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# **Research Article**

Phenotypic Identification and Antimicrobial Susceptibility Profiles of Pathogenic Staphylococcus aureus from Nigerian Naira Notes Circulating in Bwari Metropolis, Abuja, Nigeria

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#### ABSTRACT

Staphylococcus aureus is a major pathogen responsible for a wide range of infections in both community and healthcare settings. This research was aimed to isolate and phenotypically characterize pathogenic Staphylococcus aureus from Nigerian naira notes and determine their antimicrobial susceptibility profiles. The isolation and characterization of the isolates were carried out following standard bacteriological techniques. Additionally, the antimicrobial susceptibility profiles of the isolates were assessed using the Kirby-Bauer disc diffusion method. Out of the 40 naira note samples collected, 15(37.5%) were positive for S. aureus. All the isolated S. aureus were pathogenic based on the hemolysis test. The antimicrobial susceptibility test showed varied susceptibility patterns of S. aureus isolates to the tested antimicrobial agents. The result of the antimicrobial susceptibility profiles revealed that ciprofloxacin exhibited the highest susceptibility profile of 93.3%; gentamicin recorded the highest intermediate resistance profile of 40.0%. Both Zinacef and Amoxicillin had the highest resistance profiles of 100%, respectively. This study has established that naira notes habour pathogenic S. aureus, which are a threat to public health. A high resistance observed to certain antimicrobial agents in this study emphasizes the need for appropriate antimicrobial stewardship and the consideration for effective alternative treatment options to reduce pressure on the antimicrobial agents, which has been identified as one of the major causes of resistance development. Nevertheless, based on the findings in this research, ciprofloxacin is suggested as a drug of choice for the treatment of Staphylococcus aureus infections within the study area.

Keywords: Bwari- Abuja; Naira notes; Pathogenic; Phenotypic identification; Staphylococcus aureus

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# INTRODUCTION

Staphylococcus genus is a diverse group of bacteria with around 30 species (Al-Zoubi *et al.*, 2015; Makolo *et al.*, 2022; Nwabueze *et al.*, 2024; Mishu *et al.*, 2025). Staphylococcus aureus is widely recognized as the most clinically significant species with a significant environmental presence (Makolo *et al.*, 2016; Suleima *et al.*, 2019; Makolo *et al.*, 2022; Mantovam *et al.*, 2025). Staphylococcus aureus is typically a commensal organism, it can also be pathogenic causing a variety of infections, ranging from mild infections of the skin and soft tissues to more severe and potentially fatal conditions like pneumonia and sepsis (Makolo *et al.*, 2022; Nakhaee and Hafez, 2025).

Depending on who handles the note or notes, Nigerian currency notes (naira notes) with denominations of N5, N10, N20, N50, N100, N200, N500, and N1000 can be used for a variety of legal tender purposes (Orababa *et al.*, 2021). They could be used to pay bills, settle debts, trade for goods and services, and occasionally be given as gifts (Orababa *et al.*, 2021). Studies have demonstrated that money can spread a number of harmful microorganisms, including Salmonella typhi, Escherichia coli, Staphylococcus aureus, and Klebsiella pneumoniae (Kawo et al., 2019; Orababa et al., 2021).

Staphylococci and spore-forming bacilli are the most predominant bacteria associated with currency notes and transmitted between people, either directly through direct touch or indirectly through food and water sources (Okwa and Bello, 2016; Kader and Al-Rawi, 2021). This may lead to potential outbreak of diseases caused by enteric pathogens which are normally spread through the fecal-oral routes (Alemu, 2014; Paul, 2024).

Previous studies have revealed some of the factors influencing the persistence and spread of microorganisms on currency notes. These factors include: population of microorganisms, their survival ability on surfaces, counting of currency notes with saliva, improper hand washing especially after using the toilet, sneezing and coughing on the hands leading to contamination of currency notes (Kramer et al., 2013; Nwabueze et al., 2024; Mishu et al., 2025). Other factors include, the guality of material used to produce the notes and the duration/storage condition with other environmental conditions may influence the load of microbial contamination (Michael, 2013). Previous reports showed that currency notes of lower denominations are more in circulation and habour more of the infectious agents (Musa et al., 2020; Olaleve et al., 2024).

Antimicrobial resistance and susceptibility are two perspectives that describe the overall responses of microorganisms to chemical or physical agents (Bustamante et al., 2025). An organism is said to be resistant when it shows capacity to survive and replicate in the presence of a given concentration of chemical substances under a defined set of conditions (Okoro, 2021; Baquero et al., 2021). Furthermore, the emergence and spread of antibiotic-resistant S. aureus strains have limited treatment options and pose a serious challenge in clinical practice (Chambers, 2021). One of the most well-known examples of antibiotic resistance in S. aureus is methicillin-resistant Staphylococcus aureus (MRSA), which is resistant to methicillin and other Beta-lactam antibiotics (Musa et al., 2020, Makolo et al, 2022). Apart from beta-lactam antibiotics, S. aureus has also become resistant to other antibiotic classes, such as tetracyclines, fluoroquinolones, macrolides, and aminoglycosides (Musa et al., 2020, Makolo et al, 2022; Sakalauskiene et al., 2025).

Therefore, this research was embarked on to assess the current prevalence and antimicrobial susceptibility profiles of pathogenic *Staphylococcus aureus* from the Nigerian Naira Notes in Bwari Metropolis, Abuja, Nigeria, and provide current and data-based literature that will facilitate the establishment of appropriate measures to avert the potential public health challenges inherent in the use and abuse of naira notes.

# MATERIALS AND METHODS

#### Study Area

This study was conducted in Bwari Metropolis, Abuja, Nigeria. Bwari is a Local Government Area in the Federal Capital Territory (FCT), Abuja. It is a suburban area with a population of over 300,000 people (Dauda, 2024). Bwari is known for its rich cultural heritage, natural beauty and diverse population (Emeafor *et al.*, 2018). The area covers a landmass of about 1,348 square kilometers and encompasses several communities, villages and districts (Madu, 2023). Bwari is characterized by its diverse topography, featuring rolling hills, rocky outcrops, and lush vegetation (Madu, 2023). The region is also blessed with several rivers, including Usuma Dam which contributes greatly to the agricultural productivity of the area (Opara, 2024). Sample Collection

Samples of eight (8) denominations of naira note (5, 10, 20, 50, 100, 200, 500 and 100 naira notes) were collected from April, 2024 – June, 2024 weekly. Five (5) samples were collected from each denomination, making a total of fourty (40) samples collected for this study. The collection of naira notes was made by exchange from volunteers who included traders, transport workers, civil servants, students and food vendors. The samples were collected aseptically with sterile gloves into sterile polythene bags, labeled and transported to the Microbiology Laboratory of Veritas University, Bwari Abuja within an hour of collection for bacteriological analysis. Purposive sampling technique was used.

# Culture Media Preparation

The media used in this study were Nutrient Agar, Mannitol Salt Agar, Blood Agar and Mueller-Hinton Agar. All the media used were aseptically prepared with strict adherence to the manufacturers' instructions. Sterility test was conducted on all the prepared media to ensure that they are free of microbial contaminations before samples inoculation.

# Bacteriological Analysis of the samples Serial Dilution

Each of the naira note sample was picked with the aid of sterile forceps and each was soaked in a beaker containing 10m aliquots of sterile buffered (0.1%w/v) peptone water for 20 minutes at ambient temperature with regular rigorous shaking to dislodge the cells into suspension. Thereafter, the currency notes were recovered and air-dried.

The resulting solution (stock) served as test sample. Ten-fold serial dilution was carried out as described by Musa *et al.* (2019). From the appropriate dilutions, 0.1ml aliquot were removed and spread plated on Nutrient Agar and Mannitol Salt Agar and incubated at  $37^{\circ}$ C for 24hours in the incubator.

#### Morphological characterization

Isolates that appear as yellow colonies on Mannitol Salt Agar (MSA) were presumptively considered as *Staphylococcus aureus* and the suspected colonies were subcultured on Nutrient Agar slants for further characterization. The isolates were Gram stained as described by Cheesbrough, 2019).

#### **Biochemical Characterization**

The biochemical screening was conducted on all the *Staphylococcus aureus* isolates. The tests conducted were Catalase, Coagulase, Indole, Oxidase, Citrate Utilization, Urease and Hemolysis tests following standard bacteriological procedures described by Chesbrough, (2003) and Makolo *et al.* (2022).

#### **Pathogenicity Test**

The pathogenicity status of the *Staphylococcus aureus* isolates were determined using hemolysis test a crucial tool used to detect the ability of bacteria to break down red blood cells, which is an indication of their pathogenicity.

#### Antimicrobial Susceptibility Test and selection of Multidrug Resistant Isolates

The antimicrobial susceptibility screening was conducted for all the *Staphylococcus aureus* 

isolates following the guidelines of Clinical and Laboratory Standard Institute (CLSI), using Kirby-Bauer disc diffusion method. The antibiotics used (Oxoid) were gentamicin (10ug), ampiclox (30ug), ciprofloxacin (10ug), pefloxacin (10ug), zinacef (20ug), amoxicillin (30ug), streptomycin (30ug), erythromycin (10ug), septrin (30ug) and rocephin (25ug). CLSI zone diameter interpretation standards were used to measure and interpret the diameter of the zone of inhibition generated by each antibiotic disc (CLSI, 2016). According to *Makolo et al.* (2019), the results were classified as susceptible, intermediately resistant, and resistant.

#### RESULTS

# Aerobic mesophilic bacterial count of currency notes samples

The result of the aerobic mesophilic bacterial count is depicted in Table 1. The counts were recorded as too numerous to count (TNC) as they were greater than 300cfu/ml.

Table 2 showed the prevalence of *Staphylococcus aureus* isolates from naira notes. A total number of 15(37.5%) isolates of *Staphylococcus aureus* were obtained from the naira notes analyzed in this study with denomination N 50 and N 100 having the highest carriage rate of 7.5% each and denominations N 500 and N 1000 had the lowest at 2.5% each.

#### Table 1: Aerobic Mesophilic Bacterial Count of Currency Notes Samples

Denominations	Number Screened	Mean Bacterial Counts (cfu/ml)
5	5	TNC
10	5	TNC
20	/ 5	TNC
50	5	TNC
100	/ 5	TNC
200	5	TNC
500 /	5	TNC
1000	5	TNC

Key: TNC: Too Numerous to Count

Table 2: Prevalence of <i>Staphylococcus aureus</i> isolated from naira note samples in Bwa	vari. Abuia
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Denominations	Number of samples Screened	No. of samples testing positive for <i>S. aureus</i>	Percentage of samples testing positive for
_			S. aureus
5	5	2	5.0
10	5	2	5.0
20	5	1	2.5
50	5	3	7.5
100	5	3	7.5
200	5	2	5.0
500	5	1	2.5
1000	5	1	2.5
Total	40	15	37.5

The results of the pathogenicity test for *Staphylococcus aureus* isolates from naira notes established that all were pathogenic. All the isolates exhibited Beta-hemolysis (complete hemolysis) as depicted in Table 3.

Table 4 showed the results of antimicrobial susceptibility patterns of *Staphylococcus aureus* isolates from naira notes to different antibiotics tested. The result revealed that ciprofloxacin recorded the highest susceptibility profile of 93.3%, gentamicin showed the highest intermediate resistance of 40.0%, amoxicillin and zinacef recorded the highest resistance profiles of 100% each. More so, all the isolates tested were distributed into four resistance patterns (A-D) according to their resistance to different antimicrobial groups (Table 5). Among the 15

isolates tested, all were considered multiple drug resistant (MDR) as they showed resistance to two or more classes of antibiotics tested. However, Staphylococcus aureus isolate coded NS3 had the highest MAR Index of 10 (resistant to the ten antibiotics tested), followed by NS5, NS10, NS11, NS12 and NS14 that had 9 of MAR Index (resistant to nine out of 10 antibiotics tested). *Staphylococcus* aureus isolates coded NS8, NS9, NS13 and NS15 had MAR Index of 8 each (resistant to eight out of ten antibiotics tested. Staphylococcus aureus isolates coded NS2 and NS7 had a lower MAR Index of 5 (resistant to five out of ten antibiotics tested) and Staphylococcus aureus isolates with codes NS1, NS4 and NS6 recorded the least MAR Index of 4 (resistant to four out of ten antibiotics tested) (Fig.1).

Table 3: Pathogenicity profiles of *Staphylococcus aureus* isolates from naira notes in Bwari using hemolysis test

Isolate Code	Hemolysis Test Result	Type of Hemolysis
NS1	+	Beta
NS2	+	Beta
NS3	+	Beta
NS4	+	Beta
NS5	+	Beta
NS6	+	Beta
NS7	+	Beta
NS8	+	Beta
NS9	+	Beta
NS10	+	Beta
NS11	+	Beta
NS12	+	Beta
NS13	+	Beta
NS14	+	Beta
NS15	+	Beta
Total	15 (100%)	15 (100%)

 Table 4: Antimicrobial susceptibility profiles of Staphylococcus aureus (n=15) isolates from naira notes in Bwari, Abuja

Antibiotics	Susceptibility Profiles	Intermediate Resistance Profiles	<b>Resistance Profiles</b>
Pefloxacin (10ug)	9 60.0%)	3 (20.0%)	3 (20.0%)
Gentamicin (10ug)	2 (13.3%)	6 (40.0%)	3 (20.0%)
Ampiclox (30ug)	-	2 (13.3%)	13 (86.6%)
Zinacef (20ug)	-	-	15 (100%)
Amoxicillin (30ug)	-	-	15 (100%)
Rocephin (25ug)	-	5 (33.3%)	10 (66.6%)
Ciprofloxacin (10ug)	14 (93.3%)	1 (6.6%)	-
Streptomycin (30ug)	5 (33.3%)	2 (13.3%)	8 (53.3%)
Septrin (30ug)	5 (33.3%)	-	10 (66.6%)
Erythromycin (10ug)	5 (33.3%)	2 (13.3%)	8 (53.3%)

Code	Resistance Pattern	No. of isolates with the pattern	Percentage of occurrence
А	APX, Z, AM, R	4	26.6
В	CN, APX, Z, AM, R	1	6.6
С	PEF, CN, APX, Z, AM, R, S, STX, E	9	60.0
D	PEF, CN, APX, Z, AM, R, CPX, S, SXT, E	1	6.6
Total		15	100%

Table 5: Antibiotic resistance patterns of *Staphylococcus aureus* (n=15) isolates from naira notes in Bwari, Abuja

**Key:** PEF-Pefloxacin; CN-Gentamicin; APX-Ampiclox; Z-Zinacef; AM-Amoxicillin; R-Rocephin; CPX-Ciprofloxacin; S-Streptomycin; SXT-Septrin; E-Erythromycin



Figure 1: Multiple Antibiotic Resistance (MAR) Index of *Staphylococcus aureus* isolates from naira notes in Bwari, Abuja

**Key:** N1-N15 = *Staphylococcus aureus* isolates

#### DISCUSSION

This research established a high occurrence rate for pathogenic *Staphylococcus aureus* from naira notes in Bwari, Abuja, Nigeria. This finding revealed that naira notes can serve as fomites for the transformation of staphylococcal infections. The finding in this study agrees with the report of Okoro, 2019 who reported a prevalence within the range established in this study in a similar study in Umuahia, Abia State, Nigeria. However, the report of Ni' *et al.*, 2020 obtained a higher prevalence in Kano State, Nigeria. The difference could be due to the hygienic levels of the study populations and sample sizes.

The result of antimicrobial susceptibility test in this study showed that ciprofloxacin exhibited the highest susceptibility profile, gentamicin had the highest intermediate resistance profile, and both zinacef and amoxicillin showed the highest resistance profile against the isolates tested. The high level of intermediate resistance and resistance profiles recorded in this study could be as a result of indiscriminate use and abuse of the antibiotics, mutation by the isolates development of resistance genes and circulation of substandard antimicrobial agents (Abebe and Birhanu, 2023). The increasing resistance rate of pathogenic *Staphylococcus aureus* against commonly prescribed antibiotics raises concerns as it might limit effectiveness of the antimicrobial agents, leading to difficult-to-treat bacterial infections (Michalik *et al.*, 2025). Surprisingly in this study, a *Staphylococcus aureus* isolate from naira note showed resistance to all antibiotics tested. This is a serious public health concern.

The presence of pathogenic *Staphylococcus aureus* on naira notes in circulation poses a potential risk to public health (Nwabueze *et al.*, 2024; Yohana, 2024; Olaleye *et al.*, 2024). Handling contaminated naira note currencies many contribute to the transmission of Staphylococcal infections, especially in areas with poor hand hygienic practices (Mishu *et al.*, 2025; Garvet *et al.*, 2025). Therefore, the findings in this study underscores the importance of maintaining proper hand hygiene, implementing antimicrobial stewardship

programs, and creating awareness about the potential health risks associated with contaminated currencies in circulation.

#### CONCLUSION

This research established a significant prevalence of pathogenic *Staphylococcus aureus* isolates from the naira notes investigated. Also, the findings in this study revealed that ciprofloxacin could be a drug of choice for the treatment of Staphylococcal infections within the study population. This study provides valuable insights into the potential health risks associated with the use of contaminated naira notes.

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**Authors' contributions:** Makolo, D. substantially contributed to the study conceptualization and design, data analysis and interpretation, manuscript writing and reading for important intellectual content. Yusuf, O.D., Yusuf, S.S and Ummulkhair, S.R. were actively involved in data acquisition and manuscript draft. All authors read and approved the final version of the manuscript.

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