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Prevalence of pathogenic organism associated with hospital environment (a case study of Ile-Abiye hospital, Ado-Ekiti)

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Abstract

Microbial contamination of the hospital environment has continued to increase the prevalence of nosocomial infections. This study assessed the level of microbial contamination of hospital environment: a case study of Ile-Abiye Hospital, Ado-Ekiti. Swab plate method was used to collect samples from bed surfaces. Collected samples were analyzed for the presence of bacteria using standard procedures. The entire swab specimens had bacterial growth. The bacteria isolates were, *Staphylococcus aureus* 12(62.5%), *Escherichia coli* 5(12.5%), *Staphylococcus epidermidis* 3(7.5%), and *Lactobacillus fermenti* 3(7.5%), *Bacillus subtilis* 2(5%), *Bacillus cereus* 1(2.5%), and *Micrococcus luteus* 1(2.5%). The results indicated that *Staphylococcus aureus* and *Escherichia coli* were the major species contaminating the hospital floor. Contamination of hospital floor by pathogenic bacteria is a real danger to public health. The concept of environmental bacterial reservoir is a reality that requires strict compliance with current guidelines and recommendations for hand hygiene, cleaning, and disinfection of door handles in hospitals.

Keywords: Environment; Hospital; Hygiene; Microbes; Pathogens

1. Introduction

Nosocomial infections have become increasingly an emerging threat to the health care system over the past decades. They have highly been attributed to poor disinfection, decontamination, sterilization of hospital articles as well as weak antimicrobial stewardship policies (Kihla *et al*., 2014). The post-operative ward in most cases harbors patients who have undergone intrusive surgical procedures like C-sections, hip replacements and tumor among others. These procedures render them highly vulnerable to sepsis from nosocomial pathogens (Sserwadda *et al*., 2018).

Some of the consequences of this healthcare hurdle include prolonged hospital visits, increased antimicrobial resistance, and disabilities in the affected patients which greatly reduce the quality of life and productive human resource (Bazira *et al*., 2014). As such, it is very crucial to understand the roles played by the hospital equipment and the environment in the prolonged maintenance of nosocomial infections and transmission. Whereas the hospital environment may serve as reservoirs for these organisms, their transmission to patients is mostly through hand contact (Misgana and Abdissa, 2015).

Bacterial species have been estimated to cause the vast majority of these nosocomial infections followed by fungi and protozoa. Several previous studies have ventured into isolation of nosocomial infection-causing organisms but the most often encountered species include organisms like *Staphylococcus aureus*, *Escherichia coli, Pseudomonas aeruginosa, Klebsiella species* (Seni *et al*., 2013; Saito *et al*., 2014).

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Emergence of multidrug-resistant organisms has further escalated this problem especially in resource deprived nations as a result of overuse, misuse and inadequate antimicrobial stewardship policies in the health management systems. The lack of hospital antimicrobial teams and strict adherence to treatment guidelines has led to the wide usage of broad spectrum and first line antibiotics thus the exacerbation of the resistance. Implications of this resistance are prolonged hospital stays and the gross economic burden incurred in treatment with consequently high morbidity and mortality rates (Mulu *et al*., 2012). The aim of this study is to establish nosocomial bacterial contaminants often found on hospital floors of Ile-Abiye Hospital, Ado-Ekiti, Ekiti State.

2. Material and methods

2.1. Collection of samples

Forty (40) swab samples were taken from the floor of children, male and female wards at Ile-Abiye, Ado-Ekiti. The collected swab samples were transported and microbiologically processed using standard procedures.

2.2. Identification of isolates

The bacteria isolates were identified using cultural morphological and biochemical characteristic basis on their morphological characteristics, appearance and shape.

2.2.1. Gram staining technique

Smear of each bacterial isolate was prepared on a clean slide. In preparing the smear a drop of sterile distilled water was placed in the middle of the slide. A sterilized inoculating needle was used to pick from the bacterial colony and rubbed on the slide containing a drop of sterile distilled water. The bacterial cells were spread into a thin smear, air dried and heat fixed (Fawole and Oso, 2001). The heat fixed smear was stained with crystal violet for 1 minutes timing after which the slide was rinsed with water. Gram's iodine was added after 1 minute. The slide was flooded then washed with 95% alcohol until the violet colour was seen to stop running from the slide. The slide was rinsed with gentle running tap water and counterstained with safranin for 1 minutes. The slide was rinsed with water, blotted dry and examined under microscope with oil immersion. Gram positive cells appeared purple while gram negative cells appeared pink.

2.2.2. Catalase test

A thick emulsion of each test organism was prepared on a clean slide. Several drops of 3% hydrogen peroxide were added on each of the slides. A positive result was indicated by effervescence which was caused by the liberation of oxygen gas as a result of catalase production by the bacterium. There were no gas bubbles in the bacteria that do not produce catalase (Fawole and Oso, 2001).

2.2.3. Spore Staining

Heat-fixed smear of each isolate was prepared in a slide. Malachite green solution was added to the smear and steamed for 10 minutes. The stain was not allowed to dry out. The stain was then washed off with cold water. The smear was counterstained with safranin solution for 15 seconds. It was wash with water, blotted dry and examined under the microscope with the oil immersion objective (Olutiola *et al*., 2000). Spores appeared green and bacterial cells appeared red.

2.2.4. Indole test

One of the end products of the amino acid tryptophan hydrolysis is indole. Some microorganisms are capable of hydrolyzing tryptophan to give indole. Identification of Appropriate Sample and Culture Method for the Isolation of Thermophilic Bacteria from Automobile Radiators Production of indole revealed the possession of the enzyme tryptophalase by the test organism. 1% tryptone broth was prepared in different test tubes. The test tubes were inoculated with each bacterial isolate. The tubes were then incubated for 48 hours at 35 ^{o}C. After incubation, 2 ml of chloroform was added to each broth culture and was shook gently.2ml of Kovac's reagent was added to the broth culture and shook gently. The tubes were allowed to stand for 20 minutes in order to permit the reagent to rise to the top. A red colour at the reagent layer indicated indole production (Fawole and Oso, 2001).

2.2.5. Methyl red test

Peptone broth was prepared and distributed into several test tubes, the test tubes were sterilized in an autoclave at 121 ^{o}C, the test tubes were allow to cool and the organisms were inoculated into the test tubes, the tubes were incubated at

37 ^{o}C for 48hours, five drops of methyl red was added to the tubes and the tubes were shaken and examined after 5 minutes, coloured change was observed.

3. Results

The biochemical characteristic of isolates obtained from children, male and female ward for microbial contamination at Ile-Abiye Hospital, Ado-Ekiti floors were observed in tables 1, 2 and 3. The bacterial isolates from the wards were *Staphylococcus aureus*, *Escherichia coli*, *Staphylococcus epidermidis*, *Lactobacillus fermenti, Bacillus subtilis, Micrococcus luteus* and *Bacillus cereus*.

Table 4 shows the distribution of bacteria isolates from three wards (children, female and male ward) *Staphylococcus aureus* is the most prevalence isolate with 25(62.5%), followed by *Escherichia coli* 5(12.5%), *Staphylococcus epidermidis* and *Lactobacillus fermenti* 3(7.5%) respectively, *Bacillus subtilis* 2(5%) while *Micrococcus luteus* and *Bacillus cereus* 1(2.5%) respectively.

Table 1 Morphological and Biochemical characteristics of bacteria isolated from children ward

Location	Gram rxn	Shape	Cat	Coa	Sp st	Fermentation			Suspected organism
						Glu	Suc	Lac	
Floor 1	+	Cluster	+	+	–	+	+	+	*Staphylococcus aureus*
Floor 2	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*
Floor 3	+	Cluster	–	+	–	+	+	+	*Staphylococcus aureus*
Floor 4	–	Rod	+	+	–	–	–	+	*Escherichia coli*
Floor 5	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*
Floor 6	+	Cocci	+	+	+	+	+	+	*Staphylococcus aureus*
Floor 7	+	Cocci	+	–	–	+	+	+	*Staphylococcus epidermidis*
Floor 8	+	Diplococcic	+	–	+	+	–	+	*Staphylococcus aureus*
Floor 9	+	Diplococcic	+	+	–	+	–	+	*Staphylococcus aureus*
Floor 10	–	Rod	+	–	–	+	+	+	*Escherichia coli*
Floor 11	+	Cocci	+	+	+	+	+	+	*Staphylococcus aureus*
Floor 12	+	Cocci	+	–	–	+	+	–	*Staphylococcus epidermidis*
Floor 13	+	Cocci	–	+	+	–	+	+	*Staphylococcus aureus*
Floor 14	+	Cluster	+	+	+	+	+	–	*Staphylococcus aureus*
Floor 15	+	Cocci	+	+	–	+	+	+	*Staphylococcus aureus*
Floor 16	–	Rod	+	–	+	+	+	+	*Escherichia coli*

Cat= Catalase; Coa= Coagulase; Sp st= Spore staining; Glu= Glucose; Suc= Sucrose ; Lac= Lactose ; – = Negative; + = Positive

Table 2 Morphological and Biochemical characteristics of bacteria isolated from male ward

Location	Gram rxn	Shape	Cat	Coa	Sp st	Fermentation			Suspected organism
						Glu	Suc	Lac	
Floor 1	+	Cocci	+	–	–	+	+	–	*Micrococcus luteus*
Floor 2	+	Rod	+	+	+	+	+	+	*Bacillus subtilis*
Floor 3	–	Rod	–	+	+	+	–	–	*Bacillus cereus*
Floor 4	+	Cocci	–	+	+	+	+	+	*Staphylococcus aureus*
Floor 5	+	Rod	+	+	+	+	+	–	*Bacillus subtilis*
Floor 6	+	Cocci	+	+		+	+	+	*Staphylococcus aureus*
Floor 7	+	Cocci	+	+		+	+	–	*Staphylococcus aureus*
Floor 8	–	Rod	+	–	–	+	+	+	*Escherichia coli*
Floor 9	+	Cocci	+	–	–	+	+	+	*Staphylococcus epidermidis*
Floor 10	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*
Floor 11	+	Cocci	–	+	–	+	+	+	*Staphylococcus aureus*
Floor 12	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*

Cat= Catalase; Coa= Coagulase; Sp st= Spore staining; Glu= Glucose; Suc= Sucrose ; Lac= Lactose ; – = Negative; + = Positive

Table 3 Morphological and Biochemical characteristics of bacteria isolated from female ward

Location	Gram rxn	Shape	Cat	Coa	Sp st	Fermentation			Suspected organism
						Glu	Suc	Lac	
Floor 1	+	Cluster	–	+	+	+	+	+	*Staphylococcus aureus*
Floor 2	–	Rod	+	–	–	+	+	–	*Escherichia coli*
Floor 3	–	Cluster	+	+	–	–	+	–	*Staphylococcus aureus*
Floor 4	+	Rod	–	+	–	+	+	+	*Lactobacillus fermenti*
Floor 5	+	Cocci	–	+	+	+	+	+	*Staphylococcus aureus*
Floor 6	+	Cocci	+	+	–	+	+	+	*Staphylococcus aureus*
Floor 7	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*
Floor 8	+	Cluster	+	+	+	+	+	+	*Staphylococcus aureus*
Floor 9	+	Rod	–	+	–	+	+	+	*Lactobacillus fermenti*
Floor 10	+	Cocci	+	+	–	+	+	–	*Staphylococcus aureus*
Floor 11	+	Cocci	–	+	–	+	+	+	*Staphylococcus aureus*
Floor 12	+	Rod	–	+	–	+	+	+	*Lactobacillus fermenti*

Cat= Catalase; Coa= Coagulase; Sp st= Spore staining; Glu= Glucose; Suc= Sucrose ; Lac= Lactose; – = Negative; + = Positive

Table 4 Frequency of occurrence of the Bacterial Isolates

Isolate	Occurrence	Percentage of occurrence (%)
Staphylococcus aureus	25	62.5%
Staphylococcus epidermidis	3	7.5%
Escherichia coli	5	12.5%
Micrococcus luteus	1	2.5%
Bacillus subtilis	2	5%
Bacillus cereus	1	2.5%
Lactobacillus fermenti	3	7.5%
Total	**40**	**100%**

4. Discussion

From the study, the total samples were variously contaminated by bacterial agents many of which are recognized pathogens. *Staphylococcus aureus* 25(62.5%) were the most frequently isolated from all the samples collected from the wards followed by *E. coli* 5(12.5%). The result of this study agrees with the report of Muhammad *et al.* (2013) which showed that *Staphylococcus aureus* was the predominant among the isolated bacteria in hospital environments. It also correlates with the report of Genet *et al*. (2011) who reported that *Staphylococcus aureus* was the most prevalent bacteria isolated in the operating room. Either these microorganisms originated from the patients (endogenous flora) or from the staff, instruments and consumers (exogenous flora) this is difficult to be determined as endogenous flora can be transformed exogenous one (Okon *et al.*, 2012).

Staphylococcus aureus was the common microorganism from this finding. *Staphylococcus aureus* is a normal skin flora of the patients and the staff working in the wards. Droplet and nuclei contaminated with *Staphylococcus* can infect not only the wounds but also the floor, shelves, and lamps of the wards. Dust is an important factor in aerosolization of the microorganisms settled on the floor (Matthew *et al*., 2020). Unnecessary mobility of the staff during the operation can create air stream around the open wound; therefore, continuous maintenance of laminar air flow-ventilated wards offers high-quality air during surgery and the guidelines for environmental infection control in health-care facilities should be always implemented as reported by Matthew *et al*. (2020).

E. coli 5(12.5%), *Staphylococcus epidermidis* 3(7.5%), and *Lactobacillus fermenti* 3(7.5%) are bacterial pathogens frequent encountered within hospital environment as confirmed in this research. Intra hospital transmission of these bacterial pathogens can occur from transportation of patient either from the wards to the operating theatre and the specialized units. The air in the ward/or beddings and covering fabrics of the patient may have been contaminated already, in the course of the patient movement within the hospital, it is possible that the contaminated bacterial pathogens might be released either during the patient clothing/or bedding being changed without observing proper hygienic hospital procedures.

Although the role of formites in contributing to site of infections have been controversial, the fact that Onwubiko and Akande (2015) in the past confirmed the survival of unknown bacterial pathogens on inanimate objects for months and lends credence to the possibility of their causing nosocomial infections. The floor was contaminated predominantly with *S. aureus* and *E. coli*. Other contaminated bacteria isolates include; *Staphylococcus epidermidis*, *Lactobacillus fermenti*, *Bacillus subtilis*, *Bacillus cereus*, and *Micrococcus luteus*. This correlates with the study of Nwankwo and Azeez, (2017). This might be linked to the large influx of staff, students during operation and inadequate cleaning of the floor before and after operation.

5. Conclusion

The wards were contaminated with various types of bacteria. *Staphylococcus aureus* were the predominant bacterial type isolated from the entire wards, followed by *Escherichia coli*. Other bacteria isolates include; *Staphylococcus epidermidis*, *Lactobacillus fermenti*, *Bacillus subtilis*, *Bacillus cereus*, and *Micrococcus luteus*. Adherence to infection prevention practices may be paramount important.

Recommendations

The results indicate that some factors need to be considered. Some bacterial strains such as *S. aureus*, *S. epidermidis*, and *E. coli* have a greater propensity to cause contamination, especially in wards, so extensive infection control practices are necessary to prevent or contain these pathogens. The social level of incoming patients reflects the individual patient risk, which must be investigated and modified whenever possible. There is need for hospitals to encourage periodic review of the microbial flora of their environment and the antibiotic sensitivity pattern. It is also necessary that all professionals should take an active role in infection control within their organization and more resources should be provided to encourage good antibiotic practice and good hygiene in hospitals.

Compliance with ethical standards

Disclosure of conflict of interest

No conflict of interest to be disclosed.

Statement of ethical approval

The present research work does not contain any studies performed on animals/humans subjects by any of the authors'.

Statement of informed consent

Informed consent was obtained from the management of the hospital before collection of sample for the purpose of the study.

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