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The Prevalence and Antibiotic-resistant Profile of *Staphylococcus aureus* from Fomites in a Tertiary Institution in Ibadan, Oyo State, Nigeria

Emmanuel Lucky Orike [a*], Temidayo Emmanuel Olajugbagbe [b], Titilola Oyenike Animasahun [c] and Muwafiq Abdulraheem [a]

[a] Department of Microbiology and Biotechnology, First Technical University, Ibadan, Oyo State, Nigeria.
[b] Department of Microbiology, Baze University, Abuja, Nigeria.
[c] Department of Microbiology, Obafemi Awolowo University, Ile-Ife, Nigeria.

Authors' contributions

This work was carried out in collaboration among all authors. Author ELO conceived and performed the methodology. Author ELO performed the data curation and wrote the original manuscript. Author TEO performed the data analysis. Authors TOA and MA performed the reviewing and editing. All authors read and approved the final manuscript.

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**Corresponding author: E-mail: emmanuel.orike@tech-u.edu.ng;*

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ABSTRACT

The increasing prevalence of antibiotic-resistant *Staphylococcus aureus* (ARSA) poses a significant public health threat, especially in environments with high human contact such as tertiary institutions. This study was justified by the need to address the role of fomites as vectors for the transmission of ARSA, particularly in communal settings where contamination can lead to widespread infections. The investigation aimed to isolate and identify *Staphylococcus aureus* from fomites in a tertiary institution in Ibadan, Nigeria, and assess the antibiotic resistance profiles of these isolates. Fifteen samples were collected from various fomites within the institution and were cultured on Mannitol Salt Agar (MSA). Biochemical tests, including catalase, coagulase, and hemolysis assays, were used for the identification of Staphylococcus species. Antibiotic susceptibility test was conducted using the disk diffusion method to determine the resistance patterns of the isolates. Additionally, biofilm formation, which complicates infection control, was assessed using Congo red agar. The results revealed that 54% of the isolates were identified as *Staphylococcus aureus*, 33% as Staphylococcus epidermidis, and 13% as Staphylococcus hemolyticus. The isolates exhibited high resistance to commonly used antibiotics, including beta-lactams, with significant multi-drug resistance observed. Furthermore, all isolates demonstrated biofilm-forming abilities, which increase their virulence and resistance to environmental stressors. This study highlights the critical role of fomites in the transmission of antibiotic-resistant pathogens such as *Staphylococcus aureus* in communal settings. This finding underscores the urgent need for stringent hygiene practices, routine surveillance, and targeted interventions to control the spread of ARSA in environments with frequent human interaction.

Keywords: Antibiotic-resistance; Staphylococcus aureus; fomites; biofilm.

1. INTRODUCTION

Antibiotic resistance poses a significant threat to global health, and tertiary institutions play a critical role in addressing this issue (World Health Organization, 2019). These institutions must implement effective infection control measures, such as proper hand hygiene, sterilization protocols, and surveillance systems to track resistance patterns (Centers for Disease Control and Prevention, 2020). Moreover, tertiary institutions can foster a culture of responsible antibiotic use through education and awareness programs targeting students, faculty, and healthcare professionals (Spellberg et al., 2013). Tertiary institutions can also contribute to the fight against antibiotic resistance through research and development (Taconelli et al., 2019). Research centers and laboratories within these institutions can investigate novel antimicrobial compounds, alternative therapeutic strategies, and diagnostic tools (Coates et al., 2011). Collaborations between academia, industry, and healthcare sectors can facilitate the translation of research findings into clinical practice (Boucher et al., 2013). Furthermore, institutions can offer interdisciplinary courses and training programs focusing on antibiotic stewardship, pharmacology, and infectious disease management. To mitigate antibiotic resistance, tertiary institutions can adopt policies and guidelines promoting responsible antibiotic use (Dellit et al*.,* 2017). This includes implementing antimicrobial stewardship programs, restricting antibiotic prescriptions, and monitoring antibiotic consumption (McGregor et al., 2018). Additionally, institutions can establish antibiotic resistance surveillance systems to track and report resistance patterns (European Centre for Disease Prevention and Control, 2020). By taking proactive measures, tertiary institutions can protect students, staff, and surrounding communities from the dangers of antibiotic resistance.

Antibiotic resistance represents one of the most severe threats to global public health (Collignon and McEwen, 2019; WHO, 2020; Dada *et al.,* 2023). The misuse of antibiotics in healthcare and agriculture has accelerated the emergence of multidrug-resistant bacteria, complicating the treatment of infections. Among these pathogens, *Staphylococcus aureus* has drawn particular attention due to its adaptability and resistance mechanisms, including methicillin-resistant *Staphylococcus aureus* (MRSA). *S. aureus* is a common cause of both hospital-acquired and community-acquired infections, which range from mild skin infections to life-threatening conditions like pneumonia and septicemia (Akinrotoye et al*.,* 2019).

In environments such as tertiary institutions, fomites like door handles, tables, and chairs, beddings from institution clinic serve as vectors for the transmission of pathogens, including *S. aureus*. Shared facilities and close human contact in such settings can foster the spread of antibiotic-resistant bacteria, posing a significant risk to public health (Omololu-Aso et al*.,* 2022).

This study aimed to determine the prevalence of antibiotic-resistant *S. aureus* on fomites within a tertiary institution in Ibadan, Nigeria. By investigating the resistance patterns and biofilm formation capabilities of the isolated bacteria, the study will provide valuable insight into the potential public health risks posed by these pathogens.

2. MATERIALS AND METHODS

2.1 Study Location and Sample Collection

This study was conducted in Fist Technical University, Ibadan, Oyo State, Nigeria. Fifteen samples were collected from fomites (floors, door handles, tables, and chairs) in the institution's cafeterias and clinic. Sterile cotton swabs soaked in normal saline were used to swab the surfaces, and samples were immediately transported to the laboratory on ice (Musa et al*.,* 2023).

2.2 Isolation and Identification of Bacterial Isolates

The samples were cultured on Mannitol Salt Agar (MSA) and incubated at 37°C for 24 hours. Distinct colonies were sub-cultured on Nutrient Agar for purification. The isolates were identified through standard biochemical tests including Gram staining, catalase, oxidase, urease, citrate, coagulase, and hemolysis tests (Buszewski et al*.,* 2018: Wang et al*.,* 2019: Groeneveld et al*.,* 2017: Dogra et al*.,* 2018: Zhang et al*.,* 2020: Kim et al*.,* 2017: Christensen, W. B. 1946: Loeb, 1903). Sugar fermentation tests were also performed to identify bacterial species based on their metabolic profiles (Charles and Morgan, 2019).

2.3 Hemolysis Testing

The hemolytic activity of the isolates was determined by streaking on blood agar and incubated at 37°C for 24 hours. The plates were then examined for zones of hemolysis: beta-hemolysis (clear zones indicating complete lysis of red blood cells) and gamma-hemolysis (no lysis of red blood cells) (Oliveira et al*.,* 2018).

2.4 Antibiotic Sensitivity Testing

Antibiotic susceptibility testing was performed using the Kirby-Bauer disk diffusion method on Mueller-Hinton agar. The antibiotics tested included amoxicillin-clavulanate, cefotaxime, ceftriaxone, ciprofloxacin, gentamicin, erythromycin, and others. Results were interpreted based on Clinical Laboratory Standards Institute (CLSI, 2024) guidelines.

2.5 Biofilm Formation

Biofilm formation was assessed using Congo Red Agar (CRA). The medium was prepared by adding Congo red dye to nutrient agar containing sucrose. The plates were inoculated and incubated at 37°C for 24 hours. The formation of black colonies with a dry crystalline appearance was interpreted as positive for biofilm production (Peng et al*.,* 2022).

3. RESULTS

3.1 Prevalence of *Staphylococcus aureus* on Fomites

Fifty- four percent (54%) of the isolates were identified as *S. aureus*. Other identified species included *Staphylococcus epidermidis* (33%) and *Staphylococcus hemolyticus* (13%). The high prevalence of *S. aureus* on fomites highlights their potential role in pathogen transmission in communal environments. This shows the predominance of *S. aureus*, a pathogenic strain in fomites, underscoring the potential health risks associated with contact in shared spaces (Fig. 1).

3.2 Hemolysis Activity

The hemolytic activity of the isolates on blood agar shows that all *Staphylococcus aureus* and *S. hemolyticus* isolates exhibited beta-hemolysis (complete lysis of red blood cells), which is a strong indicator of virulence. In contrast, *S. epidermidis* exhibited gamma-hemolysis, meaning no red blood cell lysis. The presence of beta-hemolytic strains, particularly *S. aureus*, highlights their pathogenic potential, which could result in severe infections if transmitted through fomites (Table 1).

3.3 Antibiotic Resistance Profiles of *Staphylococcus aureus* Isolates

Staphylococcus aureus isolates exhibited extensive antibiotic resistance. The isolates were completely resistant to key antibiotics like cefotaxime (CTX), ceftriaxone (CRO), imipenem (IMP), and cefuroxime (CXM), which are frequently used in clinical settings. Resistance to these antibiotics poses a significant concern for treating *S. aureus* infections. Ciprofloxacin (CIP) and gentamicin (GN) showed some effectiveness, as a few isolates were either susceptible or exhibited intermediate resistance (Table 2).

3.4 Biofilm Formation

All isolates of *S. aureus*, *S.epidermidis*, and *S. haemolyticus* exhibited biofilm formation. Biofilms confer a survival advantage, protecting bacteria from environmental stressors and contributing to antibiotic resistance (Table 1).

Table 1. Hemolysis activity and biofilm formation of the isolates

Isolate Code	Suspected Organisms	Hemolysis	Biofilm
CFC	*Staphylococcus epidermidis*	γ	+
CFF	*Staphylococcus epidermidis*	γ	+
CFDH	*Staphylococcus epidermidis*	γ	+
CMF	*Staphylococcus aureus*	ß	+
CLF	*Staphylococcus aureus*	ß	+
CFT	*Staphylococcus epidermidis*	γ	+
CLDH	*Staphylococcus aureus*	ß	+
CMDH	*Staphylococcus aureus*	ß	+
CLT	*Staphylococcus aureus*	ß	+
CMT	*Staphylococcus hemolyticus*	ß	+
CLBH	*Staphylococcus aureus*	ß	+
CMC	*Staphylococcus aureus*	ß	+
CEFH	*Staphylococcus hemolyticus*	ß	+
CET	*Staphylococcus aureus*	ß	+
CEF	*Staphylococcus epidermidis*	γ	+

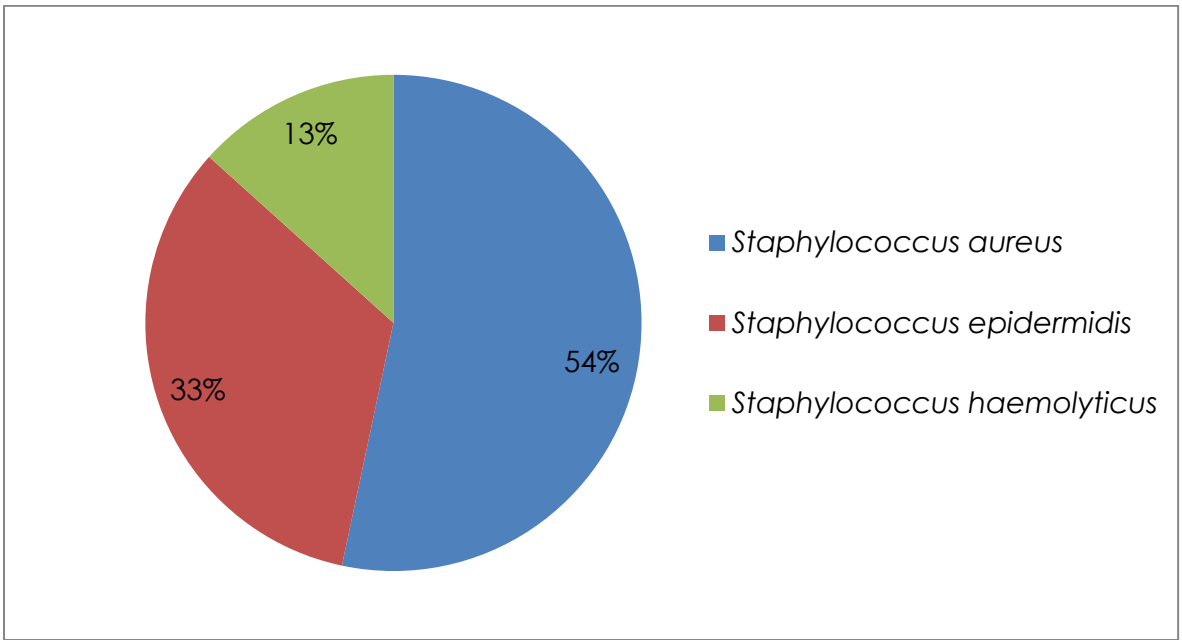

Fig. 1. Percentage of probable *Staphylococci* isolates from fomites

Table 2. Antibiotic susceptibility pattern of *S. aureus* isolates

Isolate/ Codes	CTX	CRO	IMP	CXM	OFX	ZEM	LBC	CIP	ERY	GN	AZN	AUG
Staphylococcus aureus (CMF)	R	R	R	R	S	R	S	S	R	S	I	R
Staphylococcus aureus (CLF)	R	R	R	R	R	R	S	R	I	I	S	R
Staphylococcus aureus (CLDH)	R	R	R	R	S	R	S	I	R	R	R	R
Staphylococcus aureus (CMDH)	R	R	R	R	R	R	R	R	I	S	I	R
Staphylococcus aureus (CLT)	R	R	R	R	R	R	R	R	I	R	R	R
Staphylococcus aureus (CLBH)	R	R	R	R	R	R	S	R	I	S	R	R
Staphylococcus aureus (CMC)	I	I	R	R	R	R	R	R	I	I	R	R
Staphylococcus aureus (CET)	I	S	R	R	R	S	S	R	I	S	R	I

Keys: R= Resistant, S= Susceptible, I= Intermediate, AUG= Amoxicilin Clavulanate, CTX= Cefotaxime, CRO= Ceftriaxone Sulbactam, IMP= Imipenem/Cilastatin, CXM= Cefuroxime, OFX= Ofloxacin, ZEM= Cefexime, LBC= Levofloxacin, CIP= Ciprofloxacin, ERY= Erythromycin, GN= Gentamycin, AZN= Azithromycin.

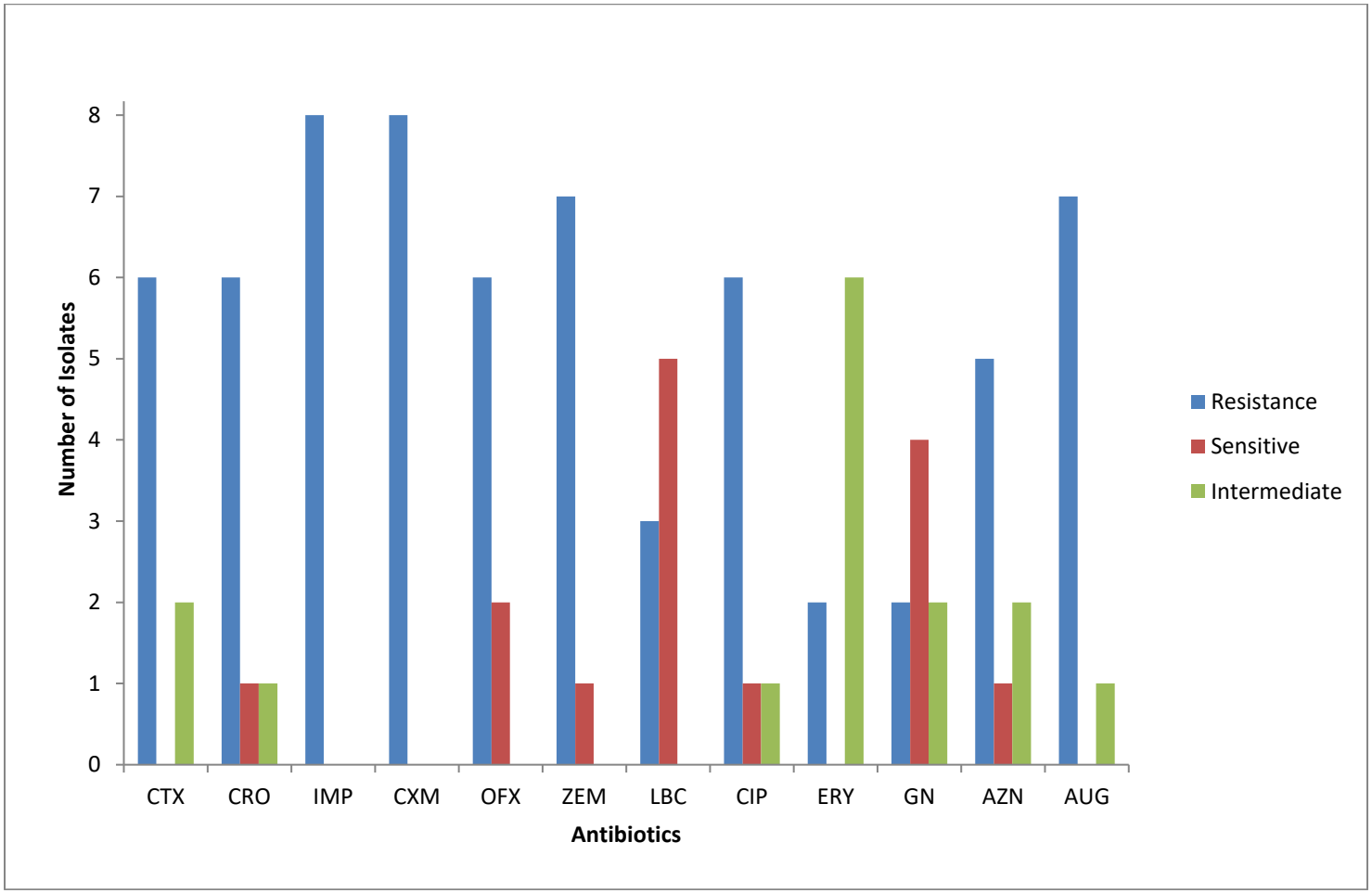

Fig. 2. Antibiotics susceptibility profile of *Staphylococcus aureus*

Keys: AUG= Amoxicilin Clavulanate, CTX= Cefotaxime, CRO= Ceftriaxone Sulbactam, IMP= Imipenem/Cilastatin, CXM= Cefuroxime, OFX= Ofloxacin, ZEM= Cefexime, LBC= Levofloxacin, CIP= Ciprofloxacin, ERY= Erythromycin, GN= Gentamycin, AZN= Azithromycin.

4. DISCUSSION

The results of this study underscore the significant presence of *S. aureus* on fomites within the institution, highlighting the risk of indirect transmission of antibiotic-resistant pathogens. The high rate of antibiotic resistance observed in this study, particularly to beta-lactams and macrolides, is consistent with global trends in antibiotic resistance (Collignon et al*.,* 2018).

The hemolysis test results indicate that the beta-hemolytic activity of *S. aureus* and *S. haemolyticus* contributes to their pathogenic potential, as they can lyse red blood cells and evade host immune responses (Otto, 2022). The gamma-hemolysis of *S. epidermidis* suggests that it is less virulent, although it still poses a risk of opportunistic infections, particularly in immunocompromised individuals.

Biofilm formation is a critical factor in the persistence of *S. aureus* on surfaces. Biofilms` protect bacteria from environmental stress, including antibiotic exposure, which may explain the observed multidrug resistance. These findings align with previous studies reporting that biofilm-forming *S. aureus* isolates are more resistant to antibiotics than their planktonic counterparts (Foster, 2021).

The resistance to cefotaxime, ceftriaxone, and other beta-lactams is of particular concern as these antibiotics are commonly used to treat *S. aureus* infections. The limited susceptibility to ciprofloxacin and gentamicin offers some hope, although the emergence of intermediate resistance calls for cautious use of these antibiotics (Akinrotoye et al*.,* 2019).

5. CONCLUSION

This study highlights the prevalence of antibiotic-resistant *S. aureus* on fomites in a tertiary institution, with all isolates forming biofilms. The widespread resistance to key antibiotics underscores the urgent need for enhanced infection control measures. Regular disinfection of communal areas, routine monitoring of antibiotic resistance patterns, and strict antibiotic stewardship are critical in reducing the spread of resistant *S. aureus* strains.

6. RECOMMENDATION

Implementation of stringent hygiene and disinfection protocols, particularly in high-contact areas such as cafeterias and clinics should be encouraged. Secondly, establishing routine surveillance programs to monitor fomite contamination and antibiotic resistance patterns is highly advised. Students and staff education on the importance of hand hygiene and the risks of fomite-mediated transmission of pathogens is very key to reduce the spread of antibiotic-resistant pathogen in tertiary institution.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Authors hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc.) and text-to-image generators have been used during the writing or editing of this manuscript.

COMPETING INTERESTS

Authors have declared that no competing interests exist

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