Journal of Advances in Microbiology

15(3): 1-8, 2019; Article no.JAMB.40632
ISSN: 2456-7116

Microorganisms Isolated from Hospital Environmental Surfaces in Akure Metropolis, Ondo State, Nigeria

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Authors' contributions

This work was carried out in collaboration between both authors. Authors OGP and AKO designed the study, author OGP carried out the research. Author AKO supervised the research, author OGP arranged the result author OGP performed the statistical analysis, wrote the protocol, author OGP wrote the first draft of the manuscript. Author OGP managed the analyses of the study. Author OGP managed the literature searches. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMB/2019/v15i330104
Editor(s):
(1) Dr. Adekunle Sanyaolu, Epidemiology Division, Nigeria Center for Disease Control, Federal Ministry of Health, Abuja, Nigeria.
Reviewers:
(1) Dr. R. Jasmine, Bishop Heber College, Tamil Nadu, India.
(2) Maria T. Roosanova, Argentina.
Complete Peer review History: http://www.sdiarticle3.com/review-history/40632

Original Research Article

Received 07 January 2018
Accepted 29 March 2018
Published 27 March 2019

ABSTRACT

Nosocomial infection is a rising problem in developing countries. Microorganisms Isolated from three private and public hospital environmental surfaces in Akure Metropolis, Ondo State, Nigeria were investigated in this study. Bacterial and fungal organisms were isolated and compared among the three hospitals. The study revealed that bacteria were the most predominant microorganisms found in the hospital environmental surfaces than fungi. *Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli, Pseudomonas aeruginosa, Klebsiella pneumoniae* and *Bacillus cereus* were the bacterial isolates while fungi include *Aspergillus fumigatus, Aspergillus niger* and *Candida albicans. Staphylococcus aureus* was found to be predominant bacteria but *Aspergillus funmigatus* was the predominant fungi. The result showed that the microbial loads of the public hospitals were higher than that of the private hospitals. The bacteria load of the male ward was found to be higher than that of the female ward while the fungal loads of each of the hospital environmental surfaces of female were higher than that of the male. The study revealed that bacteria were the most

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predominant microorganisms found in the hospital environment than fungi. *Staphylococcus aureus, Streptococcus pyogenes, Escherichia coli, Pseudomonas aeruginosa, Klebsiella pneumoniae* and *Bacillus cereus* were the bacterial isolates while fungi include *Aspergillus fumigatus, Aspergillus niger* and *Candida albicans. Staphylococcus aureus* was found to be predominant bacteria. All the hospital environmental surfaces were contaminated with one or more microorganisms in the course of the research.

Keywords: Hospital; ward; environmental; bacteria; fungi.

1. INTRODUCTION

Nosocomial infection is an infection occurring in a patient in a hospital or other health care facility in whom the infection was not present or incubating at the time of admission. It is estimated that in developing countries, nosocomial infections concern above 25% of hospitalized patients, and in the developed countries from 5 to 10% [1]. This includes infections acquired in the hospital but appearing after discharge, and also occupational infections among staff of the facility [2]. The sources of infections can be: patients, medical personnel, visitors or parts of the environment: Equipment and hospital items, also arthropods inhabiting hospitals.

Patient care is provided in facilities which range from highly equipped clinics and technologically advanced university hospitals to front-line units with only basic facilities [3]. Despite progress in public health and hospital care, infections continue to develop in hospitalized patients, and may also affect hospital staff. Many factors promote infection among hospitalized patients: Decreased immunity among patients; the increasing variety of medical procedures and invasive techniques creating potential routes of infection; and the transmission of drug-resistant bacteria among crowded hospital populations, where poor infection control practices may facilitate transmission [3].

Hospital acquired infection is an additional affliction to the patient admitted to the hospital for some serious illness and is caused by pathogens which are prevalent in hospital environment [4]. In the hospital, microbes are ubiquitous; and can reach the sick patient through various sources, such as air, water, food, contaminated equipment, linen, catheters, scopes, ventilators, contaminated disinfectants and other preparations used for treatment, visitors, infected patients, etc. [4]. Recently, the probable involvement of surfaces and equipment from the hospital environment as a disseminating source of pathogens, including resistant bacteria, has been highlighted [5]. There are no meaningful standards for permissible levels of microbial contamination of inanimate surfaces in hospital environment, but an increased microbial load on surfaces may imply the possibility of finding a pathogen [6]. Microorganisms that are often associated with hospital acquired infections are *Staphylococcus aureus, Micrococcus* sp., *Pseudomonas* sp., *Proteus* sp., *Escherichia coli*, *Enterobacter*, *Bacillus cereus*, *Cladosporium* sp., *Aspergillus* sp., and viruses [7]. *Pseudomonas aeruginosa* has been particularly incriminated in nosocomial infection because of its intrinsic resistance to most antibiotics and its ability to survive and multiply at low temperatures and in disinfectant solutions [8]. This study aimed at isolating and comparing bacterial and fungal microorganisms isolated from the environmental surfaces of three public and private tertiary healthcare institutions in Akure metropolis, Ondo state, Nigeria.

2. MATERIALS AND METHODS

2.1 Description of Study Location

This research work was carried out from September 2016 to April, 2017 in Akure metropolis, Ondo state, Nigeria. Akure covers an area of 14,798.8 ,993.7 square kilometers and lies at latitude 7°15′0″N, 7°11′N 5°11′42″E and longitude 5°11′42″E, 5°35'E. Akure is one of the 18 local government areas of Ondo State with a population of 484,798 based on the 2006 population census. It is situated in the peripheral zone of the rainforest of Ondo state. Akure is the administrative capital of Ondo state. Akure lies about 70°15' north of the equator and 50°15' east of the Meridian. It is about 700 km Southwest of Abuja and 311 km north of Lagos State. The town is situated in the tropic rainforest zone in Nigeria.

2.2 Collection of Samples

Samples were collected by swab sticks from Male Accident and Emergency Bed, Female Accident and Emergency Bed, Male Toilet, Female Toilet, Male Surgical Ward Chair, Female Surgical Ward Chair, Male Medical Ward Floor, Female Medical Ward Floor, Male Ward Air flora, Female Ward Air flora, Theatre Couch, Injection Room Tables, Neonatal Ward Couch and Maternity Ward Couch from Health Centre FUTA, Don Bosco Catholic Hospital and State Specialist Hospital Akure. The date, time, conditions and sites of sampling were noted. Basically, swabs were used, at least, for each sampling site. For sampling, swabs were moistened in 2 ml sterile saline solution and rolled several times over a surface area of around 25 cm^2, and the swab sticks were transported to the laboratory. Sampling was always done between 8-10 am.

2.3 Isolation of Microorganisms from Hospital Environment

Isolation of microorganisms from hospital environment was carried out as described by Bakkali et al. [9] with slight modification. Basically, swabs were used, at least, for each sampling site. For sampling, swabs were moistened in 2 ml sterile saline solution and rolled several times over a surface area of around 25 cm^2, and the swab sticks were transported to the laboratory. Sampling was always done between 8-10 am. A five-fold serial dilution was made and 0.1 ml of the 10^{-3} and 10^{-5} dilutions were uniformly pour-plated onto 14 cm diameter wide agar plates and of nutrient agar, Potato dextrose agar, MacConkey agar and EMB agar.

2.4 Characterization of Bacterial Isolates

The pure culture of each isolate was examined. Microscopic examination, staining techniques and biochemical tests were carried out on the isolates according to the methods described by Olutiola et al. [10] and Cheesbrough [11].

2.5 Identification of Fungal Isolates

Fungal isolates were characterized and identified based on macroscopic and microscopic details with reference to Barnett and Hunter [12].

2.6 Statistical Analysis of Data

All experiments were carried out in triplicate, and data obtained were subjected to one way analysis of variance, while the means were compared by Duncan's New Multiple Range Test at 95% confidence interval using Statistical Package for Social Sciences version 16.0. Differences were considered significant at $p \leq 0.05$.

3. RESULTS

Table 1 the bacterial load of each of the items isolated from different hospital environmental surfaces is shown in Table 1, it was observed that bacterial load of the toilet were higher than other surfaces, while the bacterial load from each of the male hospital environmental surfaces was higher than that of the female hospital environmental surfaces. It was also observed that the bacterial loads isolated from government own hospital was higher than those microorganisms isolated from private hospital.

Table 2 the fungal load of each of the fungi isolated from different hospital environmental surfaces are shown in Table 2, it was observed that fungal load of the toilet was found to be higher than other surfaces, while the fungal load isolated from each of the female hospital environmental surfaces was higher than that of the male hospital environmental surfaces. It was also observed that the fungal loads isolated from government own hospital was higher than those microorganisms isolated from private hospital.

Table 3 the rate of occurrence of different bacteria isolated from different hospital environmental surfaces is presented in Table 3. It was observed that *Staphylococcus aureus* had the highest rate of occurrence, while *Esherichia coli* had the lowest rate of occurrence out of the bacteria isolated for different hospital environment surfaces.

Table 4 the rate of occurrence of different fungi isolated from different hospital environmental surfaces is presented in Table 4. It was observed that *Aspergillus fumigatus had* the highest rate of occurrence followed by *Candida albicans* and *Aspergillus niger* which share the same number percentage positivity.

Table 1. Bacterial load of hospital environmental surfaces

Study area(source)	FUTA Health Centre (Cfu/ml)	State Specialist Hospital (Cfu/ml)	Don Bosco Hospital (Cfu/ml)
MAEB	4.1×10^4	4.6×10^4	2.0×10^3
FAEB	2.9×10^4	3.1×10^4	1.0×10^3
MT	TNC	TNC	TNC
FT	TNC	TNC	TNC
MSWC	ND	4.5×10^4	3.0×10^3
FSWC	ND	2.0×10^3	1.0×10^3
MMWF	3.3×10^4	4.1×10^4	3.0×10^3
FMWF	3.0×10^3	3.4×10^4	2.0×10^3
MWA	3.9×10^4	4.5×10^4	3.0×10^3
FWA	3.0×10^3	5.0×10^3	2.0×10^3
TC	ND	2.0×10^3	1.0×10^3
IRT	3.0×10^3	4.0×10^3	1.0×10^3
NWC	3.0×10^3	4.0×10^3	2.0×10^3
MWC	4.0×10^3	5.0×10^3	2.0×10^3

Legend: Not Determine (ND), No Growth (NG), Male Accident and Emergency Bed (MAEB), Female Accident and Emergency Bed (FAEB), Male Toilet (MT), Female Toilet (FT), Male Surgical Ward Chair (MSWC), Female Surgical Ward Chair (FSWC), Male Medical Ward Floor (MMWF), Female Medical Ward Floor (FMWF), Male Ward Air flora (MWA), Female Ward Air flora (FWA), Theatre Couch (TC), Injection Room Tables (IRT), Neonatal Ward Couch (NWC) and Maternity Ward Couch (MWC)

Table 2. Fungal load of hospital environmental surfaces

Study area(source)	FUTA Health Centre (Sfu/ml)	State Specialist Hospital (Sfu/ml)	Don Bosco Hospital ((Sfu/ml))
MAEB	2.0×10^3	1.5×10^4	1.0×10^3
FAEB	3.0×10^3	3.0×10^3	2.0×10^3
MT	4.0×10^3	4.4×10^4	3.0×10^3
FT	6.0×10^3	7.1×10^4	4.0×10^3
MSWC	ND	NG	NG
FSWC	ND	NG	NG
MMWF	1.0×10^3	2.0×10^3	NG
FMWF	2.0×10^3	4.0×10^3	1.0×10^3
MWA	2.0×10^3	3.0×10^3	NG
FWA	3.0×10^3	4.0×10^3	1.0×10^3
TC	ND	NG	NG
IRT	NG	NG	NG
NWC	2.0×10^3	3.0×10^3	1.0×10^3
MWC	NG	NG	NG

Legend: Not Determine (ND), No Growth (NG), Male Accident and Emergency Bed (MAEB), Female Accident and Emergency Bed (FAEB), Male Toilet (MT), Female Toilet (FT), Male Surgical Ward Chair (MSWC), Female Surgical Ward Chair (FSWC), Male Medical Ward Floor (MMWF), Female Medical Ward Floor (FMWF), Male Ward Air flora (MWA), Female Ward Air flora (FWA), Theatre Couch (TC), Injection Room Tables (IRT), Neonatal Ward Couch (NWC) and Maternity Ward Couch (MWC)

4. DISCUSSION

Hospital associated infections have been linked with many factors among which is the microbial quality of the indoor air of different wards and units of each hospital [13]. This type of infection occurs in 5% of all acute care hospitalization in the United State and has been reported to be responsible for the death of one out of every five thousand patients attending an American hospital [14]. In Nigeria, the rate of nosocomial infection ranges between 2.7%-3.8% [15]. Hence, this requires every possible measure to control the rise including [among other investigations) examining the quality of indoor air of the hospital wards and units. Each of the hospital environmental surfaces was contaminated with microorganisms.

Table 3. Rate of occurrence of different bacteria isolated from FUTA Health Centre, State Specialist hospital Akure and Don Bosco Hospital Akure

Bacteria	Number of surfaces tested positive	Percentage positivity (%)
Staphylococcus aureus	39	22.81
Streptococcus pyogenes	24	14.04
Escherichia coli	21	12.28
Pseudomonas aeruginosa	27	15.79
Klebsiella pneumonia	33	19.30
Bacillus cereus	27	15.79
Total	171	100.01

Table 4. Rate of occurrence of different fungi isolated from FUTA Health Centre, State Specialist hospital Akure and Don Bosco Hospital Akure

Fungi	Number of surfaces tested positive	Percentage positivity (%)
Aspergillus fumigatus	21	36.84
Aspergillus flavus	18	31.58
Candida albicans	18	31.58
Total	57	100

Bacteria were found to be more predominant than fungi, the bacteria isolated from the hospital surfaces were *Bacillus cereus, Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Staphylococcus aureus,* and *Streptococcus pyogene s*while fungi include *Aspergillus fumigatus, Aspergillus niger* and *Candida albicans. Staphylococcus aureus* was found to be predominant bacteria with the occurrence of 22.81%, this correlate with the report of Awosika et al. [16] who reported *Staphylococcus aureus* as the most frequently isolated bacterium from hospital surface. *Staphylococcus aureus* as the most frequently isolated bacterium from hospital surface has been incriminated in various diseases such as post-operative infections, urinary tract infections, skin infections, respiratory infections and food poisoning [17]. Proper control measures, such as increase in hygiene, are required to combat infections by *Staphylococcus aureus* in these hospital wards and units [16]. The occurrence of bacteria in hospitals has been commonly related to some possible sources of dissemination: bottle soap [18], hands of healthcare professionals [19], gloves and gowns [20], mobile phones [21] paper money and coins [22]. *Aspergillus fumigatus* was found to be predominant fungi with frequency occurrence of 36.84%, this correlate with the report of Cagginao et al. [23] who reported that *Aspergillus fumigatus* was the most commonly isolated [68.5%].

The bacterial load of the male ward was found to be higher than that of the female, this could be due personal hygiene of the female, this contrary with the report of Ekhaise et al. [7] who reported that a quantitative study of different hospital units showed that the children ward and female ward had the highest total bacterial count followed by the bacteriology laboratory.

The fungal loads of each of the hospital environment surfaces of female were higher than that of the male. In hospital environments, airborne molds are a potential risk for patients because of possible inhalation of conidia [24]. Because surgical procedures expose patients to infective complications, the operating theater is considered a complex habitat in which all sources of pollution have to be kept under control [25,26]. In particular, the widespread presence of *Aspergillus* spp. is the major extrinsic risk factor for invasive aspergillosis, caused by *A. fumigatus* and other species of *Aspergillus*, such as *A. flavus*, *A. niger*, and *A. terreus*, depending on the local epidemiology [27] and according to the season [28].

The microbial load of the public hospital were higher than that of the private, this tallied with the report Ekhaise et al. [7] who reported high microbial counts recorded for the public hospital (Central Hospital) as compared to private hospital (Faith Medical Center), could be due to the subsidized user fees of the public hospital,

thereby accommodating more people, as compared to the private hospital, where higher user fees are charged and are not within the reach of the poor people in the society. These findings could be explained by many factors including the number of visitors visiting the children and female wards, which exceeded visitors in other hospital units. It was noted that the amount of materials brought from outside such as personal belongings, food and fruits were more common in children and female wards. These are recognized as sources of hospital contamination. Hospital surfaces are contaminated by factors inherent to the presence of patients, such as biological fluids, sometimes associated with invasive and non-invasive techniques and hygiene. Another contamination factor would be the circulation of vectors as carrier agents for fungi and bacteria resistant to antimicrobials [29,30].

The microbial load of the theater couch and surgical ward were found to be the lowest, this could probably be due to the fact that there is high sanitary standards in this area as compared to other hospital areas and also that the theater is a restricted area, which tallied with the results of Ekhaise et al. [7] who reported that the number of microorganisms in the theater was extremely low.

Although, surfaces are not directly connected to transmission in most hospital infections, the impact of hygiene and cleaning procedures in microbial control is evident. It is suggested that microorganisms associated to hospital infections are able to survive during large periods of time, thus being a continuous source of contamination in cases where population control is not efficiently conducted [31,32].

Regular surveillance, cleaning and restriction of patient's relatives might be among the strict measures necessary to reduce the microbial load of indoor air in this hospital wards and units [16].

5. CONCLUSION

This investigation has been able to identify and prove the sensitivity patterns of micro-organism isolated from hospital environment surfaces. The study has shown that all the hospital environmental surfaces examine in the course of the study in Akure, Nigeria were contaminated with one or more microorganisms.

ACKNOWLEGEMENT

The Author wish to acknowledge Mr. ADEGOKE, Tosin Victor for his contribution towards the research.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle3.com/review-history/40632