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Surface Colonization of Health Care Facilities in Hematology by Methicillin Resistance *Staphylococcus aureus* (MRSA) at Obafemi Awolowo University Teaching Hospital Complex, Nigeria

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Abstract

Gram positive bacteria is not unique to intensive care units but has been increasingly reported in health care facilities, especially among Hematology patients worldwide. We evaluate microbiological disease etiology of *Staphylococcus aureus* in Hematology and frequency of harboring of pathogens by the facilities, medical equipment, surfaces and the possibility of cross contamination between health care workers including the antibiotic sensitivity pattern of the isolates recovered. Hundred sourced swabbed samples were taken from the facilities/equipment, rails, door knobs, personnel cell phone and other surfaces at hematology wards of the Obafemi Awolowo University Teaching Hospital Complex (OAUTHC) Ile-Ife. Swabs collected were inserted aseptically into test tubes that contain freshly prepared nutrient broth and then incubated at room temperature (37°C). After 24 h, the broth culture was inoculated into mannitol salt agar (MSA) plates using the inoculating loop. The streaking was done and the plates were incubated at 37°C for 24 h. Coagulase and DNase biochemical test later conducted to establish the *Staphylococcus aureus* isolates. Out of the 54 *Staphylococci sp.* obtained from hundred samples taken from different sources of various facilities and equipment in the wards, 25(46.3%) *Staphylococcus aureus* isolates showed total *Staphylococcus aureus* contaminant of facilities/environment/equipment where door knobs 7(28.0%) showed higher contamination rate followed by 5(20.0%) bed rails, which is higher than the % constitution of Personnel Stethoscope and cell phones which constituted 4(16.0%) and 3 (12.0%) respectively. All isolates were 98%, 64% and 28% susceptible to vancomycin, chloramphenicol and streptomycin respectively and 100% resistant to penicillin and oxacillin used. It is imperative for optimal patient care in the Hematology that effective aseptic techniques and use of sterile gloves, hand hygiene practices in handling surfaces with constant evaluation of antibiotic sensitivity pattern of pathogens for commonly used antimicrobial agents in a particular environment should be greatly maintained.

Keywords: Hematology ward; Methicillin resistance *staphylococcus aureus*; Antibiotic susceptibility; Cross infections

Introduction

Studies demonstrated a shift in the etiology of bacteraemic infections from predominance of Gram-negative rods to Gram-positive cocci [1]. Prior to the availability of Methicillin penicillin Resistant *Staphylococcus aureus* (MRSA) was the main threat to neutropenic patients and mortality rate from *Staphylococcus aureus* infections exceeded 50% [2].

Inanimate objects when contaminated with pathogenic microorganisms can transfer to a new host thereby serving as vehicles in transmission [3,4]. Fomites therefore associated particularly with hospital acquired infections (HAIs) that remain a major cause of patient morbidity and mortality [5,6]. An estimated 20% to 40% of HAIs have been attributed to cross infection via the hands of health care workers (HCWs), who have become contaminated from direct contact with the patient or indirectly by touching contaminated hospital environmental surfaces [7-9]. Fomites can therefore serve as reservoir with pathogens being spread from the inanimate environment to an animate (patient) environment via the hands of HCWs [10-12]. Stethoscopes, neckties [13-15], skin cells, hair, food, computer and ATM keyboards, pens, tables, artificial acrylic fingernails [16], bedding and clothing are common hospital sources of pathogens [4,17]. Up to 60% of hospital staff's uniforms are usually colonized with potentially pathogenic bacteria, including drug-resistant organisms [18]. Intravenous fluid (IVF) tubes/stands, catheters, water systems and life support equipment can also be carriers, when the pathogens form biofilms on the surfaces [4,19].

Transmission via hands of health care workers is likely the most common mechanism for spread of MRSA [15]. An understaffed has also been cited as a potential risk factor for hematology and Intensive Care Units (ICU). MRSA transmission, perhaps due to sacrifices in hand hygiene practices by overextended staff [20]. Additional factors associated with increased risk of nosocomial MRSA acquisition include duration

of antibiotic therapy, exposure to quinolone or macrolide antibiotics, length of hospital stay, enteral feeding, post-surgical status, and insertion of central line or urinary catheter during admission, ICU admission, and proximity to another patient with MRSA infection or colonization [21]. Ferreira et al. [22] on novel methicillin-resistant coagulase-negative *Staphylococcus* clone isolated from patients with hematological diseases at the Blood Bank Centre of Amazon, Brazil in their findings reported the potential of dissemination presented by multi-resistant *Staphylococcus* and they suggest the introduction of monitoring actions to reduce the spread of pathogenic clonal lineages of *Staphylococcus aureus* and *Staphylococcus epidermidis* to avoid hospital infections and mortality risks. The aim of this study was to evaluate microbiological disease etiology of *Staphylococcus aureus* in Hematology and frequency of pathogen harboring by facilities, medical equipment, ward surfaces and the possibility of cross contamination between health care workers including the antibiotic sensitivity pattern of the isolates recovered.

Materials and Methods

Collection of samples

A total of 100 swabs were collected from the Hematology wards of the Obafemi Awolowo University Teaching Hospital Complex. The samples taken were mainly from surfaces such as staff cell phones, Wards rails, Health personnel's stethoscopes diaphragm, hospital linens, bed rails, ward door knobs and Hospital desks. The samples were collected using wet sterile swab sticks by sweeping on the surfaces which were taken to the laboratory immediately after collection for analysis.

Methods of isolation

Swabs collected were inserted aseptically into test tubes that contain freshly prepared nutrient broth and then incubated at room temperature (37°C). After 24 h, the broth culture was inoculated into mannitol salt agar (MSA) plates using the inoculating loop. The streaking was done and the plates were incubated at 37°C for 24 h. Golden yellow color showed the fermentation of the mannitol which is a presumptive test for *Staphylococcus aureus*. Conventional methods of using catalase and coagulase tests for isolation of Gram positive coagulase positive *staphylococci* were adopted.

DNase test

A loopful of the 24 h agar culture was smeared on freshly prepared DNase agar plates and incubated at 37°C for 24 h. After 24 h, the plates were flooded with 1 N HCl and left for 5 minutes allowing for penetration before pouring away. A clear zone was observed around the colonies. This confirms *Staphylococcus aureus* isolated.

Antibiotics sensitivity test (Disk diffusion method)

The bacterial isolates were tested for their sensitivity to antibiotics by means of multiple disc diffusion method as recommended by CLSI, 2015 [23]. The commercial disc used contained the Gram positive discs. Fresh cultures of isolates

were harvested and suspended in a tube containing nutrient broth until turbidity corresponding to 0.5 McFarlan standard was obtained. Sensitivity was done on Mueller Hinton Agar. *Staphylococcus aureus* ATCC 25923 was used as a quality control for culture and antimicrobial susceptibility testing throughout the study. The antibiotics used were penicillin G (P) (10 µg), streptomycin (S) (15 µg), chloroamphenicol (C) (10 µg), oxacillin (Ox) (1 µg), gentamicin (CN) (30 µg), vancomycin (VA) (10 µg). The zones of inhibition were measured, recorded and interpreted according to the Clinical Laboratory Standard Institute provided (CLSI 2015).

Results and Discussion

It was reported in 1994 that patients with hematologic disorders, especially, leukemias and lymphomas, complicated bacterial infection frequently becomes serious. The site of infection is often unidentified, two special sites, the oral cavity and venous catheter a represented fomite, must not be overlooked. In this study, MRSA contamination of hematology facilities and in our own case the Ward surface environment at OAUTHC Ile- Ife, Nigeria was established (Table 1). Out of the 54 *Staphylococci sp* obtained from hundred samples taken from different sources of various facilities and equipment in the unit, 25(46.3%) *Staphylococcus aureus* isolates showed total MRSA undue colonization of facilities/environment/equipment where door knobs 7(28.0%) showed higher contamination rate followed by 5(20.0%) bed rails, which is higher than the % constitution of Personnel Stethoscope and cell phones which constituted 4(16.0%) and 3(12.0%) respectively.

In a retrospective study by Haqiwarra et al. [24] where 53 patients with haematological disorders whose bacterial cultures were positive for MRSA, were assessed to analyse the risk factors for MRSA infection, and the prognostic factors. In their findings, Sixteen patients showed colonization by MRSA but never developed infection(C), 16 showed colonization and subsequent infection (C-I), while 21 had MRSA infection at the time of first culture. It was inferred that poor performance status, thrombocytopenia, increased serum urea nitrogen and decreased serum cholinesterase which were more prominent than in (C) + (C-I). In this study, we did not directly assess the rate of colonization among patients but established the possibilities of cross infection from contaminated environment and these suggested inherent MRSA among patients admitted in the facilities.

However, the findings of this work similar to the report of Omololu-Aso et al. [25] conducted on selected wards in OAUTHC ranging from Male Medical and Female Medical Wards where occurrence of *Staphylococcus aureus* contamination of common equipment and others hospital fomites were confirmed. In their report, the percentage occurrence of *Staphylococcus aureus* varied from 17.85% to 25.0% in which door knobs had the highest *Staphylococcus aureus* contamination. Investigation conducted by Engelhart et al. [26] on *Pseudomonas aeruginosa* outbreak in a haematology-oncology unit associated with contaminated surface cleaning equipment at a tertiary-care centre, University of Bonn, Bonn, Germany, a total of 4.5% of samples from sanitary equipment and 20.0% of samples from

surface cleaning equipment were found to be contaminated with *Pseudomonas aeruginosa*.

Table 1: Distribution Frequency of *Staphylococcus aureus* isolated from the OAUTHC Hematology unit.

Facilities/ Equipment/ <i>Staphylococci</i> sp.	% occurrence of <i>Staphylococci</i> sp	<i>Staphylococcus aureus</i> isolated	% <i>Staphylococcus aureus</i> recovered
Ward Rails	18.5	4	16
10			
Door Knobs	18.5	7	28
10			
Bed Rails	18.5	5	20
10			
Wards Desks	12.9	2	8
7			
Staff/Patients Cell phones	18.5	3	12
10			
Stethoscopes	12.9	4	16
7			
Total: 54		Total: 25	

Fifty four percent (54.0%) *Staphylococci* sp in the hematological ward as reported in this study was not a surprise, it showed a strong indication of divers' polymicrobial contaminants that could be harbored by hospital fomites along with *Staphylococcus aureus* population leading to nosocomial infectious disease. This also supported the reports of Abraham and Jacob [27] who found that a significant percentage of cell phones were contaminated with multidrug resistant *Acinetobacter* sp and that cross contamination between hands, cell phone and patients constituted 10% of these pathogens. In our own report emphasis are strongly laid on *Staphylococcus aureus* invasion of the facilities. Noteworthy among haematological infections are MRSA infection caused by long-term use of third generation cepheems and neutropenic enterocolitis, for which the most recommended aid is surgical resection of the lesion. Skiest et al. [28] reported that Clinicians should be aware of the increasing prevalence of CA-MRSA. The

majority of patients hospitalized with community-associated *Staphylococcus aureus* infections were due to MRSA, recent antibiotics status predicted infection with MRSA and no clinical profile could reliably exclude MRSA. Some workers suggested that bacterial infection in haematology could be treated by empiric therapy and that G-CSF is effective for rapid granulocyte recovery and for prophylaxis, the laminar air flow room, oral non absorbable antibiotics, and systemic chemoprevention with colonization resistance are recommended. In this study, the isolates of *Staphylococcus aureus* recovered were tested on penicillin G (P) (10 µg), streptomycin (S) (15 µg), chloramphenicol (C) (10 µg), oxacillin (Ox) (1 µg), gentamicin (CN) (30 µg), vancomycin (VA) (10 µg). It was observed (Figure 1 and Table 2) that all isolates were 98%, 64% and 28% susceptible to vancomycin, chloramphenicol and streptomycin respectively and 100% resistant to penicillin and oxacillin used.

Table 2: Susceptibility profiles of *Staphylococcus aureus* isolated from the Haematology.

Antibiotics	Total No. of Isolates	Total No. S, R, I	Susceptible (%)	Resistance (%)	Intermediate (%)
Gentamycin	25	18S, 3R, 4I	72	12	16
Vancomycin	25	10S, 15R, 0I	98	2	0
Penicillin	25	0S, 25R, 0I	0	100	0
Oxacillin	25	0S, 25R, 0I	0	100	0
Chloramphenicol	25	16S, 5R, 4I	64	20	16
Streptomycin	25	7S, 16R, 2I	28	64	8

R: Resistance; I: Intermediate; S: Susceptible

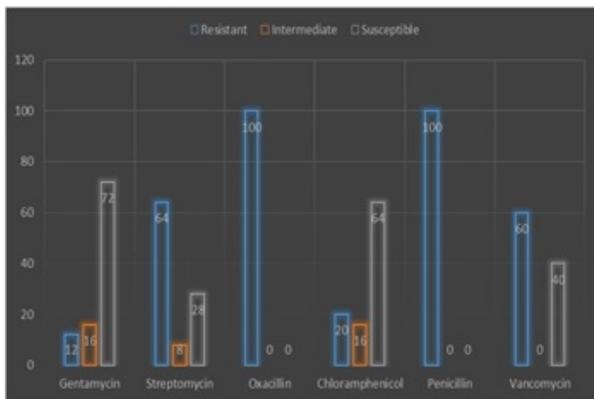


Figure 1: Antibiotic susceptibility trends of *Staphylococcus aureus* isolates recovered from hematology unit, OAUTHC Ilife, Nigeria.

The epidemiology of *Staphylococcus aureus* is dynamic and has changed significantly over the years. The proven ability of *Staphylococcus aureus* to acquire resistance gene is of a great concern among physicians worldwide. The search for novel therapeutic alternatives associated with policies to control antibiotic use and hospital-acquired infections guided by epidemiological surveillance studies should be constant habits among health professionals and hospitals as an alternative to minimize the problem.

These findings represent alarming increased rates in methicillin resistant *Staphylococcus aureus*. These results are comparable to findings in other studies [29,30]. It is imperative for optimal patient care in the Hematology that effective aseptic techniques, hand hygiene practices and use of sterile gloves in handling surfaces with constant evaluation of antibiotic sensitivity pattern of pathogens for commonly used antimicrobial agents in a particular environment should be greatly maintained.

Conclusion

Our investigation advised on the need to carefully evaluate cleaning and disinfection practices for patient care, particularly in neutropenic patients health care facilities. We suggested strongly that in order to decrease the likelihood of MRSA equipment contamination in Haematology, there must be a rise in the use of sterile pieces of equipment.

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