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Original Research Article

Evaluation of Some Selected Medical Equipment in Olabisi Onabanjo University Teaching Hospital for *Staphylococcus aureus* of Nosocomial Threat

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Abstract

Hospital is an establishment where patients that are diseased, infected, ill or injured receive medical care. A medical staff and equipment are required for the task of medical care delivery. The equipment that are employed in hospital could be classified as critical, semi-critical and non-critical depending on their design and tasks. A total of Eighty-five (85) samples of hospital equipment swab were obtained from Olabisi Onabanjo Teaching Hospital of which 40 samples of *Staphylococcus aureus* were isolated. The isolates were Gram stained followed by conventional biochemical test for the identification of *Staphylococcus aureus*. Antibiogram of the isolates was determined. The isolates exhibited resistance to ampiclox (87.5%), zinnacef (92.5%), amoxicillin (92.5%), rocephin (80%), and septrin (55%), while susceptible to pefloxacin (77.5%), gentamicin (80%), streptomycin (72.5%), and ciprofloxacin (52.5%). Some of the resistant isolates were exposed to plasmid DNA analysis and were found to be plasmid borne of varied molecular weight, which could be responsible for resistance to the antibiotics observed. There is therefore a need for regular disinfection, properly sterilization and preservation of medical equipment before and after use, which could curtailed or reduce the spread of equipment borne nosocomial infection.

Keywords: Medical equipment, *Staphylococcus aureus*, Olabisi Onabanjo Teaching Hospital. Molecular characterization.

INTRODUCTION

Hospital is an institution that are primarily designed for provision of medical treatment to the sick or injured people. Medical equipment and personnel are needed to carry out most of these functions. Hospital equipment is indispensable facilities fabricated purposely for a specific clinical function in health caring units. They are prone to colonization by microbes as a result of their configurations, functions and their placement. Arrays of bacteria that are non-nutritionally exacting can easily survive in form of biofilm colonies. Crevices in some of the equipment can harbor microbes. The domain where the facilities are kept could allow a beclouding of mist and other surviving environmental milieu on the facilities. (Schabrun and Chipase, 2006)

Medical device-associated infections are frequently caused by coagulase-negative staphylococci, especially *Staphylococcus epidermidis* and other resident microbes. The most important factor in the pathogenesis of medical device-associated staphylococcal infection, is the formation of adherent, multilayered bacterial colonization. Medical device can

be exposed to body fluids, such as blood, saliva, or urine and biological samples of clinical importance (Stamm, 2008). Hospital equipment are numerous and can be classified as critical, semi-critical and non critical depending on their functions and most of the equipment that can harbor bacteria of clinical potential includes; hospital bed, ward sinks, ward railings, wheel chairs, stethoscope, door handles, personnel examination table, defibrillators, anesthesia machine, patient monitor, sterilizers, ECG machines, surgical lights, chemical analyzer, X-ray machine, ultrasound, critical care equipment, catheters, ventilators, infusion Pump, syringe pump, suction apparatus, autoclave, blood pressure monitor, compression bandage, resuscitator, Oxygen mask, microscope, ophthalmoscope, thermometer, tweezers, weighing balance, crutches, inhaler, surgical hat, magnifier, Glucose meter, electrocardiography, scalpels, intravenous therapy, Laryngoscope, Nebulizer, sphygmomanometer-scan, hospital stretchers, thermometer and others. (Von Eiff *et al.,* 2005). Medical equipment plays an extensive role in various facets of healthcare services, encompassing prevention, screening, diagnosis, monitoring, therapy, and rehabilitation. Provision of healthcare services is nearly inconceivable without the use of these devices (Jamshidi *et al*., 2014).

For decades, *Staphylococcus aureus* has been predominately a nosocomial pathogen and is a leading cause of mortality and morbidity in hospitals. The important clinical *Staphylococcus aureus* infections are bacteremia, infective endocarditis, skin and soft tissue infections, osteoarticular infections and pleuropulmonary infections. Other clinical infections are epidural abscess, meningitis, toxic shock syndrome and urinary tract infection (Forbes *et al.*,1983). *Staphylococcus aureus* is equipped with a multitude of virulence factors that play a crucial role in its success as a pathogen responsible for a wide array of infections in both humans and animals. These virulence factors facilitate the bacterium's ability to adhere to host cells, overcome the host's immune defenses, invade tissues, trigger sepsis, and induce toxin-mediated syndromes. This forms the foundation for the persistence of staphylococcal infections even in the absence of a robust host immune response (Senn *et al*., 2016). Olabisi Onabanjo University Teaching Hospital, the study site is situated at Sagamu, Ogun State, South West Nigeria. The teaching hospital was established in the year 1986 with primary aim of teaching students from Olabisi Onabanjo University and provision of healthcare service to the indigenes of Ogun state and Nigeria as a whole. It operates a residency program to educate students, interns and qualified physicians, podiatrists, dentists, and pharmacists. The study aimed at evaluating some selected non-critical medical equipment in Olabisi Onabanjo University Teaching Hospital for *Staphylococcus aureus* capable of causing nosocomial infection.

MATERIALS AND METHODS

Collection of Samples

Eighty-five (85) saline soaked swab samples gently rubbed against the surfaces of each medical equipment were collected from various wards at Olabisi Onabanjo Teaching Hospital Sagamu, Ogun State Nigeria. The samples were then transferred aseptically to the pharmaceutical microbiology laboratory for microbiological analysis.

Bacteriological Analysis

The specimen laden swabs of equipment sampled were suspended in peptone water and incubated at 37ºC for 24 hours, thereafter sub-cultured on to Mannitol Salt Agar (OXOID^{R}) and incubated at optimum temperature for 24-48hours for the isolation of *Staphylococcus aureus.*

Ethical Consideration

Ethical approval with reference number NHREC/28/11/2017 was obtained from the Health Research Ethics Committee Office of the Hospital Management Board of the Olabisi Onabanjo University Teaching Hospital (O.O.U.T.H) before embarking on the study.

Antibiogram

This was determine by antibiotic disk-diffusion technique against the following antibiotics Pefloxacin (5µg), Gentamicin (10µg), Ampiclox (2µg), Cefuroxime (30µg), Ceftriaxone (30µg) Amoxicillin (10µg) Rocephin (30µg), Ciprofloxacin (5µg), Streptomycin (30µg), Cotrimoxazole (25 µg) Erythromycin (15µg). A volume of 0.1ml of the overnight broth culture of every isolate was pippeted into 9.9ml of the overnight broth culture of sterile distilled water in the test tube to make 10^{-2} dilution of the organism. The dilution were adjusted to 0.5 McFarland turbidity standard. From this dilution, 0.2 ml of the diluted culture was pipetted into the sterile melted and cooled (45^{0}C) Mueller Hinton Agar (Oxoid^{R}) and aseptically poured into into the sterile plate, and were allowed to set. The antibiotic multi-doses were aseptically placed in each plate and ere left for 30 minutes on the laboratory bench for pre-diffusion. The preparation were thereafter incubated at 37 for 24 hours before observation for the zones of growth inhibition which were interpreted as sensitive, intermediate or resistance as compared with CLSI (2020) standard).

Extraction of Plasmid DNA

Overnight culture were picked with an inoculating loop from Mueller Hinton agar plate into 300µL of (TENS) Tris 25Mm, EDTA 10Mm, NaOH 0.1N, and Sodium Dodecyl Sulphate 0.5%) in eppendorf tubes. The suspension was mixed by inverting the tubes 3 times until the mixture became sticky. The samples were placed on lab top ice bath cooler to prevent it from degradation of chromosomal DNA which may co-precipitate with plasmid DNA in the futher steps. 3.0M

sodium acetate pH 5.2, 150 µL was added to each sample and vortex mixed completely and centrifuged at 13.2 rpm for 5 minutes to pellet the cell debris and chromosomal DNA. The supernatant was transferred toa fresh tube, mixed with 900ul of ice-cold absolute ethanol and centrifuged for 10 minutes to pellet the plasmid DNA. (white pellet is observed)

The supernatant was discarded and the pellet was rinsed twice with 1mL of 70% ethanol and dried. The pellet was re-suspended in 40ul of TE buffer for the agarose gel electrophoresis.
(TENS composition: tris 25mM, EDTA 10Mm, NaOH 0.1N, and sodium dodecyl sulphate 0.5%)

Agarose Gel Electrophoresis

Agarose powder, 0.8g, was suspended in 100mL of Tris, Boric Acid, EDTA and brought to boiling on Bunsen burner to make 100mL of agarose gel which was allowed to cool to 45^0C before ethidium bromide was added to the preparation and gently mixed to avoid bubbles. It was then poured into electrophoresis tray with side comb inserted to form the wells, a process to the electrophoresis tank connected to 80v DC. Each sample, 10ul was mixed with a fount of bromo-phenol blue(tracking dye) and was introduced to different wells carefully. After this 0.5uL of Hind111digested DNA marker(promega) was also mixed with bromophenol blue and gently introduced to the first well, the tank was then plugged to the electrophoresis power pack which was allowed to run at 80V DC for $1^{1}/_{2}$hour.The gel was then viewed using short wave ultraviolet trans-illuminator in a dark room and photographs of the gel were taken using a cyber home digital camera and the molecular weights of their bands were calculated using Hind III digested DNA marker(promega) as standard.

RESULTS

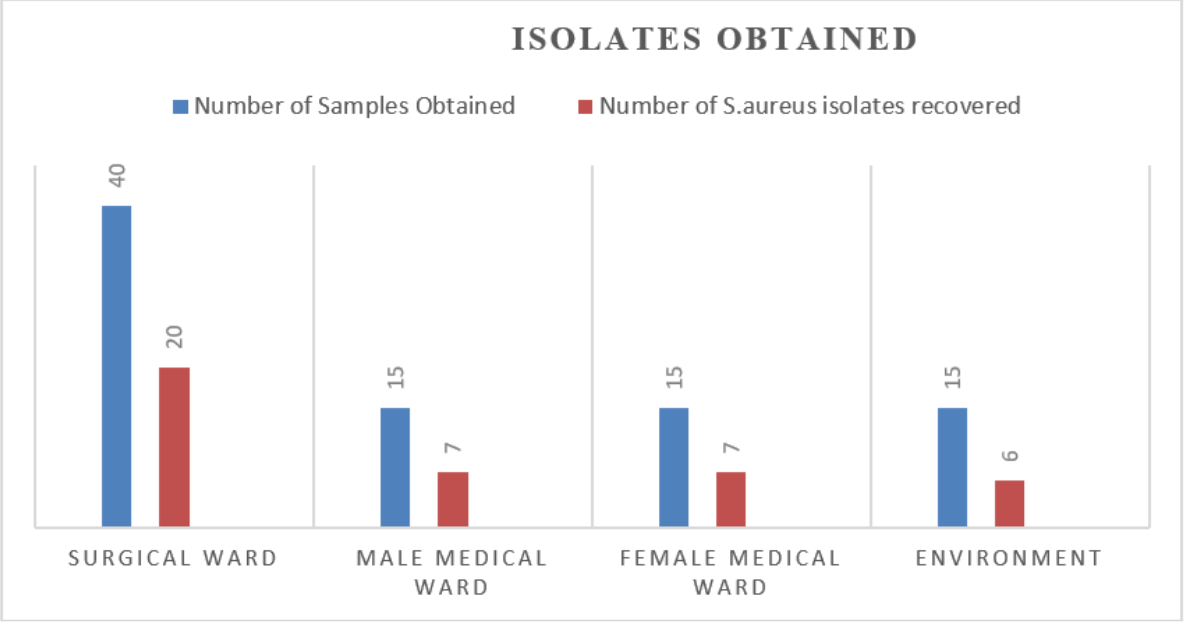


Figure 1: Samples collected across various wards within O.O.U.T.H

Table 1: Percentage and Frequency of Samples Distribution Versus Isolates Obtained in Relation to Equipment

Medical equipment	**Samples**				***% Staphylococcus aureus***			
	S . W	**M. M. W**	**F. M. W**	**E**	**S. W**	**M. M. W**	**F. M. W**	**E**
Personnel Examination Table	4	2	2	3	4(100%)	1(50%)	1(50%)	0
Hospital Chairs	5	2	2	2	3(60%)	1(50%)	1(50%)	2(100%)
Wheel chairs	5	1	1	0	2(40%)	1(100%)	1(100%)	0
Hospital beds	9	3	3	3	5(55.56%)	2(66.67%)	2(66.67%)	2(66.67%)
Ward sinks	4	2	2	0	2(50%)	1(50%)	0	0
Corridor railings	4	1	1	3	1(25%)	0	1(100%)	1(33.33%)
Door handles	4	2	2	4	2(50%)	1(50%)	0	1(25%)
Stethoscope	5	2	2	0	1(20%)	0	1(50%)	0
	40	15	15	15				

Keywords: S.W-Surgical ward M.M.W-Male medical ward F.M.W-Female medical Ward E-Environment

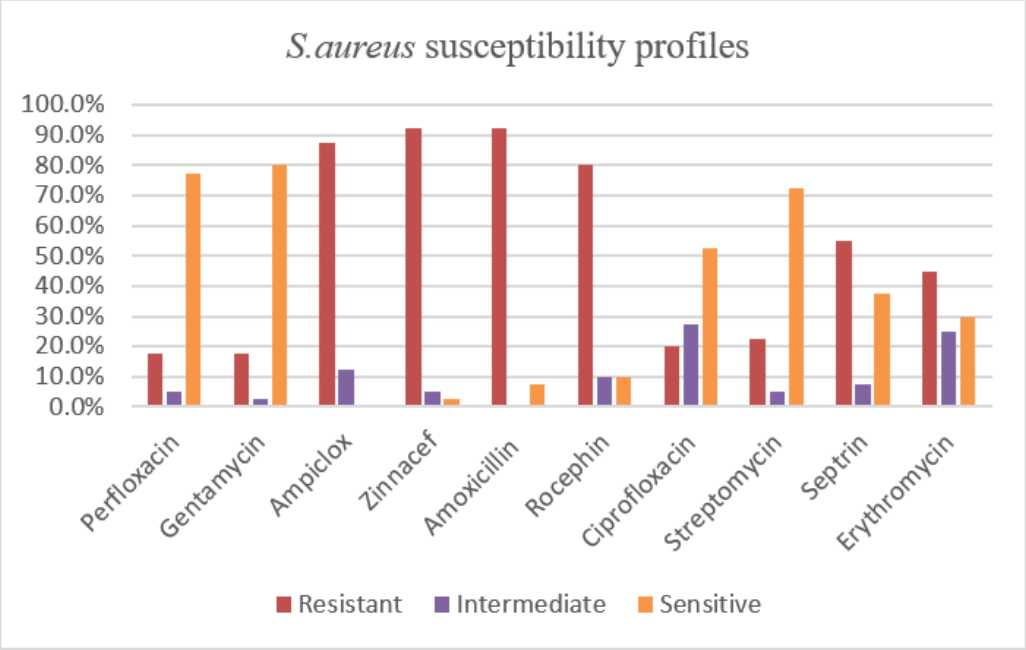
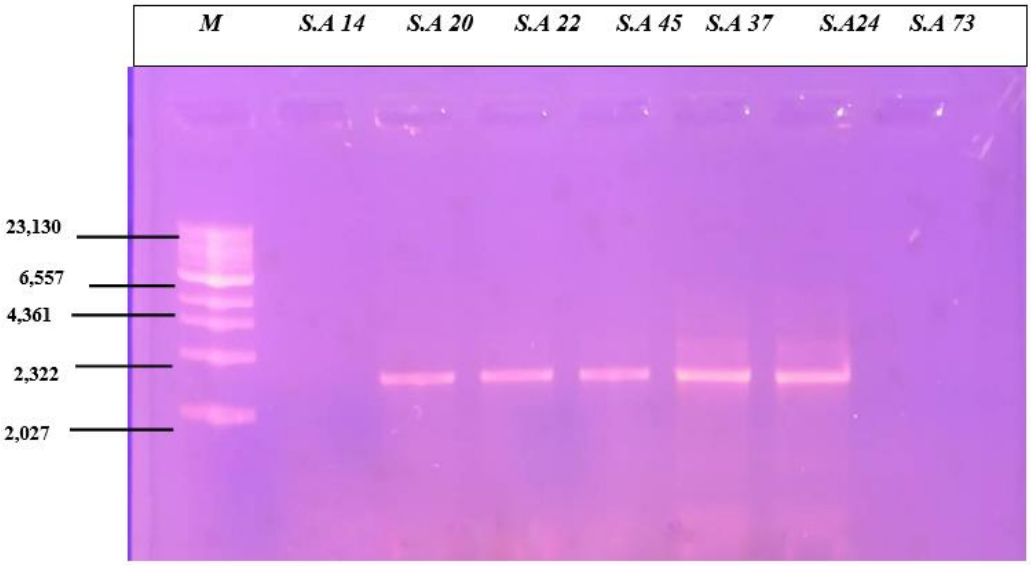


Figure 2: Graphical representation of the susceptibility of *S.aureus* to different antibiotics tested

Key: PEF-Pefloxacin(5µg) CN-Gentamicin(10µg) APX-Ampiclox(2µg) Z-Zinnacef(Cefuroxime)(30 µg) AM-Amoxicillin (10µg) R-Rocephin(Ceftriaxone sodium)(30µg) CPX-Ciprofloxacin(5µg) S-Streptomycin (30µg) SXT-Septrin ((25µg) E-Erythromycin(15µg)

The pattern of susceptibility *Staphylococcus aureus* to the antibiotics; Gentamicin (80%), Perfloxacin (77.5%),.and Streptomycin (72.5%) while resistant to Amoxicillin and Zinnacef were recorded to (92.5%) each, Ampliclox and Rocephin of (87.5%) and (80%) respectively as shown in Figure 2.



M= HIND III Digest of lambda DNA **S.A=** *Staphylococcus aureus*

Figure 3: Plasmid DNA profiles from selected resistant isolates

DISCUSSION

Hospital is an institution primarily designed to provide medical care to individuals who are ill or injured. The execution of most medical procedures in hospitals requires the presence of medical equipment and trained personnel. Hospital equipment consists of essential devices specifically designed for various clinical purposes within healthcare units. Due to their design, functions, and placement, these equipment items are susceptible to colonization by microorganisms.

In this current study, eighty-five (85) samples were obtained from obtained from medical equipment at Olabisi Onabanjo Teaching Hospital (O.O.U.T.H) of

which 40 samples of *Staphylococcus aureus* were isolated.

The ratio of distribution of the isolates from each ward visited varied from ward to ward, this could be attributed to prevailing environmental milieu around the wards but with the exception of male and female medical ward with coincidental equal number of distribution, the distribution pattern obtained in this study agree with the findings of Dadi et.al., (2021) on impact of healthcare associated infections connected to medical devices. *Staphylococcus aureus* was found to be prevalent in surgical ward due to the frequency of visitation than other wards accessed by the health care providers and patients.

The percentage distribution of isolates from equipment sampled as shows in Table 1 were found to be highest on every equipment sampled from the surgical wards and relatively equal from male medical ward and female medical ward while the relative distribution of the isolates were found to be the lowest from the environment sampled. The phenomenon recorded in percentage sample distribution in this study corroborates with the study of Mwanza and Mbohwa (2015) on assessment of effectiveness of equipment maintenance practices in public hospital.

The susceptibility pattern of *Staphylococcus aureus* isolates found in medical equipment in this study as shown in Figure 2, were recorded to be; pefloxacin (77.5%), gentamicin (80%), streptomycin (72.5%), ciprofloxacin (52.5%) while resistance to ampiclox (87.5%), zinnacef (92.5%), amoxicillin (92.5%), rocephin (80%) and septrin (55%) were observed. The episode of resistance to the antibiotics used seems alarming, which could be due to poor hospital disinfecting policy, misuse, or poor infection and disease prevention and control in health-care facilities, which agrees with the study of Odonkor and Ado, (2011) on bacterial resistance to antibiotics-recent trends and challenges.

Some of the isolates of *Staphylococcus aureus* that exhibited resistant to antibiotic were selected for plasmid DNA analysis, and were found to possess plasmid DNA of varied remarkable kilobases that ranged within 2,07kb and 23,13kb as elicited in Figure 3, which could confer antibiotic resistance to other isolates and aid the spread of resistant factors. This corroborates the study McCarthy and Lindsay (2012) on the distribution of plasmids and resistant gene in *Staphylococcus aureus.*

CONCLUSION

An indices from this study suggests that effort should be made by health care workers to ensure each medical equipment are regularly disinfected, properly sterilized and preserved as required before use to curtailed or reduce the spread of equipment borne infection transmission which could translate to treatment failures as a results of therapeutic failures.

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