Original Research Article

Microbiological surveillance of operation theatres, intensive care units and labor room of a teaching hospital in Telangana, India

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ABSTRACT

Background: Hospital-associated infections are the major cause of patient morbidity and mortality. Environmental monitoring by the microbiological testing of surfaces and equipments is useful to detect changing trends of types and counts of microbial flora. The aims of the study were to count CFU (colony forming unit) rate of indoor air, to identify bacterial colonization of surface and equipments isolated from Operation theatres, ICUs and Labor room of a teaching hospital in Telangana, India.

Methods: Air sampling of 10 OT's, 4 ICU's and 1 LR were done by settle plate method. Swabs were taken from different sites and equipments and bacterial species were isolated and identified from them.

Results: Gynaecology OT-2 and 3 showed less bacterial CFU rate of air (6 CFU/m^3) followed by Opthamology OT (16 CFU/m^3) and highest in Orthopaedics minor OT (72 CFU/m^3) and Dental OT (42 CFU/m^3 with 1 fungal CFU/m^3). Pediatric ICU showed less bacterial CFU rate (28 CFU/m^3) followed by Surgery ICU (40 CFU/m^3) and highest in Medical ICU (100 CFU/m^3 and 5 fungal CFU/m^3). Labor room showed 65 CFU/m3. Bacterial species were isolated from 48 (43%) out of total 111 swab samples taken from all OTs and ICUs. The most common isolate was Bacillus species 36 (75%) followed by micrococcus (54%). Pathogenic organisms isolated were 12 Gram negative bacilli and 6 gram positive cocci, the common isolate was *Klebsiella spp* (7 of 12).

Conclusions: The study shows that OTs were having bacterial CFU rate of air varying from 6-72 CFU/m^3 and colonized by contaminants like *Bacillus sp* and pathogens like *Klebsiella sp*. ICUs were having bacterial CFU rate of air varying from 28-100 CFU/m^3 and colonized with contaminant like Bacillus sp., as well as potential pathogens like *Klebsiella, Pseudomonas* etc. Fungal CFU were also seen both in OTs and ICUs. High level of microbial contamination indicates the needs for periodic surveillance aimed at early detection of bacterial contamination levels and prevention of hospital acquired infections.

Keywords: Air sampling, Bacterial colonization, ICU, LR, OT, Settle plate method, Surveillance

INTRODUCTION

Microbial contamination of hospital environment, especially in an operating theatre and other specialized units had continued to increase prevalence of nosocomial infections.[1-3.] Operation Theatre (OT) and Intensive care unit (ICU) acquired infections, which are often caused by antibiotic resistant bacteria poses a significant threat to patients. Hospital-associated infections are the major cause of patient morbidity and mortality.[4] Invasive procedures, high antibiotic usage and transmission of bacteria between patients due to inadequate infection control measures may explain why OTs and ICUs are "hot zones" for the emergence and spread of microbial resistance.[5] Environmental monitoring by the microbiological testing of surfaces and equipments is useful to detect changing trends of types and counts of microbial flora.[6]

Biological contaminants occur in the air as aerosols and may include bacteria, fungi, viruses, and pollens. Microbiologic air sampling is used as needed to determine the numbers and types of microorganisms, or particulates, in indoor air.[7]

Controlling airborne pathogens in health facilities is not only important for the safety of the patient, but it is also important for hospital.[8] There is a clear need for surveillance and early warning systems that can pick up signs of emerging and/or increasing microbial resistance at the local, regional and national level 5 "Microbiological surveillance" provides data about the factors contributing to infection.

Evaluation of the quality of air in operating theatres can be performed routinely by microbiological sampling and particle counting.[9,10]

Total numbers of bacteria in an empty operating theatre should be <35 cfu/m^3 with less than one colony of *Clostridium perfringens* or *S. aureus*.[11] During an operation, total air counts should be < 180 cfu/m^3.[7,11]

The present study is focused on to evaluate the level of bacterial contamination in operating theaters of hospital, intensive care units and labour room.

METHODS

The study was conducted in the department of Microbiology, Mediciti Medical College, Ghanpur, Hyderabad, Telangana, India. The theatre being sampled have been left vacant for more than one hour before sampling proceeds to avoid false-positive results due to recent theatre usage. The theatre doors are kept closed prior to and during the sampling period.

Air sampling (settle plate method)

Settle plate method was used for sampling of air by using blood agar plates from 10 OTs, 4 ICUs and 1 LR of teaching hospital in Hyderabad, Telangana, India. Blood agar plates were kept open in the OTs and ICUs at a height of 1m and 1m from the walls of the rooms. Sampling time was 1hour.

The microbial air contamination was measured by counting the number of CFU (Colony Forming Unit) per cubic meter (CFU/ m^3) of air.

Samples from surfaces and equipments (swab method)

Total 111 samples were taken from different sites and equipments of 10 OTs, 4 ICUs and 1 LR. Sterile swabs in nutrient broth were used to collect samples.

They were placed back into the broth after collection. All the samples were labeled properly and immediately transported to the Microbiology laboratory and incubated

37°C for 4 hours. Swabs taken from different sites were inoculated on Blood agar and MacConkey agar. These culture plates were incubated at 37°C under aerobic condition for 24 hours.

Isolation and identification of isolates were done as per standard guidelines. All isolates were divided in to three broad categories: 1) Normal flora e.g. Coagulase Negative *Staphylococcus* (CONS) 2) Contaminant e. g. *Bacillus sp*. 3) Pathogen e. g. *Klebsiella sp.*

RESULTS

Bacterial species were isolated from 48 (43%) out of total 111 swab samples taken from all OTs and ICUs.

The bacterial CFU/m^3 counts of air from all OTs ranged from 6 CFU/m^3 air, which was isolated from Gynacology OT-2 and 3, to 72 CFU/m^3 from Orthopaedic minor OT.

The bacterial CFU/m^3 counts of air from all ICUs ranged from 28 CFU/m^3 air, which was isolated from Pediatric ICU, to 100 CFU/m^3 from Medical ICU. Labour room shows the bacterial count as 65 CFU/m^3 as shown in Table 1.

Table 1: CFU rate of air in OTs, ICU and LR (Settle plate method).

OTs	Bacterial CFU	Fungal CFU
Gynaecology OT-1	21	1
Gynaecology OT-2	6	
Gynaecology OT-3	6	
CT & urology OT	23	
Ortho minor	72	
Ortho major	32	
General surgery major	61	
ENT/Dental	42	1
Ophthalmology	16	
General surgery minor	26	
ICUs		
NICU	45	
PICU	28	1
MICU	100	5
SICU	40	
LR	65	

Almost all OTs were found free from pathogens in swab sampling. Only 4 (40%) OTs harbored pathogens like *Klebsiella, E.coli, Enterobacter* etc. All 5 ICU harbored different bacterial species in swab sampling as shown in Table 2. Tables, Beds, trolley and BP apparatus were found to be more contaminated as shown in Figure 1.

Bacillus sp. 45% (contaminants) and micrococci 33% (normal flora) were most common isolates followed by Klebsiella (9%) (pathogens) as shown in Figure 2.

Table 2: Bacterial isolates from OTs, LR and ICUs.

| OTs	Bacterial species isolated
Gynaecology OT-1	*Klebsiella sp., Micrococci, Bacillus sp.,*
Gynaecology OT-2	*E.coli, Micrococci, Bacillus sp.,*
Gynaecology OT-3	*Klebsiella sp., Enterobacter sp., CONS, Micrococci*
CT & urology OT	*Micrococci, Bacillus sp.,*
Ortho minor	*Bacillus sp.,*
Ortho major	*Bacillus sp.,*
General surgery major	*Bacillus sp.,*
ENT/Dental	*Micrococci, Bacillus sp.,*
Ophthalmology	*CONS, Micrococci*
General surgery minor	*Klebsiella sp., CONS*
ICUs	
NICU	*Citrobacter, Pseudomonas sp., CONS, Bacillus sp.,*
PICU	*Micrococci, Bacillus sp.,*
MICU	*Klebsiella sp., Micrococci, Bacillus sp.,*
SICU	*Klebsiella sp., Pseudomonas sp., CONS, Bacillus sp.,*
LR	*CONS, Micrococci, Bacillus sp.,*
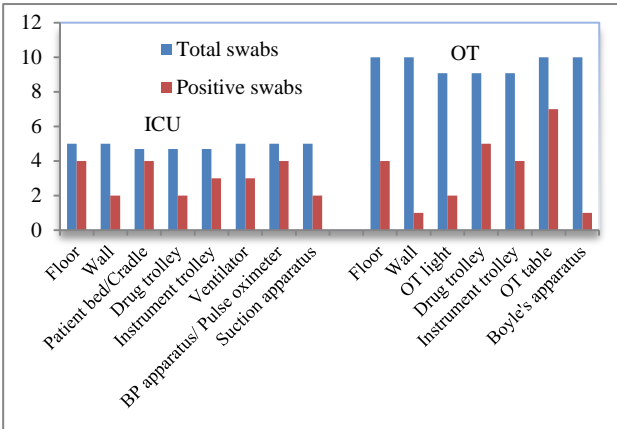
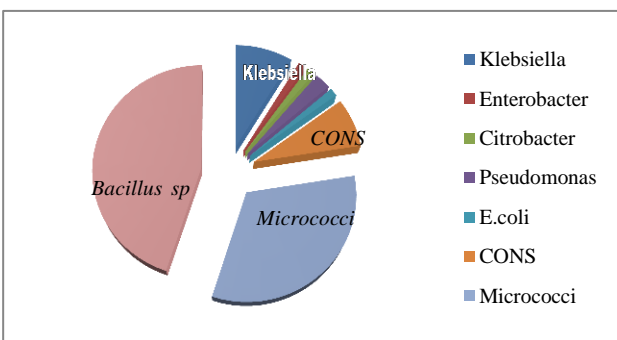**Figure 1: Swab sampling.**

Figure 2: Organisms isolated.

Total 18 potential pathogens were isolated from OTs, ICUs & LR, which included 7 *Klebsiella spp,* 6 Coagulase negative *Staphylococcus,* 2 *Pseudomonas spp,* 1 *E.coli,* 1 *Citrobacter sp* and 1 *Enterobacter sp.*

DISCUSSION

Aerobic cultures on non-selective medium should not exceed 35 colony-forming units (CFU) of bacteria and fungi per cubic meter of air for a conventional theatre and 1 cfu for an ultraclean theatre.[15] 12 All most all OTs and all ICUs and LR are exceeding the limit point in the study. Only some OTs like Gynaecology OT and Ophthalmology OT where the strict fumigation is followed daily, are not exceeding the limit. Anjali K, et al, Rumana Farooq Mir et al, etc also reported in their study that Opthamology OT was the one with least contamination.[13,14,17,18]

Bacterial air sampling in all OTs showed 6-72 CFU/m^3 most of them are between 23-42 CFU/m^3, similar results, 21-41 CFU/m^3 have been reported by Qudiesat K et al.[8] In this study, 100 CFU/m^3 are shown in MICU similar to the study by Shresta CD et al, that they reported that number of colonies of the bacteria ranged from 50 to 140 colonies in MICU and SHCU. 16 ICUs are showing heavy contamination because the fumigation procedure is not done frequently because of the presence of the patients day and night.

The bacterial pathogens were isolated comprising of Klebsiella, coagulase negative *Staphylococci spp, Pseudomonas* etc, had the highest percentage of occurrence in swab samples while in settle plate samples *Bacillus spp* and *micrococci spp*., showed highest percentage of occurrence. In study by Anjali et al.

The most common isolate was Coagulase negative *Staphylococcus species* (5.8%) followed by *Bacillus* and *Klebsiella* species (4.4%) each.[13,17] In the study by Desai SN, The most common isolate was Bacillus species 34 (91.9%) followed by S. aureus 7 (18.9%).[13,15]

Out of 10 OTs 45% were found to be colonized with contaminant Bacillus species. Similar result has been reported by Javed I et al and Sharma D et al.[4,12,16] All ICUs (100%) were observed colonized with contaminants as well as potential pathogens. OT table and drug trolleys are the most contaminated sites in OTs and beds, BP apparatus and floors are the most contaminated sites in ICUs similar to floors and tables as in study of Desai SN et al.[13,15]

According to Pasquarella et al microbiological quality of air may be considered as mirror of the hygienic condition of the operation theatres.[14,17] Surveillance of OTs and ICUs with infection control measures is helpful in controlling nosocomial infections. However, evaluation of cleaning, disinfection and sterilization methods is necessary for monitoring and to set policies. Frequency

of surveillance depends on number and type of surgeries, outbreak of post-operative infection and availability of resources.

CONCLUSION

Harboring of potential pathogens in OTs and ICUs of hospital can pose a great risk to patients. High level of microbial contamination indicates the needs for periodic surveillance aimed at early detection of bacterial contamination levels and prevention of hospital acquired infections.

Correlation between microbial burden and SSI by any method is not absolute and it indicates only the source of post-operative infection. Bacterial air count is a suitable index in predicting post-operative infections but not cost effective.

Though settle plate method may be regarded as a crude measure of airborne contamination, in places without other facilities it can still provide a simple and cost effective way of enumerating the contamination rate of horizontal surfaces at multiple points. So, settle plate method and swab method will be helpful in predicting the microbial contamination.

The quality of indoor air depends on external and internal sources such as ventilation, cleaning procedures, the surgical team and their activities. So the preventive measure may be achieved by making improvement in cleaning and by using disinfectants and needs periodic fumigation of these OTs. Newer less toxic disinfectants are alternative to formalin.

Moreover fogging cannot replace manual cleaning. Since human activity plays a major role in microbial air quality, meticulous cleaning and strict adherence to OT protocol are essential. Routine surveillance for any OT may be suggested for every two months and for septic OT every month. To prevent any contamination prior HAI develops, hospital needs to develop programmes for the implementation of good infection control practices.

REFERENCES

1. Bhalla A, Drin, D, Donskey CJ, *Staphylococcus aureus* intestinal colonization in associated with increased frequency of S. aureus on skin of hospitalized patients BMC. Infect Dis, 2007;7(105):108-23.
2. Bonten MJM, Hayden MK, Nathan C, Epidemiology of colonization of patient and environment with vancomycin-resistant *enterococci*. Lancet. 1996;348:1615-9.
3. Boyce JM, Potter-Byno, Chenevert C, King T, Environmental contamination due to methicillin-resistant S. aureus; possible infection control implication, Infect Control Hosp. Epidemiol. 1997;18:622-7.
4. Javed I, Hafeez R, Zubair M, Anwar M, Tayyib M, Husnain S. Microbiological surveillance of operation theatres and ICUs of a teaching hospital, Lahore. Biomedica. 2008;24:99-102.
5. Hanberger H, Arman D, Gill H, Jindrak V, Kalenic S, Kurcz A, et al. Surveillance of microbial resistance in European Intensive care Units: A first report from the- ICU program for improved infection control. Available at: http://liu.diva-portal.org/ smash/ get/ diva2:15024/ FULLTEXT01.pdf.
6. Sandle T. Environmental Monitoring Risk Assessment. Journal of GXP Compliance. 2006;10(2).
7. Guidelines for Environmental Infection Control in Health-Care Facilities. 2003.
8. Qudiesat K, Abu-Elteen K, Elkarmi A, Hamad M, Abussaud M. Assessment of airborne pathogens in health settings. African J Microbio Res. 2009;3(2):66-76.
9. Chacko I, Jose S, Isa A, Bhat KG. Survival of Nosocomial Bacteria in Hospital Fabrics. Ind. J. Med Microb. 2003;21(4):291.
10. Javed I, Hafeez, R, Zubair M, Anwar MS, Husnain S. Microbiological surveillance of operation theatres and ICUs of a tertiary hospital, Lahore Biomedica. 2008;24:99-102.
11. Zerr DM, Garrision MM, Allpress AL, Heath J, Christakin DA, Infection Control Policies and Hospital-Associated Infection among Surgical Patient; Variability or Association in a Multicentre Peadiatric Setting. Peadiatrics. 2005; 4:387-92.
12. Streifel AJ. Design and Maintenance of Hospital Ventilation Systems and the Prevention of Airborne Nosocomial Infections. In Mayhall CG (ed) Hospital Epidemiology and Infection Control. Philadelphia: Lippincott, Williams & Wilkins. 1999:1211-21.
13. Anjali K. Environmental Microbiological Surveillance of Operation Theatres in a Tertiary Care Hospital, Int J Cur Res. 2015;7(03):13977-80.
14. Mir RF, Singh VA, Shinu P. Pre and post fumigation bacteriological profile of various operation theatres in MMIMSR – a three year retrospective study, J Pharmaceu Biomed Sci. 2013;36(36):1887-91.
15. Desai SN, Kikani KM, Mehta SJ. Microbilogical Surveillance of Operation Theaters and Intensive Care Units of Teaching Hospital in Surendranagar, Gujarat. Gujarat Med J. GMJ. 2012;67(2):95.
16. Sharma D, Nagarajan S. A study of cleaning/ disinfecting procedures in a primary tertiary care hospital, Delhi. Health and Population Perspectives And Issues. 2001;24(4):189-97.

17. Pasquarella C, Masia MD, Nnanga N, Sansebastiano GE, Savino A, Signorelli C, et al. Microbial air monitoring in operating theatre: active and passive samplings, Ann Ig. 2004;16(1-2):375-86.