

# Nosocomial infection causing bacteria identification in tertiary care hospital of saidu sharif, Swat, Khyber pakhtunkhwa, Pakistan

## Abstract

**Introduction:** Nosocomial infection is global problem and more severe in under-developed countries. The chances of these infections become more increased in the immune compromised patients that may cause mortality or morbidity. The aim of the current study was isolation of bacterial isolates associated with Nosocomial infections from tertiary care hospital Saidu Sharif, Swat, Pakistan.

**Methods:** The study was conducted in the tertiary care hospital Saidu Sharif, Swat, Pakistan from August-December 2017. The samples were collected from medical, surgical, gynea wards and operation theater environment of the hospital. On the basis of colony morphology and biochemical test total eleven different bacterial isolates were identified.

**Results:** These isolates are *Proteus vulgaris*, *Bacillus cereus*, *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Bacillus subtilis*, *Escherichia coli*, *Klebsiella pneumonia*, *Streptococcus pyogenes*, *Micrococcus luteus*, and *Rhodococcus equi*. The data revealed that *Pseudomonas aeruginosa* is present in maximum amount in floor, wall, bed sheet, instruments, and hands before and after dressing, followed by *Staphylococcus aureus* present in floor, wall, bed sheet, instruments, before and after nurse hands.

**Conclusions:** Hygienic sanitation of the hospital and improve the hospital equipment and public awareness about nosocomial infection is needed.

**Keywords:** Nosocomial, Hygiene, Infection, Pakistan

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## Introduction

Nosocomial infection is that infection which acquired during the hospital admission. According to the World Health Organization (WHO) nosocomial infection is an infection that was found in hospitalize patient, whom the infection was not present or incubates at the time of entrance. It is global problem and more severe in under-developed countries. The chances of these infections become more increased in the immune compromised patients that may cause mortality or morbidity.<sup>1,2</sup> The risk of nosocomial infection is linked to the hygienic conditions of hospital, use of antibiotics for longer time and immune conditions of the patient.<sup>3,4</sup>

The hospital acquired infections are caused by different pathogens such as bacteria, fungi, viruses and parasites. The most common bacterial pathogens associated with hospital acquired infection are; *E. coli*, *P. aeruginosa*, *S. pneumonia*, *S. aureus*, *K. pneumonia*, *B. cereus*. These pathogens cause nosocomial infections in different ways like endogenous infection, cross infection, environmental infection.<sup>5</sup>

Prevention of nosocomial infection is challenge for both developed and under developing countries.<sup>6</sup> It can be prevented after cutting the way of transmission and remission of the source of transmission from the hospital. The source of nosocomial infection can be minimized by hygienic sanitation, proper waste management and use of sterilized medical and surgical instruments.<sup>7</sup>

Therefore, the present study was conducted to isolate and identify the bacteria, which are likely to be caused nosocomial infections in the tertiary care hospital of Saidu Sharif, Swat, Pakistan.

## Materials and methods

### Sample collection

The study was conducted in the tertiary care hospital Saidusharif, Swat, Pakistan from August-December 2017. The samples were collected from medical, surgical, gynea wards and operation theater environment of the hospital. The samples were taken through sterilized swabs from different environments of the ward. The samples were collected from ward floor and ward walls, ward washroom floor and walls, patient beds of wards, Nurse Hands before and after attending the patients, ward trays, medical and surgical instruments. The sample were labeled and brought to the laboratory of Center for biotechnology and Microbiology University of Swat, Pakistan and stored at 4°C before further processing.

### Media and culturing

Nutrient agar, blood agar media and MacConky agar medium were prepared using the standard laboratory protocol. Broad spectrum antifungal i.e., Naystatin were added to at 1ml each per liter of media for the inhibition of fungal growth. The media were poured to the sterilized petri plates in Laminar flow hood (L.F.H). The plates were incubated at room temperature for 24 hour to check and discarded the contaminated plates. The labeled swab samples were inoculated on the uncontaminated petri plates. The plates were incubated for 24 hours at 37°C. After the period of incubation the plates were examined and different appeared colonies were marked based on colony morphology. The marked colonies were sub-cultured on the fresh nutrient agar medium and incubated for 24 hours at 37°C. These

plates were observed and the appeared colonies were marked on the basis of colony morphology. The marked colonies were again sub-cultured on the fresh media plates to obtain the pure culture.

### Identification of the pure cultures

Gram staining and different bio chemical tests were used to identify the pure cultures. These biochemical tests includes; Catalase, Coagulase, Oxidase, Urease, Indole, Triple Sugar Iron (TSI) and Citrate tests.

## Results

### Isolation of bacteria from medical ward

The data revealed that various types of bacterial isolates i.e. *B. cereus*, *P. aeruginosa*, *S. epidermidis*, *E. coli*, *S. aureus*, *B. subtilis* present in different places of the ward. Maximum numbers of bacterial isolates were *P. aeruginosa* and *S. aureus* isolates from different places. Followed *B. subtilis* and *B. cereus* was isolated only of two places (Table 1).

**Table 1** Isolation of bacteria from Medical wards

Area		Bacterial Isolates
Ward	Floor	<i>Bacillus cirrus</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus epidermidis</i>
	Wall	<i>Staphylococcus aureus</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus epidermidis</i>
Wash room	Floor	<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus aureus</i>
	Wall	<i>Bacillus subtilis</i>
		<i>Staphylococcus aureus</i>
Ward bed	Bed sheet	<i>Bacillus subtilis</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus aureus</i>
		<i>Escherichia coli</i>
Instruments	Stethoscope, Catheter, syringe	<i>Bacillus cirrus</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus aureus</i>
Hands	From nurse hand before and after dressing	<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus aureus</i>
		<i>Escherichia coli</i>
		<i>Pseudomonas aeruginosa</i>
Ward Dust ben	Dust ben	<i>Staphylococcus aureus</i>
		<i>Escherichia coli</i>
		<i>Pseudomonas aeruginosa</i>
Ward table	Table and tray	<i>Staphylococcus aureus</i>
		<i>Escherichia coli</i>

### Isolation of bacteria from surgical ward

The data revealed that various types of bacterial isolates i.e. *P. aeruginosa*, *K. pneumonia*, *S. epidermidis*, *B. subtilis*, *B. cereus* and *S. aureus* present in different places of the wards. Maximum numbers

of bacterial isolates were *P. aeruginosa* isolates from floor, walls, instruments, nurse hand before and after dressing and bed sheets places. Followed by *S. epidermidis*, the least number of isolate was *B. subtilis* and *B. cereus* presented in instruments and tray (Table 2).

**Table 2** Isolation of bacteria from surgical wards

Area		Bacterial Isolates
Ward	Wall, Floor	<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus aureus</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Bacillus cereus</i>
Washroom	Wall, Floor	<i>Bacillus subtilis</i>
		<i>Pseudomonas aeruginosa</i>
Sheet	Sheet	<i>Staphylococcus epidermidis</i> <i>Staphylococcus aureus</i>
Table	Table	<i>Klebsiella pneumonia</i>
Dust ben	Dust ben	<i>Staphylococcus aureus</i>
		<i>Staphylococcus epidermidis</i>
Surgical	Before use of tray and after use tray	<i>Staphylococcus epidermidis</i>
		<i>Bacillus cirrus</i>
Instruments	Stethoscope, Catheter	<i>Bacillus subtilis</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Staphylococcus epidermidis</i>
Hands	From nurse hand before and after dressing	<i>Pseudomonas aeruginosa</i>
		<i>Klebsiella pneumonia</i>
		<i>Staphylococcus aureus</i>
		<i>Pseudomonas aeruginosa</i>

**Isolation of bacteria from gynae ward**

Various types of bacterial isolates i.e. *P. aeruginosa*, *S. pyogens*, *S. aureus*, *B. subtilis*, *K. pneumonia*, *M. luteus*, *P. vulgaris*, *R. equi* and *E. coli* present in different places of the ward. Maximum number of

bacterial isolates was *P. aeruginosa*, isolates from vaginal speculum, floor, wall, hand and bed sheet places. Followed *K. pneumonia* isolated from vaginal speculum, ward wall and bed sheet places. While both *E. coli* and *R. equi*, were isolated from one place bed sheet (Table 3).

**Table 3** Isolation of bacteria from Gynae ward

Area		Bacterial Isolates
Ward	Wall, Floor	<i>Staphylococcus aureus</i>
		<i>Klebsiella pneumonia</i>
		<i>Streptococcus pyogens</i>
		<i>Proteus vulgaris</i>
		<i>Klebsiella pneumonia</i>
		<i>Pseudomonas aeruginosa</i>
		<i>Pseudomonas aeruginosa</i>
Wash room	Floor	<i>Proteus vulgaris</i>
		<i>Streptococcus pyogens</i>
		<i>Bacillus subtilis</i>

Table Continued...

Area		Bacterial Isolates
Ward bed	Bed sheet	<i>Pseudomonas aeruginosa</i>
		<i>Klebsiella pneumonia</i>
		<i>Escherichia coli</i>
		<i>Staphylococcus aureus</i>
		<i>Micrococcus luteus</i>
		<i>Rhodococcus equi</i>
Hand	From nurse hand before and after dressing	<i>Pseudomonas aeruginosa</i>
		<i>Proteus vulgaris</i>
		<i>Micrococcus letus</i>
Vaginal speculum	Vaginal speculum	<i>Klebsiella pneumonia</i>
		<i>Bacillus subtilis</i>
		<i>Pseudomonas aeruginosa</i>

### Isolation of bacteria from operation theater

The various types of bacterial isolates i.e. *P. aeruginosa*, *S. aureus* and *E. coli* present in different places of the Operation Theater. *P. aeruginosa* and *S. aureus* isolates was more frequently isolated from floor, wall and sheet places, while *E. coli* was isolated only from walls (Table 4).

**Table 4** Isolation of bacteria from Operation Theater

Area	Bacterial Isolates
Floor	<i>Pseudomonas aeruginosa</i>
	<i>Staphylococcus aureus</i>
Wall	<i>Escherichia coli</i>
	<i>Staphylococcus aureus</i>
Sheet	<i>Pseudomonas aeruginosa</i>

### Discussion

Nosocomial infections are those infection that are not present in a patient when enters hospital. Hospital acquired infections is worldwide problem that effect the developed as well as under developing countries. It is caused by those pathogens that are easily transmitted and spread on the body e.g. *S. aureus*, *S. epidermidis*, *B. subtilis*, *P. aeruginosa*, and *Klebsiella spp.* Nosocomial infection is the major cause of mortality and mobility in the hospitalized patients. It usually causes urinary tract infections, surgical site infections and blood stream infections. The present study was conducted to find out the scenario of the nosocomial pathogens resistance.

The result of the current study shows that the most dominant causative agent of nosocomial infections is *P. aeruginosa*, found to be one of the major causes of nosocomial infections worldwide. It is found as normal micro flora of the skin and is found in frequent in the environment.<sup>8</sup> The major cause for the hospital acquired infection in the immune-compromised and burn patients are *P. aeruginosa*.<sup>9</sup> This bacterium has the property to show resistant to several antibiotics, because implementation of antibiotics inactivates enzymes and slows down the permeability rate.<sup>10</sup>

The current study revealed different bacterial species that are the causative agents of Nosocomial infection. Those species include *P.*

*aeruginosa*, *E. coli*, *S. epidermidis*, *S. aureus*, *B. subtilis*, *B. cereus*, *K. pneumonia* and *S. pyogens*. Borkow also isolated the same nosocomial infection agents from different hospitals.<sup>11</sup> A study was conducted in the teaching hospital of Tehran Iran, to show the rate of overall Hospital acquired infections.<sup>12</sup>

Mohammad isolated the nosocomial infection agents such as *S. aureus* and *S. epidermidis*.<sup>13</sup> The same agents were isolated in our present study. The current study reported the *P. aeruginosa* as the most common specie while Naidu reported *K. pneumonia* as the most common specie.<sup>7</sup> Sabra reported *E. coli* the most frequent as compared to *P. aeruginosa*.<sup>4</sup> The study conducted reported that the most frequent type of nosocomial Urinary tract infection (UTI) is *E. coli* in the patient of spinal card injury.<sup>14</sup> The previous results are similar to our results they revealed *P. aeruginosa* the most frequent Nosocomial infection agent. The result of the research conducted by<sup>15</sup> is similar to our results, they reported *P. aeruginosa*.

In the year 2010 study was conducted in the tertiary hospitals in Dhaka about the nosocomial infections, this study revealed that the frequent cause of the infections was *E. coli* followed by *Pseudomonas*, *Proteus*, *Staphylococcus* and *Acinetobacter*.<sup>16</sup> While the current study reported *P. aeruginosa* as the most frequent agent followed by the *S. aureus* and *B. subtilis*, *S. epidermidis* and *S. aureus* are the most frequent in the surgical ward. One of the important nosocomial pathogen is *P. aeruginosa* that can survive in animal medium.<sup>17</sup>

### Conclusions

In the current study various numbers of bacterial species were identified. In which *P. aeruginosa* is dominant species and found in all the collection sites of the sample collection. Other important nosocomial agents like *B. cereus*, *P. aeruginosa*, *S. epidermidis*, *S. aureus*, *B. subtilis*, *E. coli*, *S. pyogens*, *K. pneumonia*, *M. luteus*, *P. vulgaris* and *R. equi* were also identified in the current study. Molecular identification of the pathogens at specie level is needed. Further study for antibiotic sensitivity test is needed. Hygienic sanitation of the hospital and improve the hospital equipment is needed. Public awareness about nosocomial infection is needed.

### Author contribution

MA, MK, MNU, and SHS helped in manuscript writing. MNU proof read and designed the study. Final manuscript was approved by all the authors.

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## Conflicts of interest

The present study has no conflicts of interest.

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