Original Research Article

Microbiological surveillance of operation theatre’s and intensive care units in a tertiary care hospital in NCR region, New Delhi

Soumya Singh¹, Rohit Kumar²*, Mrinmoy Sarma³

¹Department of Microbiology, SCPM College of Nursing and Paramedical Sciences, Lucknow, Uttar Pradesh, India
²Department of Microbiology, Kalpana Chawla Govt. Medical College & Hospital, Karnal, Haryana, India
³Department of Microbiology, Rishiraj College of Dental Sciences and Research, Bhopal, Madhya Pradesh, India

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*Correspondence:
Dr. Rohit Kumar,
E-mail: rk95rk@gmail.com

ABSTRACT

Background: Harbouring of potential pathogens in operation theatres (OTs) and intensive care units (ICUs) of hospital is a major cause of patient’s 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. 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.

Methods: During a period of 6 months from January 2019 to June 2019, Air sampling from Operation theaters and Intensive care units were done by settle plate method. Swabs were taken from different sites and equipments and bacterial species were isolated and identified from them.

Results: A total of 1410 samples were collected from various sites of Operation theaters and Intensive care units over a period of 6 months in which 960 were surface samples and 450 were air samples. Out of 960 surface samples, 95(9.89%) and out of 450 air samples, 90 (20%) were bacterial positive. Isolated organism was divided into normal flora (CONS, Micrococci), contaminant (bacillus species) and pathogenic organism e.g. Staphylococcus aureus, Acinetobacter spp., Pseudomonas spp etc. Out of those 30 (16.20%) CONS, 50 (27.02%) Micrococci, 75 (40.50%) Bacillus spp, 16 (8.6%) Staphylococcus aureus, 10 (5.4%) Acinetobacter spp, 2 (1.08%) Pseudomonas spp, 4 (2.16%) Klebsiella, 7 (3.78%) Escherichia coli were isolated.

Conclusions: Strengthening surveillance and laboratory capacity will surely enhance infection prevention and control. Routine sampling is strongly recommended for increasing awareness to identify and control all possible sources and types of infections.

Keywords: Microbiological, Operation theater, Surgical site infection, Surveillance

INTRODUCTION

Hospital-associated infections are an important source of morbidity and mortality with postoperative, surgical site infections (SSI) being the second most common cause after urinary tract infections.¹,²

Surgical-site infection is the leading complication of surgery. Normal skin flora of patients or healthcare workers causes more than half all infections following clean surgery, but the importance of airborne bacteria in this setting remains controversial.³
Hospital acquired infections (HAIs) prolong hospital stays, create long-term disability, increase resistance to antimicrobials, represent a massive additional financial burden for health systems and cause unnecessary deaths.

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.  

Sources of infection can either be endogenous or exogenous from the theatre environment like air, surfaces, and articles in operation theatre (OT). So the preventive measure may be achieved by making improvement in cleaning by using disinfectants, needs periodic fumigation of these OTs and with routine microbial surveillance.

Microbiological surveillance” provides data about the factors contributing to infection. Environmental monitoring by the microbiological testing of surfaces and equipments is useful to detect changing trends of types and counts of microbial flora. Evaluation of the quality of air in operating theatres can be performed routinely by microbiological sampling and particle counting. The quality of indoor air depends on external and internal sources such as ventilation, cleaning procedures, the surgical team and their activities.

The present study was conducted to identify bacterial colonization of surfaces and equipment in the OTs and to determine the microbial contamination of air in the OTs of a tertiary care hospital.

**METHODS**

Prospective observational study was conducted in the Department of Microbiology, Santosh Medical College and Hospital, Ghaziabad, Uttar Pradesh, India for a period of 6 months from January 2019 to June 2019. A total number of 1410 samples from different sites of Operation theatres and ICU’s were enrolled in the study.

**Inclusion criteria**

Samples from Surgery OT, Gynaecology OT, Major OT, ENT, Ophthalmology, ICU, NICU, PICU, MICU and SICU.

**Exclusion criteria**

Operation theatre and ICU’s where septic cases were operated.

**Methods**

The operation theatre being sampled was left vacant for more than 1 hour before sampling proceeds to avoid false positive results due to recent theatre usage. The theatres were kept closed prior to and during the sampling period.

**Collection and transport of sample**

Air and Surface samples were taken from Surgery OT, Gynaecology OT, Major OT, ENT, Ophthalmology, ICU, NICU, PICU, MICU and SICU. Blood agar plates and sterile swabs were transported to Operation theatres in sealed plastic bags.

- Settle plate method (for monitoring quality of air)
- Surface swabbing.

**Air sampling**

Air sampling was done by settle plate method. Blood agar was placed at four locations in OT’s and ICU’s one meter above ground, one meter away from the wall for one hour. These were then transported to laboratory and incubated at 37°C for 24 hours under aerobic conditions. After incubation, plates were observed for growth. Isolates were then identified using standard microbiological procedures and CFU/m³ were estimated using Omeliansky formula according to which:

\[
CFU / m^3 = a \times 1000 / p \times t \times 0.2
\]

a = no of colonies on the Petri plates,  
p = the surface measurement of the plate used in cm²  
t = the time of the exposure of the Petri plates in minutes.

**Recommended conventional operation theatre values**

- An empty theatre bio load should not exceed 35 CFU/m³.  
- During surgery bio load should not exceed 180 CFU/m³.

**Surface sampling**

Moistened sterile cotton swabs in nutrient broth were used to collect samples from different sites like operation table, autoclaved instruments, drug trolley, walls, floor and light. All samples were properly labeled and immediately transported to microbiology laboratory. Swabs taken from different sites were inoculated on blood agar & MacConkey agar plates. These were incubated at 37°C for 18-24hrs under aerobic conditions. After incubation the isolates were identified by colony characteristics, gram reaction and standard biochemical tests.

**Statistical analysis**

Univariate analysis was carried out. Chi-square test was used for categorical variables (p<0.05 was considered
significant) and student t-test was carried out for quantitative variables.

RESULTS

A total of 1410 samples were collected from various sites of Operation theatres and Intensive care units from Department of Microbiology, Santosh Medical College and Hospital, Ghaziabad, Uttar Pradesh, India for a period of 6 months from July 2018 to December 2018. Out of total 1410 samples, 960 were surface samples and 450 were air samples.

Out of 960 surface samples 95 (9.89%) were positive for culture and Out of 450 air samples, 90 (20%) were positive for culture.

Out of 960 surface samples, 60 samples were taken from Ophthalmology, 120 from ENT, 120 from surgery OT, 180 from Gynaecology OT, 60 from Major OT, 60 from ICU, 60 from NICU, 90 from PICU, 90 from MICU and 60 samples were taken from SICU (Figure 1).

Out of 450 air samples (settle plate samples), 30 samples were taken from Ophthalmology, 60 from ENT, 60 from surgery OT, 90 from Gynaecology OT, 30 from Major OT, 60 from ICU, 30 from NICU, 30 from PICU, 30 from MICU and 30 samples were taken from SICU (Figure 2).

Total 960 samples were taken from different sites and equipments of OTs and ICUs. In OTs, 60 from Pt. Bed, 45 samples from floors, 180 from wall, 60 from OT lights, 50 from drug trolley, 50 from instrument trolley, 45 from OT table and 50 from anaesthesia tables, Whereas in ICUs, 25 samples from floor, 100 from wall, 80 from patient bed, 45 from drug, 45 from instrument trolley, 45 from ventilator, 25 from suction apparatus and 30 from BP apparatus and 25 from door handles (Figure 3).

![Figure 1: Department wise distribution of surface samples.](image1)

![Figure 2: Department wise distribution of air samples.](image2)

![Figure 3: Distribution of samples based on surface tested.](image3)

![Figure 4: Distribution of isolated gram negative bacilli and gram positive bacteria.](image4)

### Figures:
- **Figure 1:** Department wise distribution of surface samples.
- **Figure 2:** Department wise distribution of air samples.
- **Figure 3:** Distribution of samples based on surface tested.
- **Figure 4:** Distribution of isolated gram negative bacilli and gram positive bacteria.
Out of 960 surface samples, 95 (9.89%) and out of 450 air samples, 90 (20%) were bacterial positive. Isolated organism was divided into normal flora (CONS, Micrococi), contaminant (bacillus species) and pathogenic organism e.g. *Staphylococcus aureus*, *Acinetobacter spp.*, *Pseudomonas spp.* etc. Out of those 30 (16.20%) CONS, 50 (27.02%) Micrococi, 75 (40.50%) *Bacillus spp.*, 16 (8.6%) *Staphylococcus aureus*, 1 (0.54%) *Acinetobacter spp.*, 2 (1.08%) *Pseudomonas spp.*, 4 (2.16%) *Klebsiella*, 7 (3.78%) *Escherichia coli* were isolated (Figure 4).

In gram negative bacteria, the sensitive pattern of bacterial isolates was ciprofloxacin (65%), cotrimoxazole (80%), amikacin (88%), meropenem (40%), ceftazidime (35%), piperacillin-tazobactam (75%), levofloxacin (78%) and whereas the higher resistant shown by ceftazidime (65%) and meropenem (60%) (Figure 5).

### DISCUSSION

Microbial contamination in OT leading to postoperative infections can have serious implications for patients and their families. Any case of suspected hospital-acquired infection (HAI) is investigated by including cultures from other body sites of the patient, other patients, staff, and environment.12

Careful selection of specimens to be cultured is essential to obtain meaningful data. Infections prolong hospital stays, create long-term disability, increase resistance to antimicrobials, represent a massive additional financial burden for health systems and cause unnecessary deaths.

Thus, the solution is a well-implemented infection control program which can improve staff education and accountability, also by conducting research to adapt and validate surveillance protocols based on the reality of developing countries to achieve acceptable performance.
This can reduce the incidence of HAIs by around one-third.13

Aerobic cultures on non-selective medium should not exceed 35 colonies forming units (CFU) per cubic meter of air in an empty operation theatre and 180 CFU per cubic meter of air during an operation for conventional theatre. Microbiological contamination of air in the operating room is generally considered to be risk factor for surgical site infections in clean surgery.14

According Pasquarella et al microbiological quality of air may be considered as mirror of the hygienic condition of the operation theatres.15 The quality of indoor air depends on external and internal sources, such as ventilation, cleaning procedures, the surgical team and their activity.

In the present study, Bacterial Colony Forming Unit was ranged between 121-162 CFU/mm² from different Operation theatres and 124-151 CFU/mm² from different Intensive Care Units which was similar to study done by Kiranmai S, Madhvi K et al.16 The bacterial CFU was ranged between 17-82 CFU/mm² from OT's and 31-200 CFU/mm² respectively. While other study conducted by Javed I et al showed higher ranged between 6500 -5730 CFU/mm² from OT’s and 628-1571 CFU/mm³.17

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 method for air sampling showed highest percentage of occurrence of Micrococci and Bacillus spp. followed by CONS while in the study done by Anjali et al.18

Moreover, among the pathogens Staphylococcus aureus and Acinetobacter spp. were isolated, in a study done by Qudiesat et al.18 Staphylococcus aureus was isolated while in a study done by Kiranmai et al E.coli, Klebsiella spp. and Enterobacter spp. was isolated from air sampling.16

CONS are considered as an exogenous organism. Source of CONS in the study can be normal skin flora of medical personnel, patients and fabrics.18 S. aureus was isolated in a study done by Qudiesat et al, Desai SN, and Yadav M et al.3,18

In our study surface of Gynaecology OT were found higher pathogenic bacteria, Escherichia coli 7(3.78%), Klebsiella 4(2.16%) and Pseudomonas 2(1.08%) were isolated.

Other study showed bacterial species were isolated 15(23.4%) out of total 64 swabs samples taken from all operation theatre and ICU’s, CONS (6.25%) and Micrococci (6.25%) (Both normal flora) are the predominant isolated followed by bacillus (4.6%). Pathogenic bacteria such as Acinetobacter species (31.2%), Enterobacteriaceae species (1.56%) and Pseudomonas (1.56%) were also isolated in study conducted by Anjali et al.17

Out of 10 ICUs and OTs, 46.51% were found to be colonized with contaminant Bacillus species. Similar result has been reported by Javed I et al and Sharma D et al.9,15 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 et al.17

In Gram negative bacteria, the sensitive pattern of Bacterial isolates were ciprofloxacin (65%), cotrimoxazole (80%), amikacin (88%), meropenem (40%), ceftazidime (35%), piperacillin-tazobactam (75%), levofloxacin (78%) and whereas the higher resistant shown by ceftazidime (65%) and meropenem (60%). Similar study conducted by Anjali et al which has found 69% meropenem resistant.17

In gram positive bacteria, the sensitive pattern of bacterial isolates was erythromycin (26%), clindamycin (78%), amikacin (88%), penicillin (10%), levofloxacin (74%) and whereas the higher resistant shown by penicillin (90%) and erythromycin (74%). All bacterial isolates from air samples were 100 % sensitive for linezolid and vancomycin.

This study has several limitations. There is changing tend towards GNB and GPC isolation. It may be due to their ability to survive in adverse conditions. It may also be due to lack of proper disinfection of fumigation of facilities, overcrowding and unnecessary visiting of critical care facilities by people or due to improper ventilation of OTs. 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.

**CONCLUSION**

Strengthening surveillance and laboratory capacity will surely enhance infection prevention and control. Routine sampling is strongly recommended for increasing awareness to identify and control all possible sources and types of infections. Settle plate’s method for air and swabbing technique for surfaces are considered as crude methods but in a limited resource setup these methods are proved to be more valuable in detecting the contamination level. 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
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