



Société Ivoirienne de Microbiologie

MICROBIOLOGY AND NATURE
Journal homepage: www.microbiologyandnature.com

Bacterial contamination and antimicrobial susceptibility from the hands of health care workers (HCWs) and inanimate surfaces in the neonatal intensive care unit (NICU) at the Windhoek Central Hospital (WCH)

Kakuru Shampapi Alphons¹, Tjijenda Vanessa Fortune¹, Erastus Haindongo¹, Aboua Yapo Guillaume¹

¹Department of Health Sciences, Faculty of Health and Applied Sciences, Private Bag 13388, Windhoek, Namibia

Received Sept 27th 2019 / Revised Nov 30th 2019/ Accepted Dec 12th 2019/ Published online January 31st 2020

Abstract

Bacterial contamination of intensive care unit surfaces is of clinical concern because it is one of the major risk factors of intensive care unit-acquired infections and centre point of multidrug resistant (MDR) pathogens. This study aimed to determine the distribution of bacteria isolated from the hands of health care workers (HCWs) and inanimate surfaces in the neonatal intensive care unit (NICU) at the Windhoek Central Hospital. A cross-sectional, descriptive study carried out in NICU from September 2018 to October 2018 at the Windhoek Central Hospital in Windhoek. This study entailed the collection of 34 swab samples from high-contact environmental surfaces, as well as 2 swab samples from the hands of HCWs in NICU. Overall out of the surfaces swabbed, 52.8% yielded positive bacterial growth and 48.2% of the surfaces had no bacterial growth after 48 hours of incubation. Five different pathogens were identified, *S. aureus*, CoNS, Enterobacter species, *Pseudomonas aeruginosa* and Acinetobacter species respectively. CoNS accounted for majority of the bacterial pathogens isolated 70%, followed by Enterobacter species. The antimicrobial susceptibility pattern of clinically relevant pathogens tested showed similar patterns, with high resistant level to penicillin and cephalosporins. In the light of this, there is need therefore for thorough disinfection and conscientious contact control procedures to minimize the spread of these pathogens in the neonatal intensive care unit, where interaction between patients, HCWs, parents and caregivers is very common and frequent.

Keywords: Nosocomial infection, Inanimate surfaces, antibiotic resistance; intensive care units; multi-drug resistance

Résumé

La contamination bactérienne des surfaces des unités de soins intensifs est une préoccupation clinique car c'est l'un des principaux facteurs de risque d'infections acquises en unité de soins intensifs et le point central des agents pathogènes multirésistants au traitement médicaux. Cette étude visait à déterminer la distribution des bactéries isolées des mains des travailleurs de la santé et des surfaces inanimées au sein l'unité de soins intensifs néonataux (USIN) de l'hôpital central de Windhoek. C'est une étude descriptive transversale réalisée à l'USIN de septembre 2018 à octobre 2018 à l'hôpital central de Windhoek. Cette étude a impliqué la collecte de 34 échantillons prélevés sur des surfaces exposées à l'environnement, ainsi que de 2 échantillons prélevés sur les mains des travailleurs de la santé de l'USIN. Sur l'ensemble des surfaces nettoyées, 52,8% ont donné une croissance bactérienne positive et 48,2% des surfaces n'avaient aucune croissance bactérienne après 48 heures d'incubation. Cinq agents pathogènes différents ont été identifiés, *S. aureus*, CoNS, les espèces Enterobacter, *Pseudomonas aeruginosa* et Acinetobacter, respectivement. Le CoNS représentait la majorité des agents pathogènes bactériens isolés à 70%, suivis de l'espèce Enterobacter. Le profil de sensibilité aux antimicrobiens des agents pathogènes cliniquement pertinents testés a montré des profils similaires, avec un niveau de résistance élevé à la pénicilline et aux céphalosporines. Il est donc nécessaire de procéder à une désinfection minutieuse et à des procédures de contrôle des contacts consciencieuses afin de minimiser la propagation de ces agents pathogènes dans les unités de soins intensifs néonataux, où les interactions entre patients, agents de santé, parents et responsables de soins sont très fréquentes et fréquentes.

Mots-clés: infection nosocomiale, surfaces inanimées, résistance aux antibiotiques; unités de soins intensifs; multi-résistance aux médicaments

Corresponding author E-mail address: yaboua@nust.na

Introduction

Each year 3.6 million infants are estimated to die in the first four (04) weeks of life (neonatal period) but the majority of the neonates continue to die at home and are uncounted. There are three major causes of neonatal death, mainly infections, complications of preterm birth, and intra-partum-related neonatal deaths (birth asphyxia) and they account for approximately more than 80% of all neonatal deaths globally (Lawn et al. 2010).

Healthcare-associated infection (HAI) is a serious problem in neonates who are admitted to the neonatal intensive care unit (NICU). The HAI is associated with increases in mortality, morbidity, and prolonged length of hospital stay (Krawczenko et al. 2012). A Healthcare-associated infection is defined as an infection not present or incubating at the time of admission, with onset after 48 hours of stay. Nosocomial infections are usually a result of pathogens that are resistant to the first-line of antibiotics (Urzedo et al. 2014).

New-borns admitted to intensive care units (ICUs) are at high risk for developing nosocomial infections (NIs) because of the severity of their illness and exposure to invasive medical devices such as mechanical ventilators and central venous catheters (CVCs) and resistant microorganisms (Urzedo et al. 2014). According to Dramowski et al. (2017) hospitalized neonates are considered a vulnerable population due to their immature immune systems and frequent infectious disease exposures through contact with healthcare staff, parents, other patients, equipment, and the hospital environment (Dramowski et al. 2017).

The epidemiology and incidence of infections varies widely among NICUs (7-25.5%), depending on environmental factors and differences in clinical practice. According to Brito et al. (2010) bloodstream infections (BSI) are the most frequent neonatal infection in the NICU (45-55%), followed by respiratory infections (16-30%) and urinary tract infections (8-18%) (Brito et al 2010). It was stated that low gestational age and birth weight are the two most frequently identi-

fied individual risks for NIs. The pattern of organisms causing infections differs from place to place and over a period of time. Additionally, the emergence of resistant organisms to antimicrobial agents has become a major health threat worldwide (Ingale et al. 2017).

According to a study conducted by Gray and Omar (2013) the incidence and causes of infections varies widely among NICUs. However, the incidence of infection is higher in developing countries, where Gram-negative bacteria are usually reported to be the predominant pathogens (Gray and Omar, 2013).

According to studies done by Shah et al. (2013) and Mehar et al. (2013) in India as cited by Ingale et al (2017), the rate of infections in NICU is 21.2% and 50.9% respectively. In the United States the frequency of infection in NICU ranges from 3-6% and from 8-10% in Europe, these differences could be due to different population characteristics and varying predisposing factors (Ingale et al. 2017).

Intensive care unit acquired infections are a challenging health problem globally and they are also a major cause of mortality and morbidity worldwide. Contamination of inanimate surfaces (telephones, keyboard, medical charts, and stethoscopes) and cross-contamination in ICU has been identified as the major cause of outbreaks. Contamination within the ICU may occur either by the transfer of microorganisms contaminating the health-care worker's hands or direct patient shedding of microorganisms in the vicinity of a patient's bed (Russoto et al. 2015). It has been demonstrated that the increased incidences of HAIs are related to cross-infections from patient to patient or hospital staff to patient, poor infection control practices and to the presence of pathogenic microorganisms that are selected and maintained within the hospital environment (Abreu et al. 2013).

According to a study done by Sales et al. (2014) there is a relationship between the presence of resistant pathogens on hospital surfaces and equipment and the frequency with which they are cleaned, how they are cleaned, the proper use of disinfectants and the proper disinfecting

Both Gram-negative and gram-positive bacteria have been isolated from inanimate surfaces and are able to survive up to months on dry surfaces, with longer persistence seen under humid and lower temperature conditions. Many factors influence and affect rate of contamination and cross-contamination in the ICU. This include type of organisms, source and destination surfaces, humidity level, and size of inoculum. Other factors that play a role include hand hygiene compliance, number of nurse-staffing levels, number of colonized or infected patients and ICU structural features (Russoto et al. 2015). This study aimed to determine the distribution of bacteria isolated from the hands of health care workers (HCWs) and inanimate surfaces in the neonatal intensive care unit (NICU) at the Windhoek Central Hospital.

Materials and Methods

Study design

This study was a cross-sectional, descriptive study carried out in a neonatal intensive care unit (NICU) in October 2018 at a referral government hospital (Windhoek Central Hospital) in Windhoek, Namibia. This study entailed the collection of 34 swab samples from high-contact environmental surfaces, as well as 2 swab samples from the hands of HCWs in NICU. Positive and negative controls were run during the processing of samples to ensure validity. The study only focused on selected bacteria that were isolated and identified. Other organisms such as parasites, viruses and fungi were excluded from this study. The permission to carry out the study was granted by the Department of Medical Laboratory Sciences, Namibia University of Science, and Technology (NUST); the Ministry of Health and Social Services (MoHSS) clearance number SS 2018 was granted by the Ministry of Health and Social Services (MoHSS). Staff members and patient names were not used in this study.

Sampling and data collection technique

In this study convenience sampling method was used, whereby only identified inanimate objects/surfaces within the neonatal intensive care unit at the Windhoek Central Hospital were considered.

Inclusion criteria and exclusion criteria

Inclusion criteria: The study only focused on selected inanimate surfaces that were identified and sampled

within the neonatal intensive care unit. The study focused on selected bacteria that were isolated and identified.

Exclusion criteria: unselected surfaces were not sampled and other organisms such as parasites, viruses and fungi were excluded from this study.

Laboratory analysis

Each swab was well labeled and transported in a tube provided by the manufacturer containing SRK solution. Swabs were transported to the laboratory within 1 hour of sample collection in a cooler box (1-4°C). Upon arrival to the laboratory, the swabs directly inoculated onto 5% horse blood agar and MacConkey agar plate and incubated at 37°C for 24 hours. Discrete colonies after 24 hours of incubation were further sub-cultured onto fresh prepared plates of blood and MacConkey agar plates to obtain pure cultures. The purified cultures were Gram stained and biochemical tests were done for identification. Analytical profile Index (API) 20E was used for the identification of Gram negative bacterial isolates. For this purpose, a Pasteur pipette was used to inoculate the API strips with the suspensions of the organisms, the tubes on the strip were inoculated as per manufacturer's instructions. Whereby some tests both the tube and the cupule were filled with the suspension, for some only the tube was filled with the suspension and to others paraffin was added to the cupule to create an anaerobic environment. The Strip were incubated aerobically for 24 hours at 37°C, after the incubation period interpretation of the reactions was done by adding reagents to the respective tubes, hence the organisms were identified based on their reaction with the various substrates.

Antimicrobial susceptibility of the isolated bacteria

Antimicrobial susceptibility of the bacterial strains was tested using the Kirby-Bauer disk diffusion method on the Mueller-Hilton agar (MH) according to the Clinical Laboratory Standard Institute guidelines. Suspensions of the tested organism were prepared by picking well isolated pure colonies of the organism with a sterile cotton swab. The colonies were emulsified in a sterile normal saline solution to give a density equivalent to that of 0.5 McFarland standards. A sterile cotton swab was then dipped in the suspension and excess moisture in the swab head squeezed out on the inside of the tube. The entire surface of the MHA plate was swabbed with the test organism suspension, turning the plate 360 degrees and repeating the process three times. The antimicrobial disks were placed on the surface of the agar and gently pressed down with sterile forceps.

The plates were then incubated at 37°C for 24 hours aerobically within 30 minutes of applying the disks. After the incubation period, the zones of inhibition were measured with the aid of a Vernier caliper and the tested isolates were classified as either “resistant”, “intermediate” or “sensitive” to the respective drugs. Gram-positive isolates were tested against erythromycin (15 µg), clindamycin (2 µg), gentamycin (10 µg), ciprofloxacin (5 µg), ceftazidime (30 µg), penicillin (10 µg), tetracycline (30 µg) and novobiocin (30 µg), as for Gram-negative isolates they were tested against augmentin (10 µg), amikacin (30 µg), ceftriaxone (30 µg), ceftazidime (30 µg), cefuroxime (30 µg), cefepime (30 µg), ampicillin (10 µg), piperacillin-tazobactam (10 µg), meropenem (10 µg), imipenem (10 µg) and ertapenem (10 µg) respectively.

RESULTS

A total of 36 inanimate surfaces were pre identified based on their location and degree of usage and these surfaces were swabbed using the swabbing method to determine the diversity of bacterial species colonizing the ICU environments and HCWs' hands. Overall out of the 36 surfaces swabbed, 19 (52.8%) yielded positive bacterial growth and 17 (48.2%) of the surfaces had no bacterial growth after 48 hours of incubation. Five different pathogens were identified,

S. aureus, CoNS, Enterobacter species, *Pseudomonas aeruginosa* and Acinetobacter species respectively. CoNS (*S. epidermidis* and *S. saprophyticus*) as presented in (Table 1) accounted for majority of the bacterial pathogens identified 14 (70%), while other pathogens are as follow *S. aureus* 1 (5%), Enterobacter species 3 (15%), *Pseudomonas aeruginosa* 1 (5%) and Acinetobacter species 1 (5%) respectively. From this study as presented in (Table 2) majority of the organisms were isolated from door handles (35%), feeding bottle holder (10%), wash basins (10%), bed linen (5%), bed trolley (5%), stethoscope (5), weighing scale (5%), saturation probe (5%), saturation machine monitor (5%), CPAP line (5%) and humidifier (5%) respectively. The antimicrobial susceptibility pattern of clinically relevant pathogens tested as presented in (Table 3 to 7) showed similar pattern with high resistant level to penicillin, ceftazidime and cefuroxime, and high sensitivity to Augmentin, amikacin, ciprofloxacin, gentamycin, piperacillin-tazobactam and all the carbapenems (meropenem, imipenem and ertapenem).

Table 1: The Types and frequency of bacteria isolated from inanimate surfaces in the neonatal Intensive Care Unit at Windhoek Central Hospital (N = 20).

Bacteria isolates	Frequency (n)	Percentage (%)
<i>Staphylococcus aureus</i>	1	5
<i>Coagulase negative staphylococci (CoNS)</i>	14	70
<i>Enterobacter spp</i>	3	15
<i>Pseudomonas aeruginosa</i>	1	5
<i>Acinetobacter spp</i>	1	5

Out of the organisms that were isolated, 15 (75%) were Gram positive cocci (in clusters) and 5 (25%) of the organisms were Gram negative bacilli. Among the organisms isolated, 19 (95%) of them were isolated from inanimate surfaces and 1 (5%) was isolated from the hands of a health care worker (HCW). Of the

organisms that were isolated and identified, 5% were *Staphylococcus aureus*, 50% were *Staphylococcus epidermidis*, 20% were *Staphylococcus saprophyticus*, 15% were *Enterobacter species*, 5% were *Pseudomonas aeruginosa* and 5% were identified as *Acinetobacter species*.

Table 2: Distribution of different pathogens in the neonatal intensive care unit at the Windhoek Central Hospital.

Surfaces swabbed	Organisms isolated
Store room door (outside) handle	<i>Staphylococcus saprophyticus</i>
Store room (files)	No organism isolated
Apron	No organism isolated
Keyboard and mouse	<i>Staphylococcus aureus</i> (MRSA)
Telephone	No organism isolated
Intensive care unit door handle	No organism isolated
Acute care room door handle	No organism isolated
Baby room door handle A	No organism isolated
Baby room door handle B	<i>Staphylococcus epidermidis</i>
Isolation room door handle (Entrance door)	<i>Staphylococcus epidermidis</i>
Isolation room 1 door handle	No organism isolated
Isolation room 2 door handle	<i>Staphylococcus epidermidis</i>
Isolation room 3 door handle	<i>Staphylococcus saprophyticus</i>
Isolation room 4 door handle	<i>Staphylococcus epidermidis</i>
Basin (isolation room 3)	<i>Staphylococcus epidermidis</i>
Basin (baby room)	<i>Staphylococcus saprophyticus</i>
Bed rails	<i>Staphylococcus saprophyticus</i>
Stethoscope	<i>Staphylococcus epidermidis</i>
Bed trolley	<i>Staphylococcus epidermidis</i>
Medical charts	No organism isolated
Weighing scale	<i>Staphylococcus epidermidis</i>
Saturation probe	<i>Staphylococcus epidermidis</i>
Ventilator pipes	No organism isolated
Incubator outside surfaces	No organism isolated
Health care worker's hands A	No organism isolated
Health care worker's hands B	<i>Enterobacter spp</i>
Saturation machine monitor	<i>Staphylococcus epidermidis</i>
Humidifier rim	No organism isolated
Fridge door handle (Feeding room)	No organism isolated
Basin (Feeding)	No organism isolated
Feeding room door handle	No organism isolated
Feeding bottle holder	<i>Enterobacter spp</i> and <i>Pseudomonas aeruginosa</i>
CPAP lines from CSSD	<i>Enterobacter spp</i>
Humidifier	<i>Acinetobacter spp</i>
Ambubag- from CSSD	No organism isolated
Electro-cardiogram lead wires	No organism isolated

Most bacterial isolates 6 (30%) were isolated from door handles and the least number of bacteria 1 (5%) were isolated from keyboard and mouse, feeding bottle holder and humidifier respectively. The most common bacteria that was isolated from the inanimate surfaces

were CoNS 14 (70%). The Bacteria were only isolated from 54.1% of the swabbed surfaces and no bacteria was isolated from 45.9% of the swabbed surfaces within the neonatal intensive care unit.

Table 3: The antibiotic sensitivity profile of *Staphylococcus aureus*

Organism	Antimicrobials	Total tested	Sensitive n (%)	Intermediate n (%)	Resistant n (%)
<i>Staphylococcus aureus</i>					
	Erythromycin	1	1 (100%)	0 (0%)	0 (0%)
	Clindamycin	1	1 (100%)	0 (0%)	0 (0%)
	Gentamycin	1	1 (100%)	0 (0%)	0 (0%)
	Ciprofloxacin	1	1 (100%)	0 (0%)	0 (0%)
	Cefoxitin	1	0 (0%)	0 (0%)	1 (100%)
	Penicillin	1	0 (0%)	0 (0%)	1 (100%)
	Tetracycline	1	1 (100%)	0 (0%)	0 (0%)

Although the isolates were too few to provide a meaningful antimicrobial sensitivity analysis this organism (*S. aureus*), was sensitive to all the antimicrobial except for cefoxitin and penicillin.

Table 4: The antibiotic sensitivity profile of coagulase negative *Staphylococci (CoNS)*

Organism	Antimicrobials	Total tested	Sensitive n (%)	Intermediate n (%)	Resistant n (%)
<i>CoNS</i>		14			
	Erythromycin	14	2 (14.3%)	2 (14.3%)	10 (71.4%)
	Clindamycin	14	7 (50%)	0 (0%)	7 (50%)
	Gentamycin	14	10 (71.4)	0 (0%)	4 (28.6%)
	Ciprofloxacin	14	6 (42.9%)	0 (0%)	8 (57.1%)
	Cefoxitin	14	0 (0%)	0 (0%)	14 (100%)
	Penicillin	14	0 (0%)	0 (0%)	14 (100%)
	Tetracycline	14	10 (71.4%)	0 (0%)	4 (28.6%)

All of the *CoNS* showed 100% resistance to cefoxitin and penicillin. Most of them were sensitive to Clindamycin (50.0%), Gentamycin (71.5%), Ciprofloxacin (42.9%) and Tetracycline (71.4%) respectively. Variable resistance was seen with erythromycin (71.4%), Clindamycin (50%), Gentamycin (28.6%), ciprofloxacin (57.1%) and tetracycline (28.6%) respectively.

Table 5: The antibiotic sensitivity profile for *Enterobacter species*

Organism	Antimicrobials	Total tested	Sensitive n (%)	Intermediate n (%)	Resistant n (%)
<i>Enterobacter spp</i>					
	Augmentin	3	1 (33.3%)	0 (0%)	2 (66.7%)
	Cefoxitin	3	0 (0%)	0 (0%)	3 (100%)
	Ceftazidime	3	0 (0%)	0 (0%)	3 (100%)
	Cefuroxime	3	0 (0%)	0 (0%)	3 (100%)
	Ceftriaxone	3	0 (0%)	1 (33.3%)	2 (66.7%)
	Cefepime	3	1 (33.3%)	0 (0%)	2 (66.7%)
	Ciprofloxacin	3	3 (100%)	0 (0%)	0 (0%)
	Ampicillin	3	1 (33.3%)	1 (33.3%)	1 (33.3%)
	Gentamycin	3	3 (100%)	0 (0%)	0 (0%)
	Amikacin	3	3 (100%)	0 (0%)	0 (0%)
	Piperacillin-Tazobactam	3	3 (100%)	0 (0%)	0 (0%)
	Meropenem	3	2 (66.7%)	0 (0%)	1 (33.3%)
	Imipenem	3	3 (100%)	0 (0%)	0 (0%)
	Ertapenem	3	2 (66.7%)	0 (0%)	1 (33.3%)

Enterobacter species showed high degree of resistance to cefoxitin 3 (100%), Ceftazidime 3 (100%) and Cefuroxime 3 (100%). Among the *Enterobacter species*, 100% sensitivity was observed with gentamycin, amikacin and piperacillin-tazobactam. They were also 3 (100%) sensitive to imipenem, 2 (66.7%) sensitive to meropenem and ertapenem respectively.

Table 6: The antibiotic sensitivity profile for *Pseudomonas aeruginosa*

Organism	Antimicrobials	Total tested	Sensitive n (%)	Intermediate n (%)	Resistant n (%)
<i>P. aeruginosa</i>					
	Amikacin	1	0 (0%)	1 (100%)	0 (0%)
	Gentamycin	1	0 (0%)	0 (0%)	1 (100%)
	Cefepime	1	1 (100%)	0 (0%)	0 (0%)
	Ceftazidime	1	0 (0%)	0 (0%)	1 (100%)
	Ciprofloxacin	1	1 (100%)	0 (0%)	0 (0%)
	Piperacillin-Tazobactam	1	1 (100%)	0 (0%)	0 (0%)
	Meropenem	1	1 (100%)	0 (0%)	0 (0%)
	Imipenem	1	0 (0%)	0 (0%)	1 (100%)

Although the isolates were too few to provide a meaningful antimicrobial sensitivity analysis this organism (*Pseudomonas aeruginosa*) had an intermediate resistance to amikacin, but it was resistant to gentamycin (100%), ceftazidime (100%), and imipenem (100%). It was sensitive to cefepime (100%), ciprofloxacin (100%), piperacillin-tazobactam (100%) and meropenem (100%), respectively.

Table 7: The antibiotic sensitivity profile for *Acinetobacter species*

Organism	Antimicrobials	Total tested N	Sensitive n (%)	Intermediate n (%)	Resistant n (%)
<i>Acinetobacter spp</i>					
	Augmentin	1	1 (100%)	0 (0%)	0 (0%)
	Cefoxitin	1	0 (0%)	0 (0%)	1 (100%)
	Ceftazidime	1	0 (0%)	0 (0%)	1 (100%)
	Cefuroxime	1	0 (0%)	0 (0%)	1 (100%)
	Ceftriaxone		0 (0%)	0 (0%)	1 (100%)
	Cefepime	1	0 (0%)	0 (0%)	1 (100%)
	Ciprofloxacin	1	1 (100%)	0 (0%)	0 (0%)
	Ampicillin	1	1 (100%)	0 (0%)	0 (0%)
	Gentamycin	1	0 (0%)	0 (0%)	1 (100%)
	Amikacin	1	1 (100%)	0 (0%)	0 (0%)
	Piperacillin- Tazobactam	1	1 (100%)	0 (0%)	0 (0%)
	Meropenem	1	1 (100%)	0 (0%)	0 (0%)
	Imipenem	1	1 (100%)	0 (0%)	0 (0%)
	Ertapenem	1	1 (100%)	0 (0%)	0 (0%)

Table 7 shows that, *Acinetobacter species* was resistant to cefoxitin, ceftazidime, ceftriaxone, cefepime and gentamycin. This organism was sensitive to augmentin, ciprofloxacin, ampicillin, amikacin, piperacillin-tazobactam, meropenem, imipenem and ertapenem respectively.

Discussion

Patients in ICUs are at the highest risk for HAIs because of invasive medical procedures during their hospitalization because of their low immunity. The ICU staff and physicians can serve as vehicles for the spread of resident pathogens from different hospital wards to ICUs. Accordingly, the hands of HCWs and ICU personnel require the greatest hygiene standards, contamination of the ICU environment also plays an important role in the acquisition of nosocomial pathogens by both patients and HCWs. Investigation of the rate of bacterial contamination of the hands of HCWs and the ICU environmental surfaces could provide recommendations for preventing transmission of pathogenic bacteria to patients and personnel in health-care settings (Tajeddin et al. 2015). Normal human skin is colonized by bacteria, with total aerobic bacterial counts rang-

ing from more than 4×10^4 CFU/cm to 1×10^6 CFU/cm. The total bacterial counts on the hands of HCWs ranges from 3.9×10^4 to 4.6×10^6 CFU/cm. HCWs are often contaminated with microbial agents in the hospital environment. Contact between the contaminated HCWs and hospitalized patients in ICUs might cause serious infections. Because nearly 10⁶ skin squamous contains viable microorganisms that are shed daily from normal skin, it is not surprising that patient gowns, bed linens, bedsides and other objects in the ICU environment become contaminated (Tajeddin et al. 2015). Bacterial contamination of ICU is the major factor responsible for increased incidence of nosocomial infections, with attendant consequential effects on patient and hospital management (Yusuf et al. 2017). Inanimate surfaces have often been described as the source for outbreaks of nosocomial in-

Most Gram-positive bacteria, such as *Staphylococcus aureus*, including methicillin resistant *Staphylococcus aureus* (MRSA), survive months on dry surfaces (7 days to 7 months). Many Gram-negative species, such as *Acinetobacter spp*, *Escherichia coli* and *Klebsiella spp* are able to survive for up to 30 months on dry inanimate surfaces (Holma, 2015).

In this study, the swabbing method was used instead of the direct plating method to determine the diversity of bacterial species colonizing the ICU environments and HCWs' hands. The results of our study showed contamination of the inanimate environments by diverse groups of bacteria, including both Gram-positive bacteria (75%) and Gram-negative bacteria (25%). This is comparable to a study done by Tajeddin et al. (2015) in Iran, where Gram-positive bacteria comprised a greater percentage of the bacterial isolates (60.7%) compared to Gram-negative bacteria (39.3%) (Tajeddin et al. 2015). In this study more Gram-positives were isolated as compared to Gram negatives and these results are consistent with the results of a study done in Nigeria, where majority of the isolates were Gram positive organisms (52.2%) as compared to the Gram negatives (47.8%). The isolation of more Gram-positive organisms is because they are known to be members of the body flora of both asymptomatic carriers and sick persons. These organisms can be spread by the hand, expelled from the respiratory tract or transmitted by animate or inanimate objects (Maryam et al. 2014).

Result from this study is different from a study done in Zimbabwe in Intensive Care Units of a Tertiary Hospital in Bulawayo, in which Gram-negative bacteria (66.18%) comprised the greater percentage of the bacterial isolates as compared to Gram-positive bacteria (33.82%) (Mbanga et al. 2018). The result from this study is also in line with that of a study done at Kiwoko Hospital, a rural setting in the central region of Uganda, in which Gram-positives comprised a greater percentage of the bacterial isolates (59.4%) when compared to Gram-

negatives (40.6%) (Segujja et al. 2011). This could be due to the fact that different surfaces were sampled, it can also be attributed to irregular disinfection, difference in the types of disinfectants used, hygienic conditions and overcrowding.

Overall the bacterial contamination rates from this study was 52.8%, this is similar to a study done at a tertiary hospital in Bauchi, North-eastern Nigeria in which contamination rates of 52.8% was seen in the neonatal intensive care unit (NICU) and 40.8% in the adult intensive care unit (AICU) respectively (Yusuf et al. 2017). The contamination rate of 52.8% in this study is in line with the results of a study done by Saka et al. (2016) at University of Ilorin Teaching Hospital, Ilorin, Nigeria, in which a rate of 67.7% was reported (Saka et al. 2016) and this was slightly higher than the rate in this study. The contamination rate in this study was also similar to a study done in Uganda where a contamination rate of 57.6% was reported (Segujja et al. 2011). A much higher contamination rate of 86.1% was seen in a study done in Zimbabwe in Intensive Care Units of a Tertiary Hospital in Bulawayo (Mbanga et al. 2018). The difference could be attributed to difference in the types of disinfectants used, hygienic conditions and overcrowding.

The contamination rate in this study is also much lower, when compared to a study done in Morocco in which a contamination rate of 96.3% was reported (Lalami et al. 2016). The high contamination rate recorded in NICU in this study and other similar studies around the globe is due to reasons such as high number of neonates with different clinical conditions being admitted frequently for clinical attention and evaluation. This clinical practice requires the frequent presence and attention of the mothers for breastfeeding and health care worker, thus increasing the unit occupancy density, traffic and human activities (Yusuf et al. 2017). Variation in hand hygiene and sterilisation procedures in different neonatal intensive care units, could account for the differ-

The predominant bacterial contaminants in study as presented in (Table 1) were Coagulase Negative Staphylococci (CoNS), which accounted for (70%) of all isolates, followed by Enterobacter species which accounted for (15%), *S. aureus*, *P. aeruginosa* and *Acinetobacter* species each contributed (5%). These results are in line with that of a study done in Nigeria in which the CoNS (19.2%) were one of the predominating organisms that were isolated together with *Staphylococcus aureus* (26.8%) and *Bacilli* species (33.0%), except that the study by Yusuf et al. (2017) did not report on Enterobacter species, *P. aeruginosa* and *Acinetobacter* species. The high contamination rate with CoNS and *S. aureus* is attributed to the fact that these pathogens are normal flora of human skin, and clothing fabrics that are continuously shed during routine activity and clothing fabrics (Yusuf 2017). In this study 70% of the bacterial isolated were CoNS, this is similar to the results of a study done in an Intensive Care Unit of the University of Maiduguri Teaching Hospital in Nigeria, were CoNS accounted for 39.4% (15) of all the isolates (Barma et al. 2017). However result from our study is different from the study done in a teaching hospital (Abuth Zaria) in northern Nigeria, in which *S. aureus* (21.7%) was the predominating isolate (Maryam et al. 2014). Result from our study is different from that of a study done in the paediatric wards of the University of Ilorin Teaching Hospital in Nigeria, were *Staphylococcus aureus* (39.4%) and *Acinetobacter baumannii* (23.8%) were the two most common organisms isolated respectively. From this same study eight different organisms were isolated mainly *S. aureus* (39.4%), *A. baumannii* (23.8%), *E. faecalis* (2.5%), *E. coli* (3.1%), *K. pneumoniae* (20%), *P. aeruginosa* (1.3%) and CoNS (10%) (Saka et al. 2016). This is different from the result of our study in which five different organisms were isolated mainly CoNS (70%), *S. aureus* (5%), *Enterobacter spp* (10%), *P. aeruginosa* (5%) and *Acinetobacter spp* (5%). This difference might be due to the fact that our sample size was small (36) compared to the

sample size of (201) used by Saka et al. (2016) and it can also be attributed to difference in cleaning procedures and no strict adherence to simple hand washing technique among health care workers. From this study as presented in (Table 2) majority of the organisms were isolated from door handles (35%), feeding bottle holder (10%), wash basins (10%), bed linen (5%), bed trolley (5%), stethoscope (5%), weighing scale (5%), saturation probe (5%), saturation machine monitor (5%), CPAP line (5%) and humidifier (5%) respectively. This is different from the results of a study done in Nigeria by Maryam et al. (2014) where bacteria were mostly isolated from surfaces such as uniforms (17.4%), intravenous fluid (IVF) stand (17.4%), tables (13.0%), stethoscopes (13.0%), pens (13.0%), door handles (13.0%) and chairs (13.0%) respectively. These differences can be attributed to the fact that different surfaces were sampled.

In antimicrobial susceptibility profiles, *S. aureus* in this study (as seen in Table 3) showed a high sensitivity to erythromycin (100%), clindamycin (100%), gentamycin (100%), ciprofloxacin (100%) but it was resistant to penicillin (100%) and ceftioxin (100%). These results are in line with those reported in study by Maryam et al. (2014), where (100%) sensitivity was seen against erythromycin. In the same study *S. aureus* had a 40% and 60% sensitivity pattern to gentamycin and ciprofloxacin respectively (Maryam et al. 2014). Result from our study is also in line with the results of a study carried out in Bauchi, Nigeria where high sensitivity of *S. aureus* to erythromycin (100%), gentamycin (100%) and ciprofloxacin (100%) (Yusuf et al. 2017). The results of our study do not agree with the results of a study done at Kiwoko Hospital, a rural setting in the central region of Uganda, where 90.7% sensitivity to ceftioxin was reported (Segujja et al. 2011). This study found that CoNS (as presented in Table 4) had a moderate sensitivity 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%).

These results are not in line with those found by a study done in Nigeria where 90%, 100% and 100% sensitivity was reported against ciprofloxacin, gentamycin and erythromycin respectively (Yusuf et al. 2017). The results from this study do not agree with that of a study done in Abuja, Nigeria, where 90% sensitivity against ciprofloxacin, 80% sensitivity against gentamycin and 87% sensitivity against erythromycin was reported (Barma et al. 2017). Results from this study is also different from the results of a study done in Intensive Care Units of a Tertiary Hospital in Bulawayo, Zimbabwe where a higher resistance rate to ciprofloxacin (88.2%), a lower resistance rate to erythromycin (64.7%) and gentamycin (23.5%) was reported (Mbunga et al. 2018).

In this study (as shown in Table 5) Enterobacter species showed a high sensitivity to ciprofloxacin (100%), gentamycin (100%), amikacin (100%), piperacillin-tazobactam (100%), meropenem (66.7%) and ertapenem (33.3%) respectively. High resistance was seen with cefoxitin (100%), ceftazidime (100%), and cefuroxime (100%). Additionally Enterobacter species showed moderate sensitivity to meropenem (66.7%), ertapenem (66.7%), augmentin (33.3%), cefepime (33.6%) and ampicillin (33.3%) respectively. These results do not agree with results from a study done in Iran, where high resistance levels were reported against amikacin (88%), gentamycin (90%), ciprofloxacin (53%) and a low resistance rate was seen against ceftazidime (20%) (Ekrami et al. 2011)

The result obtained in this study (as reported in Table 6) for the antimicrobial susceptibility patterns of *Pseudomonas aeruginosa* showed high sensitivity to cefepime (100%), ciprofloxacin (100%), piperacillin-tazobactam (100%) and meropenem (100%). It was also resistant to

gentamycin, ceftazidime and imipenem. These results were not consistent with the results of a study done in Bulawayo, Zimbabwe, in which high resistance to gentamycin (62.5%), piperacillin-tazobactam (50%) and amikacin (12.5%) was reported (Mbunga et al. 2018). Similar findings were also reported in a study done in Iran, which showed a high resistant to gentamycin (79%), ceftazidime (25%) and amikacin (83%) (Ekrami et al. 2011).

Acinetobacter spp as presented (in Table 7) was resistant to ceftazidime, cefoxitin, cefuroxime, ceftriaxone, cefepime and gentamycin. Hundred percent sensitivity to carbapenems, ampicillin, ciprofloxacin, amikacin and piperacillin-tazobactam was observed. These findings are in line with the findings from a study done in Iran, in which 100% (12/12) of *Acinetobacter* isolates were resistant to ceftazidime, imipenem and gentamicin (Shamsizadeh et al. 2017). This is due to the fact that, they produce a wide variety of β -lactamases, enzymes that degrade the β -lactam ring. This enzyme confers resistance to penicillins, cephalosporins, and carbapenems. They resist broad-spectrum cephalosporins by AmpC cephalosporinase enzyme that is encoded in chromosome (Nandi and Arjuna, 2017).

Conclusion

The isolation of pathogenic bacteria from inanimate surfaces in this study indicates that they can be vehicles for disease transmission. In the light of this, there is a need therefore for thorough disinfection and conscientious contact control procedures to minimize the spread of these pathogens in the neonatal intensive care unit, where interaction between patients, HCWs, parents and caregivers is very common and frequent. It is also necessary to encourage the effective use of disposable hand gloves between patients and to avoid touching inanimate surfaces with gloved hands, which may be acting as sources of infection.

The hospital environment is a complicated ecosystem and many interventions are needed for optimal infection control. Lack of a universal procedure for surveillance of nosocomial infection, poor hand hygiene, and high level of bacterial contamination on hospital environmental surfaces are the most important problems in hospital settings.

Acknowledgments

We would like to acknowledge the staff of the Windhoek Central Hospital Neonatal Intensive Care Unit and University of Namibia (School of Medicine) for their assistance in the research.

References

- Abreu AC, Tavares RR, Borges A, Mergulhao F, Simoes M (2013). Current and emergent strategies for disinfection of hospital environments. 68: 2718–2732. Doi:10.1093/jac/dkt281.
- Barma MM, Nasir IA, Babayo A (2017). Bacterial pathogens and their antibiotic susceptibility pattern in Intensive Care Units of the University of Maiduguri Teaching Hospital, Nigeria. *J Med Trop* 2017; 19:16-20. Doi: 10.4103/2276-7096.207587.
- Brito DVD, de Brito CS., Resende DS, Moreira DÓJ, Abdallah VOS, and Filho PPG (2010). Nosocomial infections in a Brazilian neonatal intensive care unit: a 4-year surveillance study. 43(6):633-637.
- Dramowski A, Aucamp M, Bekkera A, and Mehtar S (2017). Infectious disease exposures and outbreaks at a South African neonatal unit with review of neonatal outbreak epidemiology in Africa. 57 (2017) 79–85. <http://dx.doi.org/10.1016/j.ijid.2017.01.026>.
- Ekrami AR, Kayedani A, Jahangir M, Kalantar E, Jalali M (2011). Isolation of common aerobic bacterial pathogens from the environment of seven hospitals, Ahvaz, Iran. *Jundishapur J Microbiol.* 4 (2): 75-82.
- Gray J, and Omar N (2013). Nosocomial infections in neonatal intensive care units in developed and developing countries: how can we narrow the gap? 83 (2013) 193-195. <http://dx.doi.org/10.1016/j.jhin.2012.12.006>.
- Holma T (2015). Characterization of bacteria causing outbreaks by using molecular methods in a clinical microbiology laboratory. ISBN 978-951-51-1801-1.
- Ingale HD, Kongre VA, Bharadwaj RS (2017). A study of infections in neonatal intensive care unit at a tertiary care hospital. 4(4):1349-1356. DOI: http://dx.doi.org/10.18203/2349_3291.ijcp20172664.
- Krawczenko IS, Jankowska A, Kurylak A (2012). Healthcare-associated infections in a neonatal intensive care unit. Volume 8 (5): 854-858. DOI: 10.5114/aoms.2012.31412.
- Lalami AEO, Touiler H, El-Akhal F, Ettayebi M, Benchmesi N, Maniar S, Bekkari H (2016). Microbiological monitoring of environment surfaces in a hospital in Fez city, Morocco. *J. Mater. Environ. Sci.* 2016; 7 : (1):123-30.
- Lawn JE, Kerber K, Laryea CE and Cousens S (2010). 3.6 Million Neonatal Death. What Is Progressing and What Is Not? 34:371-386. Doi:10.1053/j.semperi.2010.09.011.
- Maryam A, Hadiza US, Aminu UM (2014). Characterization and determination of antibiotic susceptibility pattern of bacteria isolated from some fomites in a teaching hospital in northern Nigeria. Vol. 8(8): pp. 814-818, 2014. DOI: 10.5897/AJMR2013.6512.
- Mbanga J, Sibanda A, Rubayah S, Buwerimwe F, Mambozda K (2018). Multi-Drug Resistant (MDR) Bacterial Isolates on Close Contact Surfaces and Health Care Workers in Intensive Care Units of a Tertiary Hospital in Bulawayo, Zimbabwe. Vol 27(2): 1-15, 2018. DOI: 10.9734/JAMMR/2018/42764.
- Nandi D and Arjuna A (2017). Acinetobacter main cause of hospital acquired infections: a review. Volume 10 (5). DOI: <http://dx.doi.org/10.22159/ajpcr.2017.v10i5.17511>.
- Russotto V, Cortegiani A, Raineri SM, Giarratano A (2015). Bacterial contamination of inanimate surfaces and equipment in the intensive care unit. 3:54 (2015). DOI 10.1186/s40560-015-0120-5
- Saka KH, Akanbi AA, Obasa TO, Raheem RA, Oshodi AJ et al. (2016). Pathogenic aerobic bacterial contaminants on non-critical hospital surfaces within paediatric ward of a Nigerian Hospital. *J Med Microb Diagn* 5: 241. Doi: 10.4172/21610703.1000241.
- Sales VM, Oliveira E, Célia R, Gonçalves FR, Carvalho de Melo C (2014). Microbiological analysis of inanimate surfaces in an Intensive Care Unit and patient safety. Doi.org/10.12707/RIII1293.
- Segujja F, Mwambi B, Drago CK, Lubowa NM, Mungambwa J, Wabuyi P (2011). Characterization and Antimicrobial Susceptibility Patterns of Isolates from Ward Fomites. Vol 14(4): 1-16, 2016, Article no.BBJ.27861. DOI: 10.9734/BBJ/2016/27861.

Shamsizadeh Z, Nikaeen M, Esfahani BN, Mirhoseini SH, Hatamzadeh M, Hassanzadeh A (2017). Detection of antibiotic resistant *Acinetobacter baumannii* in various hospital environments: potential sources for transmission of *Acinetobacter* infections. Volume 22 (44). DOI 10.1186/s12199-017-0653-4.

Tajeddin E, Rashidana M, Razaghia M, Javadia SSS, Sherafata SJ, Alebouyeha M, Sarbazic MR, Mansourid N, Zaliba MR (2015). The role of the intensive care unit environment and health-care workers in the transmission of bacteria associated with hospital acquired infections. <http://dx.doi.org/10.1016/j.jiph.2015.05.10>.

Urzedo JE, Levenhagen MMMD, Pedroso RS, Abdallah VOS, Sabino SS and Brito DVD (2014). Nosocomial infections in a neonatal intensive care unit during 16 years: 1997-2012. 47(3):321-326. Doi.org/10.1590/0037-8682-0101-2014.

Yusuf JB, Okwongo OK, Mohammed A, Abubakar KS, Babayo A, Barma MM, Ibrahim S, Sulaiman AI, Hafiz H, Bello ZS (2017). Bacterial Contamination of Intensive Care Units at a Tertiary Hospital in Bauchi, Northeastern Nigeria. *American Journal of Internal Medicine*. Vol. 5 (3), 2017, pp. 46-51. Doi: 10.11648/j.ajim.20170503.13.