Sweden, 2018	Quasi-experimental study	Use of LAF units to reduce airborne bacterial contamination	Mobile LAF units effectively reduce airborne bacterial contamination during neurosurgeries, providing ultra-clean air for clean neurosurgeries that are prone to SSI	Absence of a standardized CFU sampling position; more samples from the surgical site were desired; temperature and humidity were not measured

Table 8. *Cont.*

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Tan, H., Othman, M. H. D., Kek, H. Y. et al. [42]	Malaysia, 2024	Experimental study	Use of localized exhaust systems combined with air curtain technology to reduce airborne particle settlement	The adoption of local extraction and air curtain systems in operating rooms could significantly improve infection control, enhance patient safety and improve the quality and outcomes of healthcare	Sole reliance on surface area of exposure as the primary metric for assessing risk related to patient wounds; study performed using a constructed OR, and thus, the complexity introduced by the variability in staff positioning and movement were not assessed
Alsved, M., Civilis, A., Ekolind, P. et al. [43]	Sweden, 2018	Experimental study	Use of temperature-controlled airflow (TcAF) ventilation systems to improve air quality and reduce airborne bacterial contamination	TcAF and LAF effectively remove bacteria from the operating room air, while TcAF uses less energy and provides a more comfortable working environment compared to LAF	Only one of many existing designs of LAF was studied; using CFU as a measure of airborne microbial load means certain bacterial cells may be missed
Lind, M. C., Sadrizadeh, S., Venås, B. et al. [44]	Norway, 2019	Experimental study	Implementation of a ventilation strategy involving low-velocity wall-mounted diffusers to minimize airborne particle migration during door openings	The installation of a ventilation unit with low-velocity wall diffusers can significantly reduce the migration of contaminants into the operating rooms during door opening activities	The placement of wall diffusers in the anteroom can be problematic; potential impact of installing airflow barriers inside the OR as this may interfere with existing airflows
Wang, C., and Sadrizadeh, S. [45]	USA, 2018	Experimental study	A novel ventilation strategy for operating rooms, designed to improve air quality and reduce airborne contamination	The simulation results confirmed the superiority of LAF and TAF to TMA in delivering high-purity air and also showed that TAF can serve as an effective alternative to LAF	Study was performed on the basis of a specific case; the ventilation systems were evaluated under different airflow rates; assumptions adopted in the simulation regarding the number of staff, their clothing and activity level as well as the OR foot traffic may significantly underestimate the CFU concentration in the ORs

* The reference appears multiple times in the tables as it addresses multiple risk factors.

3.8.2. Artificial Intelligence

Artificial Intelligence (AI) has entered the field of SSI prevention in recent years by improving air quality. More specifically, Colella et al. [46] propose a decision support system (DSS) based on a fuzzy inference system (FIS) for monitoring air quality in the operating room. The goal is to maintain the surgical environment and optimize subsequent management decisions, thus preventing SSIs. This is a promising future solution; however, it seems necessary to further test and develop this system to address limitations related to the type and number of data.

A system that uses AI to enhance the infection control by maintaining optimal air quality was also developed by Jamali et al. [47]. The authors propose a sophisticated decision support system that integrates fuzzy inference system (FIS) with an adaptive fuzzy neural inference (ANFIS) to optimize air quality through controlling the distribution of air flow. This is an innovative approach, which exploits the potential of the combination of fuzzy logic and neural networks with promising results for the control of air pollution.

Key information regarding the articles cited in this section are presented in Table 9.

Table 9. Key information per article regarding "Assessment of Methods to Overcome Identified Challenges—Artificial Intelligence".

Authors	Country, Year of Publication	Type of Study	Objective	Key Findings	Limitations
Colella, Y., Valente, A. S., Rossano, L. et al. [46]	Italy, 2022	Experimental study	Implementation of a fuzzy inference system (FIS) for monitoring and assessing indoor air quality to reduce airborne contamination	The fuzzy inference system (FIS) effectively monitors operating room air quality, reducing airborne contamination and preventing SSIs by analyzing input data and physician movements	- The apparatus used to measure input parameters could not determine which and how many aerosolized particles were live bacteria - If a new parameter were introduced to monitor air quality, all inference rules in the fuzzy system would need to be reconfigured
Jamali, N., Gharib, M. R., and Koma, B. O. [47]	Iran, 2023	Computational modelling study	To develop and evaluate a neuro-fuzzy decision support system for optimizing indoor air quality in operating rooms	The neuro-fuzzy system successfully identified optimal air quality parameters, providing a decision-making tool for real-time adjustments in operating rooms	- The limitation in terms of input type and number can be a significant factor for future research - Deep learning and meta-heuristic optimization algorithms could be improved to estimate indoor air quality more accurately

Statistical methods used in each study are presented in Table 10. Effect measures and validation methods are also included in the same table.

Table 10. Effect measure, statistical methods and validation per article in order of citation in the "Results" section.

Authors	Effect Measures—Statistical Analysis	Statistical Validation
Squeri, R., Genovese, C., Trimarchi, G. et al. (2019) [14]	Rho of Spearman	p-value < 0.05
Masia, M. D., Dettori, M., Deriu, G. M. et al. (2020) [15]	Range, IQR, frequencies	N/A
Gradisnik, L., Bunc, G., Ravnik, J. et al. (2024) [16]	N/A	N/A
Montagna, M. T., Rutigliano, S., Trerotoli, P. et al. (2019) [17]	Spearman's correlation coeficient	p-value < 0.05
Birgand, G., Toupet, G., Rukly, S. et al. (2015) [18]	IQR, Odds Ratio	p-value < 0.05, CI 95%
Parvizi, J., Barnes, S., Shohat, N., and Edmiston Jr, C. E. (2017) [19]	N/A	N/A
Stauning, M. A., Bediako-Bowan, A., Bjerrum, S. et al. (2020) [20]	Odds Ratio, Wilcoxon-rank-sum-test	p-value < 0.05, CI 95%
McHugh, S. M., Hill, A. D. K., and Humphreys, H. (2015) [21]	N/A	N/A
Pada, S., and Perl, T. M. (2015) [22]	N/A	N/A
Popp, W., Alefelder, C., Bauer, S. et al. (2019) [23]	N/A	N/A
Andersson, A. E., Petzold, M., Bergh, I. et al. (2014) [24]	Incidence Rate Ratio	p-value < 0.05, CI 95%
Knudsen, R. J., Knudsen, S. M. N., Nymark, T. et al. (2021) [25]	Median values, Mann–Whitney U test	p-value < 0.05, CI 95%

Table 10. *Cont.*

Authors	Effect Measures—Statistical Analysis	Statistical Validation
Ouyang, X., Wang, Q., Li, X. et al. (2023) [26]	Odds Ratio, Risk Ratio	p-value < 0.05, CI 95%
Stauning, M. T., Bediako-Bowan, A., Andersen, L. P. et al. (2018) [27]	Estimate	p-value < 0.05, CI 95%
Fernández-Rodríguez, D., Tarabichi, S., Golankiewicz, K. et al. (2024) [5]	t-test, Mann–Whitney U test	p-value $\leq$ 0.05
Sadrizadeh, S., Pantelic, J., Sherman, M. et al. (2018) [28]	N/A	N/A
Cao, G., Storås, M. C., Aganovic, A. et al. (2018) [30]	N/A	N/A
Sadrizadeh, S., Tammelin, A., Ekolind, P. et al. (2014) [29]	N/A	N/A
Fu Shaw, L., Chen, I. H., Chen, C. S. et al. (2018) [31]	r	p-value < 0.05, CI 95%
Zhang, Y., Cao, G., Feng, G. et al. (2020) [32]	Mean values	Relative difference in mean values and standard deviation
Kasina, P., Tammelin, A., Blomfeldt, A. M. et al. (2016) [33]	Mann–Whitney U-test, median values	p-value
Cao, G., Pedersen, C., Zhang, Y. et al. (2021) [34]	N/A	N/A
Lange, V. R. (2022) [35]	N/A	N/A
Wistrand, C., Söderquist, B., and Sundqvist, A. S. (2021) [36]	Mann–Whitney U test, Kaplan-Meier curves, x^2-test or Fisher's exact test	p-value < 0.05, CI 95%
Wistrand, C., Westerdahl, E., and Sundqvist, A. S. (2024) [1]	Z	p-value < 0.05, CI 95%
Mullen, A. N., and Wieser, E. (2024) [37]	IQR, x^2, Z-test, Student's t-test	alpha of 0.05 p-value
Wahdan, M. M., El-Awady, M. Y., Abo ElMagd, N. M. et al. (2021) [38]	x^2, Risk Ratio	p-value < 0.05, CI 95%
Messina, G., Spataro, G., Catarsi, L. et al. (2020) [39]	Wilcoxon rank test for paired data	p-value < 0.05, CI 95%
Sadrizadeh, S., and Holmberg, S. (2015) [40]	Mean values	Standard deviation (graphically)
von Vogelsang, A. C., Förander, P., Arvidsson, M. et al. (2018) [41]	Odds Ratio, Mann-Whitney U-test	p-value < 0.05, CI 95%
Tan, H., Othman, M. H. D., Kek, H. Y. et al. (2024) [42]	Linear regression	R^2
Alsved, M., Civilis, A., Ekolind, P. et al. (2018) [43]	Mann–Whitney U test, Spearman rank-order correlation test	p-value < 0.05, CI 95%
Lind, M. C., Sadrizadeh, S., Venås, B. et al. (2019) [44]	N/A	N/A
Wang, C., and Sadrizadeh, S. (2018) [45]	N/A	N/A
Colella, Y., Valente, A. S., Rossano, L. et al. (2022) [46]	N/A	N/A
Jamali, N., Gharib, M. R., and Koma, B. O. (2023) [47]	Error Ratio	Accuracy, Precision

4. Discussion

This systematic review underlines the correlation between the microbial load in the air of operating rooms and the incidence of SSIs. The international literature shows that there is a direct correlation between microbial air load and SSIs, as pathogens suspended above the surgical site settle in the wound and infect it. Studies have shown that airborne pathogens settling in surgical wounds are a primary cause of SSIs, thus highlighting

the need for stringent air quality control measures. However, all relative studies have some limitations. These include sampling procedures [16,18] and also not taking into consideration confounding factors [14,17,18] such as the frequency of door openings, the number of staff within the operating room and the way they move, the frequency of air changes per hour, elements of equipment, and staff clothing systems, potentially biassing the results.

4.1. Efficiency of the Current Airflow Systems

Laminar airflow systems have been a focus in the prevention of SSIs; however, their efficiency is still controversial. Studies have produced conflicting results: some demonstrate reduced microbial load and SSIs [23–25], while others show no significant benefit [22,26] or even possible risks [21]. These conflicting outcomes derive from differences in sampling locations, unaccounted confounders, and small sample sizes. This lack of consensus indicates that the performance of different air flow technologies should be evaluated against specific contexts, considering a number of variables such as the type of procedure, the design of the room and the behaviour of the personnel. There is a clear need for extensive research in the filed in order to establish specific guidelines as far as the airflow systems is concerned. This review identifies temperature-controlled air flow as a promising alternative, pointing to ways in which comparative studies might be used to develop refined ventilation guidelines.

4.2. Environmental and Behavioural Factors

Throughout the literature reviewed, door openings and movements of personnel appear to be critical disruptors to sterile conditions. These call for stricter operation and facility designs that limit such disturbances. Inventions like automatic sliding doors and restricted entry may be significant contributors to reducing microbial contamination of the surgical field. Furthermore, training staff on the best movement patterns and use of attire will improve compliance with minimizing contamination risks. Regarding the rate of air changes per hour, the studies included in this review call for an update of the international guidelines issued by the WHO [31,32]. However, these results are based on limited testing leading to a small number of measurements. This means that more studies are needed before considering updating those guidelines.

4.3. Emerging Technologies

Advanced filtration systems, such as High-Efficiency Particulate Air (HEPA) and ultraviolet-based systems, show considerable promise in microbial burden reduction. Further modification through mobile laminar airflow units and local exhaust could enhance the surgical field sterility. Large-scale, multicentre trials would be necessary for the establishment of the efficacy and cost-effectiveness of these systems.

Artificial Intelligence (AI) is becoming increasingly recognized as a powerful tool for air quality management, offering decision support through technologies like fuzzy logic and neural networks. These systems seem to be rather promising, yet their application to varied clinical environments needs extensive testing against current limitations regarding data variability and algorithmic precision.

4.4. Gaps in Knowledge and Future Directions

Despite the progress that has been made, the previously mentioned weaknesses continue to pose a threat for surgical patients.

One of the major issues is the clear distinction in the evidence of LAF systems that necessitates the investigation of other factors as well. Very few studies have considered the

combined effect of multiple interventions, which limits our ability to optimize operating room conditions.

In recent years, very few case studies have been conducted in this field and the great majority of them concern adults and common surgeries, not children or specialized procedures, despite the fact that such populations perhaps present unique risks and needs.

A study, apart from involving various fields such as engineering, epidemiology, and clinical expertise, should also focus on the innovation and validation of solutions to be conducted in pediatrics. The capability of technologies to provide real-time monitoring of staff and environmental sensors is an added plus that should be given attention.

4.5. Implications for Practice and Policy

The pathogenesis of SSI is multifactorial and include both patient- and procedure-specific factors [21]. Contamination of the surgical field by suspended particles is attributed in 30% of cases to direct sedimentation of the particles in the wound and in 70% of cases to sedimentation on the instruments and hands of the surgeon and their subsequent transport to the wound. Therefore, surgical contamination is mainly due to airborne particles, some of which may carry microorganisms [7]. This highlights the importance of air quality control in the operating room and the need to set international standards for air contamination limits in the operating room. This could be achieved by performing repeated sampling over time, thus creating a historical database for each type of surgery, which can lead to the calculation of the appropriate limits.

This review also brings out the lack of international standards for air sampling during surgery, which is underlined as a limitation in several included studies. Therefore, choice of media, sampling volume, frequency, sampling position and incubation time vary among the studies conducted worldwide, thus making it difficult to compare results obtained from different samplers.

Other aspects that need to be addressed in terms of guidelines include air change rates, staff training and proper use of equipment.

Investments in technology and infrastructure should be made. These must be scalable, cost-effective and aim at diverse healthcare settings.

5. Limitations

Despite the recognition of the potential added value on the validity of this systematic review, a risk of bias assessment was not performed for the included studies. This decision was driven by the heterogeneity of the study types included, the lack of a generally accepted consensus methodology about whether it is appropriate to do so and the emphatically discouraged use of scales that yield a summary score.

Regarding the heterogeneity, the decision to include different types of studies in this review was made consciously as the aim was to cover—as much as possible—the broad spectrum of risk factors for microbial air contamination in the operating room.

A limitation of this review is its lack of registration in the International Prospective Register of Systematic Reviews (PROSPERO) or a similar database.

6. Conclusions

This review, to our knowledge, is the first to summarize the content of the existing international literature on all major factors affecting air quality in the operating room. It emphasizes the interrelated functions of ventilation systems, environmental controls, personnel conduct and innovative technologies in preserving sterile surgical settings in order to prevent SSIs.

It highlights the difficulty in maintaining air quality within operation theatres and how it directly influences SSIs. It calls for the implementation of international guidelines regarding air contamination limits in operating rooms, as well as air sampling methods. Further research is needed in order to assess the efficacy of different air flow systems such as LAF, and emerging technologies bases on AI such as FIS and ANFIS. Filling the gaps in existing knowledge and adopting innovative technologies can help the healthcare sector reduce the burden of SSIs significantly, and thus enhance patient outcomes along with the optimization of resource utilization.

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