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Study of Bacterial Contamination in the Operating Room and Newborn Intensive Care Unit: Effects of the Predominantly Used Antibiotics at Sabratha Teaching Hospital



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Abstract

Contamination by bacteria in the operating room (OR) and newborn intensive care units (NICU) has continued to increase the incidence of nosocomial infections and a center point of multidrug-resistant (MDR) pathogens. This study was to examine the spread of bacteria isolated from inanimate surfaces in the operating room and neonatal intensive care units at Sabratha Teaching Hospital and find out the effectiveness of the most antimicrobial used. Methods: At the Sabratha Teaching Hospital in Sabratha, Libya, a cross-sectional, descriptive study was conducted in the OR and NICU from February to May 2024. A total of 22 swab samples were collected from high-contact environmental surfaces. Isolation, identification, and antibiotic sensitivity patterns were performed via standard techniques. Results: After 48 hours of incubation, (54.5 %) of the swabbed surfaces showed positive bacterial growth and 45.5% showed no bacterial growth, of which 25% had gram-positive bacteria and 75% had gram-negative bacteria. Staphylococcus saprophyticus 3 (25%) was the identified gram-positive bacterial isolate, and Klebsiella spp. 5 (55.5%) and Pseudomonas aeruginosa 4 (44.4%) were the probable gram-negative bacterial isolates. The antimicrobial susceptibility pattern of clinical pathogens tested Staphylococcus epidermides and Klebsiella spp showed similar patterns which were sensitive to most antimicrobials used, while Pseudomonas aeruginosa was highly resistant to Cotrimoxazole, Cefatoxime, Cefixitin, Ceftazidime, Ceftricillin, and Calvu amoxicillin. Conclusion: This study confirms the need for rigorous disinfection protocols and hand hygiene to reduce the spread of these pathogens in the OR and NICU.

Keywords: Antibiotic Resistance, Intensive Care Unit, Nosocomial Infection, Operating Room, *Staphylococcus Epidermides*.

INTRODUCTION

Patients treated at a hospital or other healthcare institution but not present at the time of admission may contract nosocomial infections, sometimes known as "hospital-associated infections" (HCAIs). These conditions may arise while treating other conditions or after the patient is released from the hospital. It also covers illnesses that employees contract at work (Haque *et al.* 2018). In rich nations, seven out of every 100 hospitalized patients may have one of these infections linked to healthcare, while ten out of every 100 patients in impoverished nations may have one (Haque *et al.* 2020).



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Bacteria may cause injury to patients in operating rooms (ORs) and heavily taxed hospitals (Weinstein 1998). Depending on the quantity of bacterial pathogens present, various sources have different effects on the degree of bacterial contamination. Similarly, the degree of contamination can be decreased if hospital staff members pay attention to hygiene standards and a hospital's environment, particularly, its cleanliness and disinfection are the primary factors in lowering the danger of microbial infection (Mora and Alanta 2001). The breathing pattern in active operating rooms and intensive care also helps reduce infection levels (Okon *et al* 2012). Hospitals are reservoirs of different types of microbes; many pathogenic bacterial species have colonized frequently touched areas (Shiferaw *et al*. 2013). Infectious mechanisms, high antibiotic use, and patient-to-patient bacterial transmission due to inadequate infection control measures may explain why operating rooms and intensive care units are "hot zones" for the presence and spread of microbial resistance (Bhalla *et al*. 2007). The impact of bacterial contamination on operating rooms and hospital care and its overall impact on hospital infection control is important for the patient and the healthcare team (Tacconelli *et al*. 2018, Rochford *et al*. 2018).

A significant public health issue that contributes to higher rates of morbidity and mortality as well as longer hospital stays is the morbidity associated with infections in newborn care. According to published research, the infection frequency linked to healthcare in newborn intensive care units (NICUs) varies from 8.7% to 74.3% (Kajiyazdi *et al.* 2021, Chiguer *et al.* 2019, Cura *et al.* 2016).

The neonatal period, which lasts for four weeks and accounts for approximately 3.6 million deaths annually, is primarily caused by infections, complications from premature birth, and intrapartum-related neonatal deaths (birth asphyxia) (Lawn *et al.* 2010). Because of the severity of their illness and their exposure to medical devices such as central venous catheters (CVCs) and mechanical ventilators, as well as resistant microorganisms, newborns admitted to intensive care units (ICUs) are quite dangerous for nosocomial infections (NI) (Chiguer *et al.* 2019). The types of organisms that cause infections vary geographically and over time, and the emergence of antibiotic-resistant organisms has become a major global health concern (Ingale *et al.* 2017). There is a link between the presence of pathogens on hospital surfaces and equipment and the frequency with which they are cleaned, how they are cleaned, appropriate use of disinfectants, and appropriate decontamination. Gram-negative and Gram-positive bacteria have been isolated from inanimate surfaces and can survive persistently for months on dry surfaces at humid and lower temperatures (Sales *et al.* 2014).

The rate of contamination in operating rooms and critical care units is influenced by numerous factors. These include the inoculum size, moisture content, source and target surface, and organism species. The setting of the critical care unit, the number of nurses, the number of patients colonized or infected, and hand hygiene compliance are other significant factors (D'Alessandro *et al.* 2021).

These infections are more serious and cause more deaths in intensive care if caused by resistant bacteria, especially infections from venous catheters or ventilator infections. Nosocomial infections are caused by various reasons, including hospital staff and ventilation and breathing systems. Treatment for bacterial infections is typically individualized and based on the patient's clinical picture. Consequently, it's essential to identify every strain of the bacteria that could be causing nosocomial infections. More antibiotics are advocated when information on bacterial occurrence in the critical care unit and operating room is lacking and their antimicrobial resistance is unclear. This can result in the improper use of antibiotics. It not only provides no

treatment benefits but also creates problems for patients. The misuse of antibiotics is known to cause *Clostridium difficile* bacteria, which causes diarrhea. Understanding the mechanisms of bacterial incidence and drug resistance in the intensive care unit and operating room is considered the first step in treating infectious diseases (Bobo and Dubberke 2010, Esfahani *et al.* 2017). Although the number of beds in intensive care units is low, the number of hospital-acquired infections is high. Nowadays, controlling the risk of nosocomial infection is becoming a priority (Merzougui, *et al* 2018). Since infectious diseases are transmitted from patients in intensive care and account for almost half of all infections, controlling and reducing the spread of infectious diseases in these regions provides significant economic benefits.

In relation to the above introduction, this study aimed to investigate abiotic surface infections in the newborn intensive care unit (NICU) and operating room (OR) to evaluate the effectiveness of the most commonly used antibiotics.

MATERIALS AND METHODS

Study design and setting

This cross-sectional study was conducted from February to May 2024 in the operating room and newborn intensive care unit (NICU) of the Sabratha Teaching Hospital in Sabratha, Libya.

For this study, 22 swab samples were taken from high-contact surfaces, including [gowns, instrument boxes, tables of instruments before operations, bedside, reception desks, liquid soup, betadine, incubator sides, neonatal beds, suction tips, ventilators, ambu bags, digital weighing machines, waterspouts, and staff tables].

Sample collection

Sterilized cotton swabs were labeled, collected, and placed in a tube containing nutritional broth an hour after the samples were obtained. The swabs were inoculated onto nutrient agar in the lab and kept overnight at 37°C. MacConkey agar was used for subculture, and blood agar plates were purified by aerobic 24-hour incubation at 37 °C.

Identification of Isolated Bacteria

Standard microbiological methods, such as colony morphology, and Gram stain are used to differentiate between Gram-positive and Gram-negative bacterial isolates. Comprehensive biochemical analyses using oxidase, coagulase, and catalase were performed to determine all pure isolates (Cheesbrough, 2009).

Antibiotic susceptibility of the isolated bacteria

The Clinical and Laboratory Standards Institute (CLSI) guideline (Yusuf *et al.* 2017) states that modified Kirby-Bauer disk diffusion is employed for antibiotic susceptibility testing following isolation and identification. The standard medium for the Kirby-Bauer method of susceptibility testing is Muller Hinton Agar (MHA). A filter paper tablet containing a known concentration of the antibiotic in a precise amount is used to calculate the minimum inhibition concentration (MIC). (Weinstein *et al.* 2018). One pure colony of bacteria was combined with one drop of sterile normal saline to create the bacterial suspension. The bacterial suspension was then compared to the McFarland standard to identify the concentration of the bacteria.

The test organism suspension was swabbed across the whole area of the MHA plate. After rotating the platter 360 degrees and doing so three times, let the dishes air dry fully. Using tongs, after adding the antibiotic tablets to the contaminated plates, the dishes were incubated for one whole day at 37°C. To evaluate the isolates' susceptibility to different antibiotics, inhibition regions were also measured around each antibiotic tablet using the provided ruler. Some areas around the disc showed no microbial growth, indicating that the antibacterial agent is effective; Microbes were growing

around the disc in other regions, suggesting that there was no zone encircling it and that the antibacterial chemical was inert.

In this study, antibiotic disks used were Cotrimoxazole 0.75 mg (COT 0.75), Amikacin 1 mg (AMK 1), Cefatoxime 1mg (CTX 1), Cefoxitin 1 mg (FOX 10), ciprofloxacin 5 mg (CIP 5), Ceftazidime 1 mg (CAZ 1), imipenem 10 mg (IMI 10), Polymixin 30 mg (POL 30), Linezolid 3 mg (LNZ 3), Meropenem 1mg (MER1), Ceftricillin 3 mg (CTR 3), Clavu amoxicillin 1 (AUG 1), Gentamycin 1mg (GEN 1), manufactured by the Bioanalyse company.

RESULTS

After 48 hours of incubation, 12 samples (54.5%) out of the 22 swabs that were analyzed for the presence or absence of bacteria exhibited evidence of growth, while the other 10 samples (45.4%) showed no growth.

Additionally, the findings demonstrated that 9 samples (75%) contained Gram-negative bacteria and 3 samples (25%) included Gram-positive bacteria. The potential Gram-positive organism isolated was *Staphylococcus epidermides* 3 (100%). The majority of *Staphylococcus epidermides* in the operating room have been isolated from the staff table, operating bed, and reception table. The potential Gram-negative organisms isolated were *Pseudomonas aureuginosa* 4 (44.47) and 5 (55.5%) *Klebsiella* spp isolates from the suction tip, liquid soap, baby incubator heater 3, neonate bed, and ambu bag. *Pseudomonas aeruginosa* isolates, however, were recovered from the suction tip, operation bed, outside surface of the incubator, and the water of the baby incubator 3.

Table (1). The Types and frequency of bacteria isolated from inanimate surfaces in the operating room and neonatal intensive care unit at Sabratha Teaching Hospital

Type of isolates	Gram-positive (n=3)	Gram-negative (n=9)	
	3(25%)	9(75%)	
	Gram-positive isolates Frequency	Percentage	
Staphylococcus epidermides	3	25%	
	Gram-negative isolates		
Klebsiella spp	5	55.5%	
Pseudomonas aeruginosa	4	44.47%	

The isolated bacteria's antibiotic susceptibility tests, as indicated in Tables 2, 3, and 4, revealed an even pattern with a high level of resistance to Cotrimoxazole, Cefatoxime, Cefixitin, Ceftazidime, Ceftricillin, Ceftricillin, and Clavu amoxicillin.

Table (2). The antibiotic sensitivity profile of *Staphylococcus epidermides*

organism	Antimicrobial	Total tested	Sensitive	Intermediate	Resistance
			n(%)	n(%)	n(%)
Staphylococcus epidermides	Cotrimoxazole	3	3(100%)	0 (0%)	0(0%)
	Cefatoxime	3	3(100%)	0(0%)	0(0%)
	Ciprifloxacin	3	3(100%	0(0%)	0(0%)
	Polymixin	3	3(100%)	0(0%)	0(0%)
	Linezolid	3	3(100%)	0(0%)	0(0%)
	Vancomycin	3	3(100%)	0(0%)	0(0%)
	Ceftricillin	3	0(0%)	3(100%)	0(0%)

Although the isolates were too few to provide meaningful antimicrobial sensitivity analysis, the organism (*Staphylococcus epidermides*) had intermediate resistance to ceftricillin but was sensitive to all other antimicrobials.

Table 3: The antibiotic sensitivity profile of Klebsiella species

Organism	Antimicrobial	Total tested	Sensitive n(%)	Intermediate n(%)	Resistance n(%)
	Cotrimoxazole	5	5(100%)	0 (0%)	0(0%)
·······································	Cefatoxime	5	5(100%)	0(0%)	0(0%)
	Cefoxitin	5	5(100%)	0(0%)	0(0%)
	Ceftazidime	5	5(100%)	0(0%)	0(0%)
	Polymixin	5	5(100%)	0(0%)	0(0%)
	Gentamycin	5	5(100%)	0(0%)	0(0%)
	Clavu amoxycillin	5	5(100%)	0(0%)	0(100%)

Although the isolates were too few to provide a meaningful antimicrobial sensitivity, the organism (*Klebsiella* spp) was sensitive to all the antimicrobials.

Table 4: The antibiotic sensitivity profile of Pseudomonas aeruginosa

Organism	Antimicrobial	Total tested	Sensitive n(%)	Intermediate n (%)	Resistance n(%)
	Cotrimoxazole	4	0(0%)	0 (0%)	4(100%)
Pseudomonas aeruginosa	Amikacin	4	4(100%)	0(0%)	0(0%)
	Cefatoxime	4	0(0%)	0(0%)	4(100%)
	Cefixitin	4	0(0%)	0(0%)	4(100%)
	Ciprofloxacin	4	4(100%)	0(0%)	0(0%)
	Ceftazidime	4	0(0%)	0(0%)	4(0%)
	Meropenem	4	4(100%)	0(0%)	0(100%)
	Ceftricillin	4	0(0%)	0(0%)	4(100%)
	Clavu amoxicillin	4	0(0%)	0(0%)	4(100%)

Pseudomonas aeruginosa showed a high degree of resistance to Cotrimoxazole 4 (100%), Cefatoxime 4 (100%), Cefixitin 4(100%), Ceftricillin 4(100%), and Clavu amoxicillin 4(100%). Among *P. aeruginosa*, 100% sensitivity was observed with Amikacin, Ciprofloxacin, Ceftazidime and Meropenem.

DISCUSSION

Recent studies have shown that microbial contamination of the OT and ICU environment can lead to colonization and infection of patients. Due to their weakened immune systems from invasive medical operations during their hospital stay, patients in intensive care units are most vulnerable to hospital-acquired infections or HAIs. Physicians and intensive care unit staff may act as carriers for the transfer of infectious diseases from other hospitals to the intensive care unit. Therefore, the hands of ICU staff and healthcare workers (HCWs) must follow the tightest hygiene guidelines. Furthermore, the ICU environment's pollution greatly contributes to nosocomial illness acquisition in both patients and healthcare workers. Examining the frequency of bacterial contamination on healthcare workers' hands and the surfaces in the intensive care unit could yield suggestions for limiting the spread of dangerous bacteria to patients and staff in medical environments (Tajeddin *et al.* 2016).

Hazardous infections could arise from contaminated healthcare workers' contact with hospitalized patients in intensive care units. The presence of live bacteria in the squamous layer of skin, which is constantly shed from healthy skin, is the reason behind the contamination of bed linens, patient robes, and other items in the intensive care unit (Tajeddin *et al.* 2016). One of the main causes of the increased rates of nosocomial infections in NICUs is bacterial contamination. During the care of diseases, non-critical medical equipment and inanimate surfaces may contain bacteria for an extended period and come into touch with patients and medical staff (Darge *et al.* 2019, Nseir *et al.* 2011). On dry surfaces, the majority of gram-positive bacteria, including methicillin-resistant Staphylococcus aureus (MRSA), can persist for months (From seven days to seven months). Numerous Gram-negative bacteria, including *Klebsiella* species, *E. coli*, and *Acinetobacter* species, on dry surfaces, can survive for up to 30 months (Suleyman *et al.* 2018).

According to our findings, a wide range of bacteria including, Gram-positive (25%) and Gram-negative (75%) bacteria, were present on non-living surfaces. In contrast to research done in Zimbabwe in tertiary hospitals in Bulawayo's intensive care units, Gram-negative bacteria (66.18%) constituted the majority of bacterial isolates (33.82%) as opposed to Gram-positive bacteria (Mbanga *et al.* 2018). This study is comparable to one carried out in the NICU of the Manipal Teaching Hospital in Pokhara, Nepal, where the majority of bacterial isolates were Gram-negative (66.3%) as opposed to Gram-positive (33.6%) (Bhatta DR. 2021).

The findings of this study differ from those of one conducted in the obstetrics and gynecology department of Al-Jala Hospital in Tripoli, Libya, where a higher proportion of bacterial isolates (68.9%) were Gram-positive than Gram-negative (31.03%) (Ben Ashur *et al.* 2022). This could be the result of sampling from various surfaces; it could also be the result of sporadic disinfection, variations in the disinfectant kinds used, unhygienic conditions, and crowding.

This study's overall bacterial contamination rate of (54.5%) is comparable to a study conducted at Windhoek Central Hospital in Windhoek, Namibia, where the neonatal intensive care unit (NICU) showed contamination rates of (52.8%) (Alphons *et al.* 2020). The study's (52.8%) contamination rate is consistent with findings from another study conducted at the intensive care section of a tertiary hospital in Bauchi, northeastern Nigeria, which recorded (52.8%) contamination rates for neonates (Alphons *et al.* 2020).

An investigation conducted in the tertiary hospital in Bulawayo, intensive care units, Zimbabwe, revealed a significantly greater contamination incidence of 86.1% (Mbanga *et al.* 2018). The contamination rate in this study is also much lower when compared to a study done in Morocco and Libya which reported a contamination rate of 96.3% and 95% respectively (Ben Ashur *et al.* 2022, Lalami *et al.* 2016). The high contamination rate recorded in NICU in this study and other similar studies around the globe is caused by several factors, including the large number of newborns who are often admitted for clinical care and assessment and have a variety of clinical problems. The constant presence and attention of nursing mothers and healthcare professionals is necessary for this therapeutic practice, which raises the intensity of human activity, mobility, and unit occupancy (Yusuf *et al.* 2017).

Variations in hand hygiene and sterilization procedures in neonatal intensive care units could account for the differences. In this study, the most common bacterial contaminants shown in (Table 1) were *Klebsiella* spp which accounted for (55.5%) of all isolates, followed by *Pseudomonas aeruginosa* which accounted for (44.4%) and *Staphylococcus epidermides* contributed (25%). These results align with the findings of a study in Benghazi, Libya, where *Klebsiella*. Spp and *Pseudomonas aeruginos*a were the most prevalent isolates. Other studies showed that *Klebsiella* spp and *Pseudo-*

monas aeruginosa bacteria were the most prevalent in such areas (Ensayef et al. 2009, Tajeddin et al. 2016).

The OR and NICU surfaces' microbial flora do not differ significantly from those of other hospital environments. Furthermore, preventing nosocomial infections in the NICU is made more difficult by the high susceptibility of premature and immunocompromised neonates. Colonization of NICU surfaces by opportunistic nosocomial pathogens like *Klebsiella* spp, *Staphylococcus epidermides*, and *Pseudomonas aeruginosa* are important for high-risk hospitalized patients like nosocomial infection. Additionally, there are significant dangers for babies, including low birth weight and congenital and neonatal defects.

In antimicrobial susceptibility profiles, *Staphylococcus epidermides* in this study (as seen in Table 2) showed high sensitivity to Cotrimoxazole (100%), Cefatoxime (100%), ciprofloxacin (100%), Polymixin (100%), Linezolid (100%), and Vancomycin (100%), but it was intermediate sensitive to Ceftricillin (100%). In another study, there is a difference in *Staphylococcus epidermides* sensitivity, which was moderately sensitive against gentamycin (71.4%), tetracycline (71.4%), clindamycin (50.0%) and Ciprofloxacin (42.9%). A low sensitivity against erythromycin (14.3%), with an intermediate resistance of (14.2%) was noted. These microorganisms were highly resistant to cefoxitin (100%) and penicillin (100%) (Alphons *et al.* 2020). These results were similar to those found in a study done in Nigeria where 90%, 100%, and 100% sensitivity was reported against ciprofloxacin, gentamycin, and erythromycin respectively (Yusuf *et al.* 2017).

In this study (as shown in Table 3) *Klebsiella* species showed a high sensitivity to Cotrimoxazole (100%), Cefatoxime (100%), Cefoxitin (100%), Ceftazidime (100%), Polymixin (100%), Gentamycin (100%), and Clavu amoxicillin (100%).

The result obtained in this study (as reported in Table 4) for the antimicrobial susceptibility patterns of *Pseudomonas aeroginosa* showed high sensitivity to Amikacin (100%), ciprofloxacin (100%), and meropenem (100%). But it was resistant to Cotrimoxazole (100%), Cefatoxime (100%), Cefixitin (100%), Ceftazidime (100%), and Clavu amoxicillin (100%). These results were similar to a previous study done in Windhoek which reported a high sensitivity to cefepime (100%), ciprofloxacin (100%), piperacillin-tazobactam (100%), and meropenem (100%). It was also resistant to gentamycin, ceftazidime, and imipenem (Alphons *et al.* 2020). Similar findings were also reported in a study done in Iran, which showed high resistance to Gentamycin (79%), Ceftazidime (25%), and Amikacin (83%) (Ekrami *et al.* 2011). These results were not consistent with the results of a study done in Bulawayo, Zimbabwe, in which high resistance to Gentamycin (62.5%), Piperacillintazobactam (50%), and Amikacin (12.5%) was reported (Mbanga *et al.* 2018).

CONCLUSION

One of the most important complications of hospitalized patients in OR and neonates is nosocomial infection in the NICU where contamination rates are very high. In the NICU and OR, (54.5%) of non-living surfaces and equipment were contaminated with germs. The isolation of pathogenic bacteria in this study such as *Staphylococcus epidermides* and *Klebsiella* spp indicate that they can be vehicles for disease transmission. Because of this, the operating room and neonatal intensive care unit where patient interaction with healthcare workers (HCWs), parents, and relatives is frequent and common, need rigorous disinfection and careful contact control procedures to reduce the spread of these pathogens.

Various interventions are required for the best possible infection control in the complex ecosystem, that is, the hospital setting. The most significant issues in hospital settings include the lack of a

standard protocol for nosocomial infection surveillance, high levels of bacterial contamination on hospital environmental surfaces, and poor hand hygiene.

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