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MICROBIAL CONTAMINATION IN THE SELECTED OPERATING THEATRES AT KENYATTA NATIONAL HOSPITAL IN KENYA

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ABSTRACT

Background: Surveillance of operating theatres is essential to characterize and aid control and prevention of nosocomial infections spread to surgical patients due to pathogenic bacteria and fungi.

Aim: To characterize pathogenic bacteria and fungi in different operating theatres at Kenyatta National Hospital.

Methods: A total of 1,372 samples from 12 operating theatres were collected from December 2017 to February 2018. Surface samples were collected using sterile wet swabs from different equipment while exposed agar plates obtained aerial samples. One thousand two hundred (1,200) study samples and 172 study controls were processed. Colony-forming unit per cubic metre and settle plate methods enumerated and characterized bacterial and fungal isolates.

Findings: Coagulase-Negative *Staphylococci* 86(44.5%) and *Staphylococci* aureus 44(22.8%) predominated swab samples; air contaminant in agar plates was dominated by *Staphylococcus epidermidis* 185(73%) and *coliforms* (21%); Aspergillus spp 81(71.64%) was the major fungal isolate with Aspergillus fumigatus alone constituting 36 (31.9%).

Conclusion: The aforementioned microbial isolates contaminate and colonize the operating theatres. This may initiate infection to surgical patients. Thus, it is necessary to maintain and frequently monitor operating theatres to minimize growth and proliferation of pathogenic bacteria and fungi.

INTRODUCTION

There has been a rise in the volume of surgeries performed worldwide as a medical intervention. According to Rose et al., the global population of about 6.9 billion in 2010 needed at least 321.5 million surgical procedures (1). Similarly, according to data derived from 56 countries in 2004, about 187 to 281 million surgery operations were conducted, translating to one operation for every 25 human beings alive (2). This large volume of surgeries performed globally has implications to public health concern, as the annual volume of childbirths is merely half of the surgery operations conducted internationally (3). This demands high quality surgical care by the theatre personnel in the operating room to ensure survival and wellbeing of patients undergoing surgery (4). Surgical procedures as part of medical treatment are supposed to be conducted in a clean operating theatre and uncontaminated environment, since bacterial and fungal contamination of the environment increases the prevalence of contributing nosocomial infections, to surgery complications (5,6). The operating theatre environment plays an important role in the causation of postoperative infections, constituting a third of all nosocomial infections which can be minimized through preventive measures practised by theatre personnel and other infection control units (7-9).

A survey conducted in 14 countries by WHO attributed 8.7% prevalence of nosocomial infections to the hospitalization of patients in 55 hospitals (10). The nosocomial infections prolong hospital stay of affected patients, drain healthcare resources and may lead to loss of lives (1,11).

A number of bacterial and fungal species are associated with nosocomial infections, often with devastating effects to surgical patients. For instance, more than 1.4 million people worldwide have developed nosocomial bacterial infection complications due to resistant strains of the so-called 'superbugs.' Methicillinresistant Staphylococcus aureus (MRSA) is one of the superbugs and is a strain of bacterium which does not respond to methicillin drugs penicillin) (semi-synthetic and other antibiotics like oxacillin and flucloxacillin among others (12). Likewise, dissemination Candida of spp, in particular to immunocompromised patients is quite disturbing, as some Candida spp are resistant to conventional antifungal drugs, raising possibilities of losing lives of patients while undergoing treatment (13,14).

The colonization of theatre personnel as as patients and visitors in the well healthcare facilities with MRSA and Candida spp facilitates transmission of infections (12, 15).In these addition, Aspergillus spp spores in poorly aerated healthcare facilities may serve as a source of nosocomial infection to surgical patients (16). MRSA and Candida spp can be harboured in the mucosal membranes of the throat, nostrils, and even on the skin in these asymptomatic carriers. From the asymptomatic persons, MRSA enters the susceptible patients through the damaged or diseased skin, which may lead to dermatitis, eczema or chronic wounds; inadvertent spread of Candida spp from unprotected carriers to susceptible patients may result in candidiasis manifesting in different forms such as oral thrush. Likewise, the route for *Aspergillus* spores entry is mainly inhalation, resulting in nasopharyngeal colonization, ensuing pulmonary infection to the susceptible patients. The unclean theatre environment, its surfaces or shared patient equipment and other items are also reservoirs for the aforementioned nosocomial infection (15–17).

Notwithstanding efforts by public health and infection control units, nosocomial infections have persisted to arise in hospitalized patients. Lowered immunity in hospitalized patients is a factor contributing to persistent nosocomial infections. Medical procedures and invasive techniques during surgical procedures create potential routes for pathogenic invasion; hence, the transmission of drug-resistant pathogens among patients (10). Modern surgical procedures and therapeutic interventions are also sources of nosocomial fungal and bacterial infections (18).

Reducing the burden of nosocomial infections is a necessity, and the operating theatres are at the centre of such efforts, surgical procedures, with since both environmental impact and equipment like anaesthetic apparatus, operating devices or instruments, theatre personnel among others occurring there may be a source of sepsis to patients (7). Infection Control Units and other mandated departments in hospital facilities, both in developing and developed countries ought to continue playing an essential role to avert mortalities caused by nosocomial infections. Use of preventive measures and treatments designed to lower infection in healthcare facilities and theatres ought to be prioritised in reducing such infections (17).

Taken together, infection control and surveillance are supposed to be conducted periodically to assess the general status of theatres, necessary for infection prevention. The meticulous effort by use of infection control measures in theatres is essential to minimize postoperative sepsis. This is particularly useful in developing countries where general health care systems are quite low. To this end, the study endeavoured to characterize the major bacterial and fungal isolates in some of the operating theatres at Kenyatta National Hospital. This was expected to establish the functionality of the infection control measures deployed at this healthcare facility-largest referral hospital in East and Central Africa.

MATERIALS AND METHODS

Study Setting Area

The study was performed at Kenyatta National Hospital (KNH), situated at Upper Hill area of Nairobi, the capital city of Kenya. The hospital has a 2000 bed capacity and receives patients from all the 47 Kenyan county referrals and their associated county hospitals and patient referrals from East and Central African countries.

Study population

KNH has 12 operating theatres, one receiving area, and one recovery ward. Each of the 12 theatres has one operating bed, one scrubbing area, one anaesthetic room, 6 sterile instruments setting areas as each is shared by two theatres and two sterile instruments stores serving all theatres with three sluice rooms each serving 4 theatres.

Study design and sample collection

This was a hospital-based cross-sectional study where 1200 study samples and 172 internal study controls were collected twice per month between December 2017 and February 2018. This was to ensure adequate coverage of at least all the operating theatres. The study samples and internal study controls in each specified area were collected early in the morning when all areas had been cleaned adequately ready for the daily operating procedures. The study samples and internal study controls were obtained in the following designated theatres: Emergency theatre 1, Reproductive health theatre 2, Urology theatre 3 and 6, Orthopaedic theatre 4 and 5,Specialized theatre 7, Ear, Nose and Throat theatre 8, Neurology theatre 9, Amenity theatre 10, Cardiothoracic theatre 11 and Paediatric theatre 12.

Swab method

A swab soaked in the sterile nutrient broth was used to collect samples from the 12 selected operating theatres, anaesthetic machines, recovery room patient monitors, operation tables, suction bottles and receivers used operating for room These were retrieved from procedures. operating beds, anaesthetic machines, suction bottles, and post-anaesthetic care unit patient monitors. All the samples were labelled and properly immediately transported in a sterile manner to the Department of Medical Microbiology, University of Nairobi, where standard bacterial and fungal identification and characterization was done.

Settle plate method

Sterile agar plates facilitated the collection of airborne microbial organisms. This was done by exposing the agar plates to air freely circulating in the 12 designated operating theatres. The agar plates were placed in specific areas (sterile setting area, sterile theatre stores, scrubbing areas, operating theatres, receiving areas, postanaesthetic care unit, and anaesthetic room and sluice rooms), and exposed for 30 minutes to maximize exposure time thereby allowing the collection of air sediment biological particles. After this exposure, the agar plates were taken back using a sterile technique, covered and ferried to the Department of Medical Microbiology, University Nairobi for standard of microbiological laboratory processing.

Internal quality controls

The quality of exposure agar plates was assessed by including closed sterile agar plates which were not exposed and incubated alongside sampling agar plates for exposures from the 12 operating theatres. Sterile swabs were used and also soaked in sterile normal saline. Positive controls included known bacterial cultures of: Staphylococcus aureus (ATCC29213/ATCC25922), E.coli (ATCC25922), and Pseudomonas aeruginosa (ATCC25923), obtained from the Department of Medical Microbiology, University of Nairobi, where bacterial and fungal growth and identification was conducted.

Standard Bacterial and Fungal Laboratory processing

Internal study controls and study samples isolated from different theatre environment and equipment surfaces were streaked on blood agar plates and Mac-Conkey agar for bacterial growth, and Sabouraund Dextrose Agar for fungal growth. These culture plates were incubated under aerobic conditions for 48 hours at 37°C for bacteria and at 25°C for 5 days for fungi. After incubation, the colonies were counted, and identification of isolates done based on their macroscopic colonial morphological characteristics and biochemical tests. Microbial growth count was expressed in terms of colony-forming unit per cubic metre (cfu/ m³), calculated by the aid of the following formula: $cfu/m^3 = a \times a^3$ 1000 p × t × 0.2; where a = number of colonies on settle plate, p = surfacemeasurement of plate used, t = time of exposure of settle plate.

Data management and analysis

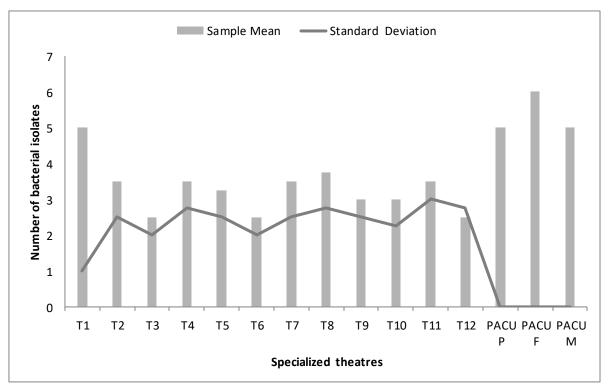
Data was entered, edited, and analysed by Microsoft Excel 2007 (Microsoft Corporation, Redmond, WA, USA). Descriptive statistics incorporated sample collection sites and the different samples collected.

lable 1								
Microbial isolates Swabbed during the study period								
Microbial Isolates	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6		
	N (%)							
No growth	16(31.4)	22(43.1)	21(41.2)	16(31.4)	16(31.4)	22(43.1)		

RESULTS

Tabla 1

Coagulase-negative staphylococci	15(29.4)	14(27.5)	20(39.2)	9(17.6)	11(21.6)	11(21.6)
Staphylococcus aureas	5(9.8)	5(9.8)	3(5.9)	6(11.8)	11(21.6)	7(13.7)
Micrococcus spp	-	2(3.9)	-	7(13.7)	2(3.9)	1(2.0)
Klebsiella spp	3(5.9)	2(3.9)	1(2.0)	-	1(2.0)	-
Pseudomonas spp	2(3.9)	-	1(2.0)	1(2.0)	3(5.9)	-
Candida spp	1(2.0)	-	-	-	1(2.0)	1(2.0)
Bacillus spp	2(3.9)	1(2.0)	-	-	1(2.0)	-
E. feacalis	-	1(2.0)	-	-	2(3.9)	-
E. coli	-	-	1(2.0)	-	-	-
Granulicatella elegans	-	-	-	1(2.0)	-	-
Coagulase-negative staphylococci and Staphylococcus aureas	4(7.8)	2(3.9)	3(5.9)	4(7.8)	2(3.9)	5(9.8)
Coagulase-negative <i>staphylococci</i> and <i>Micrococcus spp</i>	-	-	-	2(3.9)	-	-
Coagulase-negative staphylococci and Klebsiella spp	-	1(2.0)	1(2.0)	-	-	-
Coagulase-negative <i>staphylococci</i> and <i>Candida spp</i>	1(2.0)	-	-	-	-	-
Coagulase-negative <i>staphylococci</i> and <i>Bacillus spp</i>	1(2.0)	-	-	-	-	-
Coagulase-negative <i>staphylococci</i> and <i>E. feacalis</i>	-	-	-	-	-	1(2.0)
Staphylococcus aureas and Micrococcus spp	-	-	-	2(3.9)	1(2.0)	-
Staphylococcus aureas and Klebsiella spp	-	-	-	-	-	1(2.0)
Staphylococcus aureas and Pseudomonas spp	-	-	-	1(2.0)	-	-
Staphylococcus aureas and E. feacalis	-	-	-	-	-	1(2.0)
Staphylococcus aureas and E. coli	-	-	-	1(2.0)	-	-
Pseudomonas spp and Candida spp	-	-	-	1(2.0)	-	-
Candida spp and Bacillus spp	1(2.0)	-	-	-	-	-
E. feacalis and E. coli	-	1(2.0)	-	-	-	-
Staphylococcus aureas, Klebsiella spp and E. feacalis	-	-	-	-	-	1(2.0)



Only Candida spp was isolated singly or part of other swabbed bacterial isolates.

Figure 1: Bacterial growth pattern in different theatres within the entire study period

KEY:T1-Theatre 1 (Emergency);T2-Theatre 2 (RH);T3-Theatre 3 (Urology);T4-Theatre 4 (Orthopaedics);T5-Theatre 5 (Orthopaedics);T6-Theatre 6 (Urology);T7-Theatre 7 (Specialized);T8-Theatre 8 (Ear, Nose and Throat);T9-Theatre 9 (Neurology) T10-Theatre 10 (Amenity);T11- Theatre 11 (Cardiology) ;T12-Theatre 12 (Paediatric);PACU P-Post anaesthetic care unit for paediatrics; PACU F-Post anaesthetic care unit for female; PACU M-Post anaesthetic care unit for male

There was no significant difference in mean growth between different specialized theatres.

	Agar Plate Bacterial isolates				-
1 St Week		Up to 5	6-15	Above 15	
	Microbial Isolate	cfu/m ³	cfu/m ³	cfu/m ³	Total
	Staphylococcus albus	41	8	-	49
	Environmental Bacilli	1	-	-	1
	Coli-form	7	-	-	7
	No Growth	19	-	-	19
	Staphylococcus albus	30	3	-	33
2 nd	Environmental Bacilli	-	-	-	-
Week	Coli-form	5	-	1	5
	No Growth	31	-	-	31
	Staphylococcus albus	13	28	1	42
3rd	Environmental Bacilli	-	-	-	-
Week	Coli-form	7	-	-	7
WCCK	No Growth	19	-	-	19
	pseudomonas aeruginosa	-	1	-	1
	Staphylococcus albus	37	7	-	44
4^{th}	Environmental Bacilli	4	-	-	4
4 th Week	Coli-form	15	-	-	15
	No Growth	26	-	-	26
	pseudomonas aeruginosa	-	-	-	-
	Staphylococcus albus	16	1	-	17
5 th	Environmental Bacilli	3	-	-	3
5 th Week	Coli-form	4	-	-	4
	No Growth	42	-	-	42
	Pseudomonas aeruginosa	-	-	-	-
6 th Week	Environmental Bacilli	5	-	-	5
	Coli-form	15	1	-	16
	No Growth	46	-	-	46
	Pseudomonas aeruginosa	-	-	-	-
	Staphylococcus albus	1	-	-	1
	Fungi	2	-	-	2
Total	1. (439

 Table 2

 Agar Plate Bacterial isolates analysis in the three months study period

Key: CFU-Colony forming unit; M³-cubic metres;-means nil

A	Agar plate Bacterial isolate		during study period Settle rate	
		Colony-Forming Units		
	Up to 5 Cfu's	Above 5 Cfu's	Up to 0.5	Above 0.5
Study Area	N (%)	N (%)	N (%)	N (%)
PACU, Stores and Rece	iving			
area	7(53.8)	6(46.2)	13(100.0)	-
Theatre 1	12(54.5)	10(45.5)	20(100.0)	-
Theatre 2	19(65.5)	10(34.5)	28(100.0)	-
Theatre 3	11(73.3)	4(26.7)	13(100.0)	-
Theatre 4	9(90.0)	1(10.0)	9(100.0)	-
Theatre 5	8(100.0)	-	8(100.0)	-
Theatre 6	11(73.3)	4(26.7)	13(100.0)	-
Theatre 7	10(66.7)	5(33.3)	15(100.0)	-
Theatre 8	9(64.3)	5(35.7)	12(92.3)	1(7.7)
Theatre 9	13(76.5)	4(23.5)	15(93.8)	1(6.3)
Theatre 10	11(100.0)	-	10(100.0)	-
Theatre 11	10(90.9)	1(9.1)	10(100.0)	-
Theatre 12	13(81.3)	3(18.8)	15(93.8)	1(6.3)
Setting area 1 and 8	3(75.0)	1(25.0)	4(100.0)	-
Sluice room 1 and 2	3(75.0)	1(25.0)	4(100.0)	-
Setting area 2 and 7	2(50.0)	2(50.0)	4(100.0)	-
Sluice room 3 and 4	6(85.7)	1(14.3)	7(100.0)	-
Setting area 3 and 6	5(71.4)	2(28.6)	7(100.0)	-
Setting area 4 and 5	1(100.0)	-	1(100.0)	-
Sluice 5 and 6	5(100.0)	-	5(100.0)	-
Sluice room 7 and 8	5(71.4)	2(28.6)	7(100.0)	-
Sluice room 9 and 10	2(40.0)	3(60.0)	5(100.0)	-
Setting area 9 and 12	5(83.3)	1(16.7)	6(100.0)	-
Setting area 10 and 11	2(100.0)	-	1(100.0)	-
Sluice room 11 and 12	4(50.0)	4(50.0)	7(100.0)	-

 Table 3

 Avar nlate Bacterial isolate levels in KNH Theatres during study period

Key: PACU-Post Anaesthetic care unit; CFC-Colony forming unit;-means nil

Fungal isolates during three months period							
Isolated fungal species	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	
Acremonium spp	-	-	2	2	2	1	
Aspergillus fumigatus	15	-	-	_	-	-	
Aspergillus niger	1	-	-	-	-	-	
Aspergillus spp	2	-	14	15	2	12	
Aspergillus spp and candida spp	-	-	1	_	-	-	
Aspergillus terreus	4	-	-	-	-	-	
Candida spp and Aspergillus spp	-	-	-	-	-	1	
Penicillin spp and Fusarium spp	-	-	1	-	6	-	
Yeast	-	-	-	-	1	-	

 Table 4

 Fungal isolates during three months period

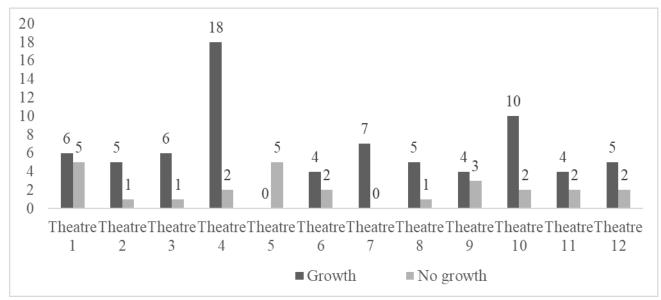


Figure 2: Fungal growth pattern across different operating theatres

DISCUSSION

This study observed Coagulase-negative staphylococci (44.5%), staphylococci aureus (22.8%), and Granulicatella elegans (bacterial species) in the selected operating theatres at Kenyatta National Hospital. Gelaw et al. also coagulase-negative staphylococci isolated (68.3%) and staphylococci aureus (30.7%) from the hospital environment, patients and staff at the University of Gondar Hospital, Northwest Ethiopia. Contamination and colonization of the theatre environment by the aforementioned bacterial species may contribute to nosocomial infections (19). Similarly, a Nigerian study observed higher levels of coagulase-negative Staphylococci (28.3%) compared to staphylococci aureus (0.83%) in surgical wards in Obafemi Awolowo University Teaching Hospital Complex, Ile-Ife, Nigeria. Conversely, the level of Pseudomonas aeruginosa (23.3%) by the Nigerian study was slightly higher compared to the level of Pseudomonas aeruginosa (4.1 %.) observed by this study (20). Moreover, A. Mishra et al. observed coliforms in delivery theatres, attributing it to the normal flora in the gut which may contaminate the theatres during delivery (11). Furthermore, J.lapins et al. linked coagulase-negative *staphylococci* to skin lesions, as it was identified in suppurativa lesions of study participants. The levels of aforementioned the bacterial species contaminate and colonize the theatre environment, contributing to nosocomial infections (21). In addition, J. P. Casalta et al. characterised Granulicatella elegans using broad-range PCR primers, linking it with infectious endocarditis (22).

This study also isolated Aspergillus spp (39.8%), with Aspergillus fumigatus alone constituting (31.9%) of the Aspergillus species isolated. Candida spp (<1%) was the least observed fungal isolate. The mixed growths of Penicilium spp (9.7%) were also identified. Similarly, A. Gniadek et al. observed predominant presence of Aspergillus species among other pathogenic hospital operating fungi in theatre environment in one of the hospitals at Kraków in Poland. The study implied negligence to decontamination processes by the hospital due to the significant increase of Aspergillus species (23).Moreover, E.O.Akinkunmi et al. observed Candida spp

in the surgical wards in Obafemi Awolowo University Teaching Hospital Complex, Ile-Ife, Nigeria among bacterial isolates (20). Oliver et al. observed *Penicilium spp* and *Aspergillus spp*, and demonstrated that the effectiveness of air-handling systems in the hospital environments dramatically reduced presence of the aforementioned fungal species (24).

The studied microbial isolates in the selected operating theatres had mean settle rate of < 0.5. The colony-forming units were less than 5cfu/m³ in all the operating areas where bacterial and fungal isolates were collected, and less than 30% between 6cfu/m³ and 15cfu/m³, which was within acceptable clean operating theatres as set by Dancer (25).

Microbial levels within the operating theatre may differ over time depending on the contaminants. Therefore, what was captured over the 3 month study period may be limited in the representation of the entire period of theatre status. Moreover, this study only concentrated in the main operating theatres at Kenyatta National Hospital excluding peripheral operating theatres which may have different microbial distribution.

CONCLUSION

This study observed bacterial and fungal isolates levels less than 10cfu/m³ and settle rates were also less than 0.5, which were within internationally acceptable levels. This implies infectious control measures deployed by Kenyatta National Hospital are functional and need to be maintained. However, the presence of pathogenic Staphylococci spp and Aspergillus spp evidenced by this study necessitates more effective cleaning and disinfection of equipment and floors to minimize the spread of the aforementioned pathogenic microbes to susceptible surgical patients. Furthermore, regular surveillance of microbial loads on equipment and theatre personnel in the operating theatres at Kenyatta National Hospital is necessary to aid the monitoring and institute control measures to avert any spread of aforementioned pathogenic microbes to susceptible surgical patients.

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