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Evaluation of Microbial Contamination for Pre- and Post-Surgical Procedures in Hospital Operating Rooms: A Systematic Onsite Sampling

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The risk of surgical site infections (SSIs) is highly associated with the level of microbial contamination settled on a patient's wound. Low or zero microbial contamination could reduce the risk of patients contracting SSIs. To date, the state of microbial contamination in an operating room (OR) for pre- and post-surgery is rarely examined. This exploration is crucial as a patient undergoing surgery tends to contract the SSI if the microbial contamination is high. This study aims to identify the number of microbes present in the OR for pre- and post-surgical procedures. Microbial sampling was performed on 15 ORs in 6 private hospitals located on the western coast of Peninsular Malaysia. Before the microbial sampling, cleanroom performance testing (CPT) was performed to ensure the compliance of operating parameters with the International Organization for Standardization (ISO) Class 7. The CPT was conducted based on ISO 14644-1, Institute of Environmental Science Recommended Practice (IES-RP-CC006.2), and National Environmental Balancing Bureau (NEBB) Procedural Standards for Certified Testing of Cleanrooms. The CPT procedures involved verifying the cleanroom operating parameters: air change rate, differential room pressure, relative humidity (RH), and air temperature. The CPT shows that the air change rate, differential room pressure, RH, and air temperature fell within the recommended operating conditions, ranging from 28/h - 45/h, 6.8 Pa - 22.3 Pa, 51.8 % – 58.9 %, 18.2 °C – 21.6 °C, respectively. The microbes were measured in two phases: pre- and post-surgical procedures. The present study confirms that the microbial contamination for post-surgical procedures increased significantly compared to pre-surgical procedures. The increase in microbial contamination of pre- and post-surgical procedures ranged from 49 to 81 colony-forming units (CFU).

1. Introduction

An operating room (OR) is a specialized and confined healthcare facility that performs surgical procedures on patients (Bali, 2021). It requires a highly sterile and clean environment to minimize patients' SSI risk (Kamar et al., 2015). ORs are designed and maintained to reduce the presence of microbes or bacteria. Strict protocols for infection control, such as disinfection procedures (Guideline, 2015), observing the cleanroom dress code (Hafiani et al., 2022), and utilizing a specialized ventilation approach (Tan et al., 2023), are mandatory. The OR usually accommodates surgeons, assistant surgeons, anaesthesiologists, circulating nurses, and assisting nurses (Kamar et al., 2020). Kamar et al. (2020) also found that occupants positively correlate with the number of microbes identified in an OR.

The microbes in an OR mainly originate from the flakes shed from occupants' skin (Wong et al., 2022). The microbes typically detected include *Methicillin-Resistant Staphylococcus Aureus* (MRSA), *Coagulase-Negative*

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Staphylococci (CoNS), *Pseudomonas aeruginosa*, *Acinetobacter baumannii*, *Klebsiella pneumoniae*, *Escherichia* coli (Tan et al., 2022b). These microbes can cause SSIs when settled on a patient's wound. A previous study reported that approximately 11.4 % of patients who undergo surgical procedures contracted SSIs (Amenu et al., 2011). In 2017, a study identified that the SSI varied from 0.5 % up to 10.1 %, depending on the type of surgical procedure (Guideline, 2019).

Previous studies have only conducted microbial sampling before performing (pre-) surgical procedures in an OR (Tan et al., 2022a). Tan et al. (2022a) examines the correlation between the concentration of microbes and particulate matters (PM) and disclosed that that the PM 5 and PM 10 have a positive correlation with microbes of 7 % and 10 %, respectively. Dreikausen et al, (2023) discovered that the highest number of microbes identified in the air of OR was 9 colony-forming unit (CFU), and 0 microbe was identified on the surface of surgical instrument. Christina et al. (2021) reported that the microbes concentration could be as high as 110 CFU/m^3 and 18 CFU/m^3 during the surgical procedure was conducted and under at rest condition, respectively. To the best of the authors' knowledge, no study has revealed the state of the art of microbial contamination conditions for pre- and post-surgical procedures. The proposed exploration is important to evaluate the suitability of subsequent surgical procedures by considering the risk of the patient contracting an infection. This study aims to develop a systematic procedure for assessing microbial counts under pre- and post-surgical procedures. The samplings of microbes were conducted in 15 ORs that utilize cleanroom technology, rated as ISO Class 7 cleanrooms.

2. Methods

2.1 Details of the operating rooms

In the present study, the 15 ORs belong to 6 private hospitals in Peninsular Malaysia. All 15 ORs are classified as ISO Class 7 cleanrooms designated for general and orthopedic surgeries. Each OR has six air-supply diffusers that are installed with high-efficiency particulate air (HEPA) filters, which could provide clean air to the occupants. Under this filter setting, the delivered air is 99.97 % free of particle size larger than 0.3 μm (Wong et al., 2019). The diffusers are mounted on the ceiling, and the fresh air is supplied uniformly and vertically downwardly. The arrays of diffusers are arranged in the barrier form, which extends by a minimum of 305 mm beyond the footprint on each side of the operating table (Standard, 2008).

2.2 Systematic onsite measurement

This study aims to develop a systematic procedure for conducting onsite measurement according to the following guidelines/ standards: IEST-RP-CC006.2 (Standard, 1997), and NEBB Procedural Standards for Certified Testing of Cleanroom (Standard, 2009). All 15 ORs were purged at least 12 hours before the onsite measurements, and samplings were conducted. All the data were logged under the at-rest condition in compliance with the ISO 14644-1 Standard (1999). Only one person (who conducted the sampling) was allowed in the OR during the onsite measurement. Likewise, the door was closed throughout the measurement procedures. Personnel movements shall be kept as minimal as possible to ensure that the operating parameters are under a steady-state condition. The personnel shall wear a cleanroom attire covering a cleanroom suit, cleanroom boots, surgical mask, cleanroom cap/ hoods (antistatic feature), and nitrite gloves (advisable but not mandatory).

The Alnor EBT 731 Balometer equipped with a flow hood was employed to quantify the airflow rate in the OR. The selected flow hood was made to fit the ceiling-mounted diffusers to ensure air-tight condition adequately. A similar balometer was used to measure the room pressure differential. The accuracy of the balometer on airflow and differential pressure measurements are ±3 % and ±0.025 Pa. The RH and air temperature were quantified using the Testo 625 digital Thermo-Hygrometer (Testo Inc., Lenzkirch, Germany). The measurement accuracy of the thermo-hygrometer on RH and air temperature measurements is ±2.5 % and ±0.5°C. To ensure the reliability of devices, all the measurement devices were calibrated in compliance with the ISO 9001: 2015 standard. The ACH was computed via Eq(1) (Standard, 2009):

$$Air\ change\ rate\ per\ hour\ (ACH) = \frac{Total\ supply\ air\ \left(\frac{m^3}{min}\right) \times 60}{Room\ volume\ (m^3)} \quad (1)$$

The microbial samplings procedures were initiated after verifying that the cleanroom operating parameters fulfilled the ISO Class 7 specification. An active air sampling approach was used to withdraw the surrounding air. The contaminants were settled on the 90-mm diameter Petri Plates filled with Tryptic Soy Agar (TSA) and Saboraud Dextrose Agar (SDA). The TSA was used to obtain the total bacteria concentration, while the SDA was used to determine the total fungal count. All the samples were outsourced for laboratory analysis. The

onsite measurement of airflow rate at the air supply diffuser, room pressure differential, and microbes sampling are shown in Figure 1a, Figure 1b, and Figure 1c, respectively.

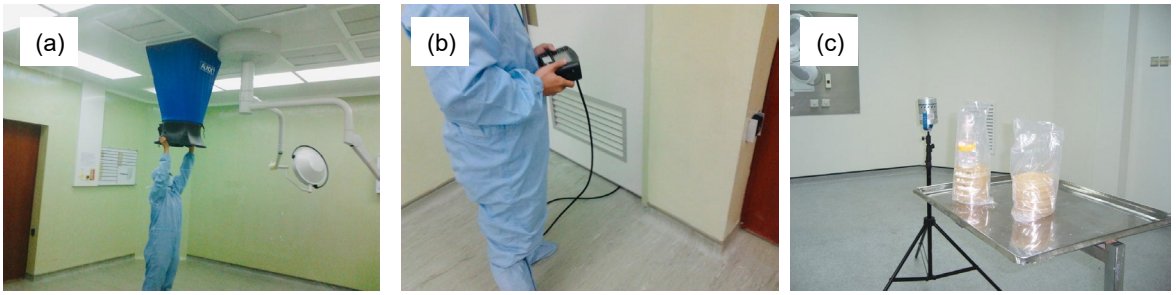

Figure 1: Example of onsite measurement of (a) airflow rate at the air supply diffuser, (b) room pressure differential, (c) microbes sampling using active air sampling approach (Tan et al., 2022a)

3. Results and discussions

The cleanroom parameters were verified before the microbial samplings. The purpose was to ensure that the ORs have achieved good operating conditions as prescribed in ISO class 7 cleanroom specifications. The measured air temperature and RH in 15 ORs are presented in Figures 2a and 2b.

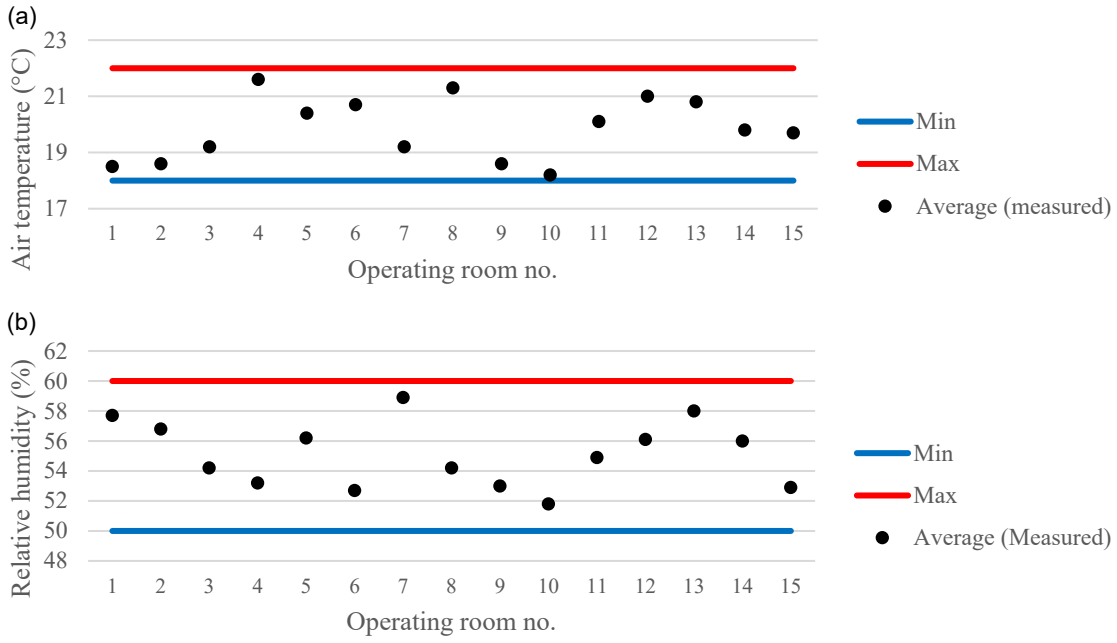

Figure 2: The measured of average (a) air temperature and (b) RH in the 15 ORs

Referring to Figure 2a, the average air temperature in 15 ORs fell within the recommended range of 18 °C and 22 °C. The lowest recorded air temperature was 18.2 °C, while the highest was 21.6 °C. Higher air temperatures above 22 °C could promote the growth of microbes (Qiu et al., 2022). Recently, Qiu et al. (2022) found that an increment of air temperature by 8 °C could elevate microbial growth by 50-60-fold. In contrast, a low air temperature could cause thermal discomfort to the occupants and hypothermia "thermal risk" to the patient.
Figure 2b shows that the RH was within the suggested range of 50 % and 60 %. The lowest RH was identified in OR 10, with an average RH of 51.8 %. The highest RH was observed in OR 7, with an average RH of 58.9 %. A high RH of over 60 % has been reported to profoundly affect microbes' growth rate (Frankel et al., 2012). In contrast, a low RH could cause a greater accumulation of electrostatic charges (Desco, 2013). For instance, walking indoors with 55 % RH could yield a charge of 7.5 kV (Desco, 2013).

Other cleanroom operating parameters, such as air change rate and room pressurization, were below the acceptable range. In an OR, the ACH shall be larger than 20/h (Tan et al., 2022a), while the present study found that the ACH in 15 ORs is from 28/h to 45/h. A low ACH might reduce the effectiveness of contaminant removal and potentially increase the contaminant recirculation in the OR. Subsequently, this scenario could cause a higher contaminant settlement risk for the patient. For the room pressurization, it is suggested that the pressure in the OR shall be larger than 5 Pa compared to the adjacent zones (Tan et al., 2022a). The purpose is to prevent the contaminant at the adjacent zones from penetrating the OR when there is any door opening/ closing activity. In the present study, all ORs have room pressurization in the 6.8 Pa - 22.3 Pa range. The present study verified that all 15 ORs fulfilled the ISO Class 7 specifications. Table 1 shows the microbial counts under pre- and post-surgical procedures. This study sampled two different types of microbes, bacteria and fungi.

Table 1: Number of microbial counts before and after a surgical procedure

Operating room	Parameter	Before (cfu)	After (cfu)	Increment (cfu)
1	Total bacteria count	2	67	65
	Total fungi count	-	-	-
	Total microbial count	2	67	65
2	Total bacteria count	6	57	51
	Total fungi count	-	-	-
	Total microbial count	6	57	51
3	Total bacteria count	7	49	42
	Total fungi count	-	-	-
	Total microbial count	7	49	42
4	Total bacteria count	3	52	49
	Total fungi count	-	-	-
	Total microbial count	3	52	49
5	Total bacteria count	-	62	62
	Total fungi count	-	-	-
	Total microbial count	-	62	62
6	Total bacteria count	-	54	54
	Total fungi count	-	-	-
	Total microbial count	-	54	54
7	Total bacteria count	1	69	68
	Total fungi count	-	-	-
	Total microbial count	1	69	68
8	Total bacteria count	-	81	81
	Total fungi count	-	-	-
	Total microbial count	-	81	81
9	Total bacteria count	5	66	61
	Total fungi count	-	-	-
	Total microbial count	5	66	61
10	Total bacteria count	4	73	69
	Total fungi count	-	-	-
	Total microbial count	4	73	69
11	Total bacteria count	-	49	49
	Total fungi count	-	-	-
	Total microbial count	-	49	49
12	Total bacteria count	2	61	59
	Total fungi count	-	-	-
	Total microbial count	2	61	59
13	Total bacteria count	4	48	44
	Total fungi count	-	-	-
	Total microbial count	4	48	44
14	Total bacteria count	3	77	74
	Total fungi count	-	-	-
	Total microbial count	3	77	74
15	Total bacteria count	1	58	57
	Total fungi count	-	-	-
	Total microbial count	1	58	57

As observed in Table 1, the number of microbes increased significantly after the surgical procedures. Based on the microbial sampling analysis, only bacteria were present in the ORs, while no fungi were detected for pre- and post-surgical procedures. This occurrence could be due to the light nature of bacteria, which ensures these microbes remain airborne and disperse according to the airflow movement. Fungi are heavier in density, less likely to remain airborne, and tend to settle on surfaces faster due to the gravitational force acting on them. Similar findings were reported by Tan et al. (2022a), who performed a correlation study between particulate matter and microbes in the ORs. The microbial measurement for pre- surgery shows good agreement with past study, with the microbe's concentration of less than 10 CFU (Tan et a., 2022a). This concentration is a pre-requisite factor of an OR to be claimed as an ISO Class 7 cleanroom.

Based on the microbes increment column in Table 1, the lowest and highest increments were 42 cfu and 81 cfu. This scenario should be avoided as high microbial counts in ORs could elevate the risk of the patient contracting SSI. According to the policies and procedures on infection control of the Ministry of Health Malaysia (Standard, 2010), the highest allowable counts of bacteria and fungi in an OR shall be less than or equivalent to 35 cfu. The microbe's concentration in some ORs has slightly exceeded the allowable concentration. This concentration (<180CFU/m^3) has been claimed to be an acceptable microbe's level in a working OR (Fu et al., 2018). It is suggested that the investigation could consider the possibility of factors that increase the number of microbes in the ORs, i.e., duration of surgical procedures, number of occupants, ACH in the ORs, and types of activities performed, among others. A recent study claimed that the higher the number of occupants in the OR, the more bacteria are shed into the air and spread with airflow (Andersson et al., 2012). The concentration of microbes in the air also depends on the traffic flow and frequency of doors opening (Wistrand et al., 2021). A higher air change rate and unidirectional airflow supply could reduce the microbes present in the OR via dilution approach (Kek et al., 2023).

4. Conclusions

The article presented a systematic procedure for measuring microbial contamination in 15 ORs under pre- and post-surgical procedures. Before the onsite sampling work, the 15 ORs are verified to operate under the cleanroom specifications as stated in the policies by the Ministry of Health Malaysia. The ACH for all ORs lies within 29/h- 45/h, which fulfilled the minimum requirement of more than 20/h. The relative humidity lies within the range of 51.8 % to 58.9 % and meets the 50 % to 60 % requirement. Likewise, the air temperature in the ORs was in the suggested range, which falls between 18.2 °C and 21.6 °C. The present study confirms that microbial contamination for post-surgical procedures increased significantly compared to pre-surgical procedures. The increment of microbial contamination of pre- and post-surgical procedures ranged from 42 to 81 cfu. Such an increment also indicated that the risk of patient contracting the SSI would be higher when the duration of surgical procedure is getting longer. Appropriate engineering approaches such as ventilation strategies, air change rate, and airflow settings could be an interesting area to be explored. In future studies, the authors recommend conducting microbial samplings in pre-, during, and post-surgery (under "operational" conditions) to reflect real-time contamination in the ORs. Likewise, a larger sample size is suggested to obtain more reliable data to reduce the margin of error.

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