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Original Research

Prevalence and antimicrobial susceptibility profile of *Staphylococcus aureus* isolated from marketed milk and cheese in Ilorin, Nigeria

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ABSTRACT

The irrational use of drugs, especially antimicrobials as growth promoters, and in the management of disease conditions without proper diagnosis has led to a higher incidence of multidrug resistance/antimicrobial resistance, which is a leading cause of death both in humans and animals. Improper diagnosis, irrational use of drugs and lack of adherence to withdrawal periods are factors incriminated in the incidence of multidrug resistance pathogens. This study was aimed at examining the prevalence, isolation, and antimicrobial susceptibility profile of *Staphylococcus (S.) aureus* in marketed milk and cheese in Ilorin, Kwara State, Nigeria. Using a cross-sectional study design which involved sampling of milk and cheese (n = 340) from four markets (Gambari, Ojaoba, Sango and Ipata) in Ilorin metropolis, *Staphylococcus aureus* (*S. aureus*) was isolated using standard microbiological procedures. Antimicrobial susceptibility testing of all positive isolates was done using the disk diffusion method. The prevalence of *S. aureus* was 3.5% in nono milk and 0.0% in cheese. The *S. aureus* isolated showed 100% resistance to ampicillin, ceftazidime and oxacillin and a high resistance rate to cefoxitin (91.7%), ceftriaxone (83.3%) and tetracycline (83.3%). Ciprofloxacin, gentamicin, and azithromycin showed lower resistance (25%) to the tested *S. aureus* isolates. The *S. aureus* isolates displayed six different resistance patterns and were resistant to three or more antibiotics. Particularly, three isolates were pan-resistant. This study emphasizes the importance of good management and hygiene practices throughout the milk processing value chain to ensure the quality and safety of the final product for consumers.

Keywords: Antimicrobial susceptibility testing; Cheese; Milk; Prevalence; *Staphylococcus aureus.*

INTRODUCTION

Staphylococcus aureus (*S. aureus*) is a member of a family of microscopic Gram-positive bacterial organisms (Park & Seo, 2019). It can cause serious infections such as skin abscesses, bacteremia, pneumonia, heart valve diseases, and bone infections in humans and animals (Tong *et al*., 2015). *Staphylococcus aureus* is an ubiquitous commensal bacterium commonly cultured from the skin, nasal and mucus membranes of healthy humans and animals (Lozano *et al*., 2016). *Staphylococcus aureus* can also stay longer on the contacted surface long after initial contact. The spread of this organism is mostly through contact with infected fluid or tissue such as milk by contaminated hands. It is a major foodborne pathogen (Kadariya *et al*., 2014; Odetokun *et al*., 2023) and there is a need to pool more information regarding the way food products, especially milk and cheese, are handled from farm to fork.

Foodborne diseases are of great public health significance (Kadariya *et al*., 2014; Odetokun *et al*., 2018; Odetokun *et al*., 2023). In many countries, strains of *S. aureus* remain a major cause of food poisoning (Cha *et al*., 2006). *Staphylococcus aureus* is a pathogenic bacterium commonly found in dairy products, including milk and cheese. It is known to produce various toxins that can cause foodborne illnesses in humans. Given the significant role that milk and cheese play in the daily diet of people in Nigeria, understanding the prevalence and antimicrobial susceptibility of *S. aureus* in these products is crucial for safeguarding public health. *Staphylococcus aureus* under favorable conditions can infect the host resulting in severe conditions (Lozano *et al*., 2016). In dairy cows, the infective nature of *S. aureus* (inhabiting epithelia, mucous membranes, and skin) and perseverance in the herd have been incriminated as a major obstacle in chronic mastitis of the mammary gland

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(Rainard *et al*., 2018). Foodborne intoxication caused by *S. aureus* is one of the most common bacteria-caused infections in many countries, especially in developing countries in which food of animal origins such as milk and milk products make a very good substrate for the growth of *S. aureus* (Okpo *et al*., 2017). Ensuring the safety of food products, especially those consumed daily like milk and cheese, is paramount. Contamination of these dairy products with *S. aureus* can lead to foodborne illnesses, ranging from mild gastrointestinal discomfort to severe infections (Badawy *et al*., 2022; Sayed-Ahmed *et al*., 2020).

The zoonotic *S. aureus* is of public health significance (Odetokun *et al*., 2018). The advent of resistant strains especially the methicillin-resistant *Staphylococcus aureus* (MRSA) has become an important subject of interest in recent years due to its deleterious effects on human and animal health (Lozano *et al*., 2016; Odetokun *et al*., 2022). *Staphylococcus aureus* is one of the leading bacterial causes of death due to antimicrobial resistance (AMR) (Murray *et al*., 2022). MRSA was published under the "WHO List of priority pathogens" as a Priority 2: High among other AMR pathogens (World Health Organization (WHO), 2017). The rise in antibiotic resistance is a global concern (Murray *et al*., 2022), and monitoring the antimicrobial susceptibility profile of pathogens such as *S. aureus* is essential. Understanding the resistance patterns of *S. aureus* isolated from milk and cheese in Ilorin, Nigeria, can contribute to the local and national efforts to combat antibiotic resistance.

Ensuring the safety and quality of food products, including dairy, is critical for maintaining consumer confidence in the food supply chain (Odetokun *et al*., 2021; Wu *et al*., 2021; Odetokun *et al*., 2022). When consumers have access to information about the prevalence of *S. aureus* and its susceptibility to antibiotics in milk and cheese, they can make more informed choices about their food consumption. This can have positive implications for both public health and the dairy industry in Ilorin. While studies on the contamination of milk and cheese by foodborne pathogens like *S. aureus* exist globally (Kou *et al*., 2021; Alembo & Torka, 2023) and elsewhere in Nigeria (Olufemi *et al*., 2018; Omoshaba *et al*., 2018; Aliyu *et al*., 2019; Oludairo *et al*., 2020; Esonu *et al*., 2021; Hassan *et al*., 2021; Aliyu *et al*., 2022; Ghali-Mohammed *et al*., 2022), localized data specific to Ilorin, Nigeria, are limited. This study will contribute valuable data that can be used for region-specific interventions and guidelines. Understanding the unique local factors that influence the prevalence and antimicrobial susceptibility of *S. aureus* can inform targeted control measures. The findings of this study can serve as a basis for the development or enhancement of food safety regulations and policies in Nigeria, especially in the context of dairy product production and distribution. Regulatory authorities can use this information to implement effective monitoring and control strategies to ensure the safety of these products. Therefore, this study was aimed at examining the prevalence, phenotypic isolation, and antimicrobial susceptibility profile of *Staphylococcus aureus* in marketed milk and cheese in Ilorin, Kwara State, Nigeria.

METHODS

STUDY DESIGN

An epidemiological survey using a Cross-sectional study design was applied in carrying out this study.

STUDY AREA

The survey was carried out in Ilorin, the capital of Kwara State. Ilorin is located in Northcentral Nigeria (Fig 1) on the coordinates: Latitudes 8^o 32'48"N to 8^o 34'30"N and Longitudes 4^o 32'15"E to 4^o 34'83"E. With a total area of about 13.6 km^2, it has an elevation of 319m above sea level (Bitrus, 2021).The current Ilorin's population is estimated at 1,030,498 (World Population Review (WPR), 2023). Ilorin is home to a conflux of tribes including Yoruba, Fulani, Hausa, and Nupe among other Nigerian tribes. The Fulani tribe of Nigeria is well recognized for cattle rearing as well as the sale of milk and milk products most commonly indigenous cheese (wara) in Ilorin. The chosen study area has notable markets for these milk and cheese products and is situated between the Northern and Southern states of Nigeria allowing for a high number of consumers purchasing these products frequently. It is therefore believed to have the highest number of retail points for milk and cheese in the state.

SAMPLE SIZE

The sample size was calculated using the formula for cross-section surveys: $n = [(Z_{1-\alpha/2})^2 * P_{exp} * (1- P_{exp})]/d^2$where "n" is the required sample size, which indicates the number of milk samples to be collected in the study area. "$Z_{1-\alpha/2}$" is the degree of confidence, typically set at 1.96."P_{exp}" is the expected prevalence and d is the desired absolute precision. In this study, an expected *Staphylococcus aureus* prevalence of 33.1% in milk (Esonu *et al*., 2021), and an absolute error margin of 5% were used in the calculation of the sample size. Therefore, n = 330. At least 330 samples were obtained across the markets in the study area.

SAMPLE COLLECTION

From December 2021 to February 2022, we randomly collected 290 milk and 50 cheese samples at an interval of one week from vendors at four different market places for this study. These include Oja-oba, Gambari, Ipata, and

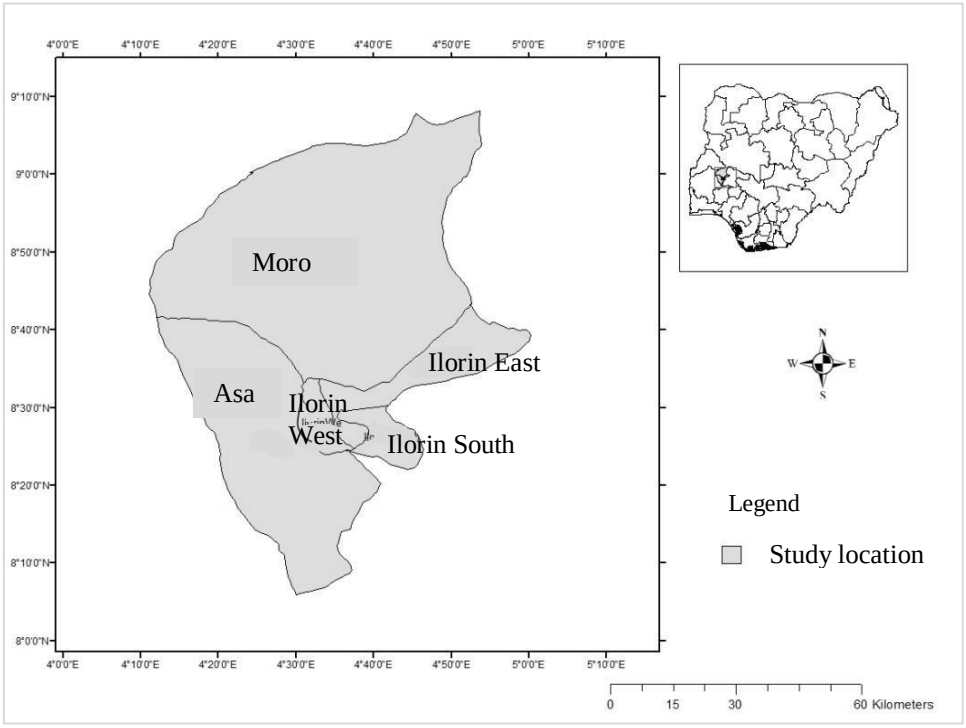

Figure 1: Map showing the Local Government Areas constituting the Ilorin Metropolis

Sango/Kulende markets. These markets were purposefully selected because they represent the places where milk and cheese are popularly sold in Ilorin. Cheese samples were collected separately in sterile foil paper and labelled, and milk samples were collected in sterile glass bijou bottles with tight caps. Both types of samples were then transported in transport coolers with ice packs to the Food Safety Laboratory, Department of Veterinary Public Health and Preventive Medicine, University of Ilorin for laboratory analysis. Upon arrival at the laboratory, the samples were immediately evaluated within 1-2 hours after collection.

ETHICAL APPROVAL

The Faculty of Veterinary Medicine Ethical Review Committee approved the study with the number UREC/FVM/15/32TA008.

PHENOTYPIC ISOLATION OF *S. AUREUS* FROM SAMPLES

Milk and cheese samples collected were processed according to the bacterial culture and identification procedures described by Odetokun *et al.* (2018) with some modifications. Briefly, 1ml of each milk sample was inoculated into 9 ml of Mueller Hinton broth (MHB) (Oxoid, UK) with 6.5% pure sodium chloride and incubated at 37ºC for 24 hours. A loopful of the inoculated broth of each sample was then cultured on Mannitol salt agar (MSA) (Oxoid, UK) for 24 hours and incubated at 37ºC. Subculture of characteristic colonies (tiny, creamy, non-mucoid, spherical, and yellowish) from the MSA to 5% sheep blood agar was carried out and incubated at 37ºC for 24 hours. Subsequently, tiny colonies showing clear zones due to beta hemolysis on the blood agar were further subcultured to nutrient agar (Oxoid, UK) and incubated at 37ºC for 24 hours after which positive colonies were stored in glycerol for further analysis.

The cheese samples collected were cultured for *S. aureus* as follows. Ten grammes of each cheese sample were broken down into small pieces and homogenized in the sterile bags containing 1ml of MHB. Then, the homogenized mixture was transferred into 9ml of MHB containing 6.5% pure sodium chloride and incubated at 37ºC for 24 hours. This was further processed using the same methods described for the milk samples.

CHARACTERIZATION OF *S. AUREUS* FROM CULTURE

The colonies stored in glycerol were subjected to further tests to confirm *S. aureus* using Gram's staining, catalase, and coagulase tests.

With Gram staining, positive *S. aureus* colonies presented as spherical (cocci) clusters appearing grapelike with a purple colour under the microscope (Ranzani *et al*., 2020). The slide coagulase test done identified positive *S. aureus* isolates as producing the enzyme coagulase. Clumping of the test organisms after rocking depicted a positive result for *S. aureus* (Sperber & Tatini, 1975). A catalase test was done to identify *S. aureus* which produces the enzyme catalase (Reiner, 2013).

ANTIMICROBIAL SUSCEPTIBILITY PROFILING OF *S. AUREUS*

Antibiogram of the positive isolates was determined using the disk diffusion method according to the Clinical and Laboratory Standards Institute (CLSI) recommendations. Positive colonies were picked and emulsified in normal saline and compared with 0.5 McFarland turbidity standard. The isolate emulsions were inoculated onto Mueller-Hinton agar (Oxoid, UK) and antibiotic discs were placed at

equidistance using the antibiotic disc dispenser (Oxoid, UK). The plates were incubated for 18 hours at 35°C. The diameter of the zone of inhibition was measured using a vernier caliper and interpreted according to standard guidelines (CLSI, 2022).

The isolates were tested on a panel of twelve antimicrobial disks with the following concentrations: Nalidixic acid (NA, 30µg), Ceftazidime (CAZ, 30µg), Gentamicin (CN, 30µg), Chloramphenicol (C, 30µg), Streptomycin (S, 10µg), Ciprofloxacin (CIP, 5µg), Ampicillin (AMP, 10µg), Tetracycline (TE, 30µg), Azithromycin (AZM, 15µg), Cefoxitin (FOX, 30µg), Oxacillin (OX, 1µg), Ceftriaxone (CRO, 30µg) to determine the sensitive, intermediate and resistant expression of the cultured isolates. The disks were chosen based on the fact that they are the commonly prescribed antibiotic used for the treatment of infections in human and animal hospitals, available over the counter and have been reported in previous studies on *S. aureus* (Omoshaba*et al*., 2018).

DATA MANAGEMENT AND STATISTICAL ANALYSIS

For this study, descriptive statistics was used. The prevalence of *S. aureus* isolated was reported in percentages (%) based on sample type and market, resistant patterns were also reported..

RESULTS

PREVALENCE OF *S. AUREUS* IN NONO MILK AND CHEESE

The result of the prevalence of *S. aureus* is presented in Table I. A prevalence of 3.5% was observed. A total of 12 *S. aureus* were isolated from nono milk obtained from the vendor at the Sango market in the study area while samples from the other markets were negative for *S. aureus.* None of the cheese samples were positive for *S. aureus.*

ANTIMICROBIAL SUSCEPTIBILITY PROFILE OF *S. AUREUS* ISOLATES FROM NONO MILK

Table II shows the antimicrobial susceptibility profile of the *S. aureus* isolates to different concentrations of antibiotics. The tested isolates displayed varying resistance levels to the antimicrobials. *S. aureus* isolates showed the highest percentage resistance (100.0%) to ampicillin, ceftazidime, and oxacillin. These isolates (91.7%) expressed resistance to cefoxitin which is a third-generation cephalosporin antimicrobial, while 83.3% of the isolates were resistance to ceftriaxone and tetracycline. Nalidixic acid had an average (50%) resistance level while chloramphenicol and streptomycin resulted in 33.3% resistance. The least resistance level (25.0%) was observed with ciprofloxacin, gentamicin, and azithromycin. However, three-quarters of the isolates (75.0%) were susceptible to gentamicin.

Table I: Antimicrobial susceptibility profile of suspected *S. aureus* isolated from Nono milk and cheese

	n	Number of positive samples	Number of negative samples	Prevalence (%)
Sample type				
Nono milk	290	12	278	3.5
Cheese	50	0	50	0.0
Total	340	12	328	3.5
Market				
Gambari	99	0	99	0.0
Ipata	5	0	5	0.0
Oja Oba	26	0	26	0.0
Sango	210	12	198	3.53
Total	340	12	328	3.53

RESISTANCE PATTERNS EXPRESSED BY *S. AUREUS* ISOLATES

Table III presents the resistance patterns expressed by the isolated *S. aureus* to the tested antibiotics. The *S. aureus* isolates displayed six different resistance patterns. These isolates were resistant to three or more antibiotics with three isolates being particularly resistant to all the 12 antibiotics tested. Four isolates displayed the most similar resistance pattern in the form of AMP, CAZ, CRO, FOX, OX, TE. Three isolates showed a resistance pattern of AMP, AZM, C, CN, CAZ, CIP, CRO, FOX, NA, OX, S, TE. All isolates were resistant to ampicillin, ceftazidime, and oxacillin.

DISCUSSION

In this study, we found an overall prevalence of 3.5% for *S. aureus* in milk samples. Notably, all of these isolates exhibited complete resistance to ampicillin, ceftazidime, and oxacillin, with six distinct resistance patterns identified. However, 75.0% of the isolates were susceptible to gentamicin. Over the years, milk and milk products have been identified as potential sources of bacterial pathogens that can spread to humans (Aliyu *et al*., 2019). Despite the relatively low prevalence of 3.5% observed in our study, it is important to underscore the significance of *S. aureus* as a foodborne and clinically relevant microorganism. It can potentially be transmitted from animals to humans through the consumption of contaminated food items like milk and cheese. The presence of *S. aureus* in the milk samples obtained from the Sango market is attributed to suboptimal milking, handling, and processing practices during the production of milk and cheese. These factors may

Table II: Antimicrobial susceptibility profile of *S. aureus* isolates from Nono milk

Antibiotics	Symbol	Potency/Conc. of antibiotics	Resistance (%)	Intermediate (%)	Susceptible (%)
Nalidixic acid	NA	30µg	50.0	50.0	0.0
Ceftazidime	CAZ	30µg	100.0	0.0	0.0
Gentamicin	CN	30µg	25.0	0.0	75.0
Chloramphenicol	C30	30µg	33.3	8.3	58.3
Streptomycin	S	10µg	33.3	8.3	58.3
Ciprofloxacin	CIP	5µg	25.0	16.7	58.3
Ampicillin	AMP	10µg	100.0	0.0	0.0
Tetracycline	TE	30 µg	83.3	16.7	0.0
Azithromycin	AZM	15µg	25.0	33.3	41.7
Cefoxitin	FOX	30µg	91.7	0.0	8.3
Oxacillin	OX	1µg	100.0	0.0	0.0
Ceftriaxone	CRO	30µg	83.3	8.3	8.3

Nalidixic acid (NA, 30µg), Ceftazidime (CAZ, 30µg), Gentamicin (CN, 30µg), Chloramphenicol (C, 30µg), Streptomycin (S, 10µg), Ciprofloxacin (CIP, 5µg), Ampicillin (AMP, 10µg), Tetracycline (TE, 30µg), Azithromycin (AZM, 15µg), Cefoxitin (FOX, 30µg), Oxacillin (OX, 1µg), Ceftriaxone (CRO, 30µg).

compromise the viability of the pathogen at the time of collection (Ghali-Mohammed *et al*., 2022; Ghali-Mohammed *et al*., 2023).

The safety of milk for human consumption is of vital concern worldwide. In Nigeria, as in many other countries, ensuring the quality and safety of milk and milk products is a crucial aspect of public health. Several studies have investigated the prevalence of *S. aureus* in milk sold for human consumption across various regions of Nigeria. In a previous study, 5% prevalence for *S. aureus* was reported in fresh milk. Notably, *S. aureus* occurrence was higher in nono (12.1%) and kindirmo (10.6%) compared to fresh raw milk (5.9%). Furthermore, the source of water (well water) used for cleaning utensils at the nono and kindirmo selling points was identified as a risk factor associated with the presence of *S. aureus* in these products (Aliyu *et al*., 2019) which is also comparable to other low prevalence found in similar studies carried out in parts of Kaduna (Okpo *et al*., 2017). Higher prevalence of 14.0% (Olufemi *et al*., 2018), 26% (Omoshaba *et al*., 2018), 31.5% (Oludairo *et al*., 2020), 38.1% (Esonu *et al*., 2021), and 47.1% (Hassan *et al*., 2021), of *S. aureus* were obtained in milk sold for human consumption in studies carried out in other parts of Nigeria. The reported prevalence in these studies has shown significant variations, raising important questions about the safety and quality of milk in the country. The variability in *S. aureus* prevalence in milk meant for consumption could be due to geographic variation, dissimilar sample size and sample collection, different levels of milk hygiene and handling practices, varying laboratory isolation methods, and the health of animals producing the milk. Varying climate conditions, agricultural practices, and livestock management systems can influence the detection of *S. aureus* in milk produced for public consumption (Odetokun *et al*., 2023).

Table III: Antimicrobial susceptibility profile of *S. aureus* isolates from Nono milk

S/N	n	Resistance pattern
1	1	AMP, CAZ, OX
2	1	AMP, CAZ, FOX, OX
3	4	AMP, CAZ, CRO, FOX, OX, TE
4	2	AMP, CAZ, CRO, FOX, NA, OX, TE
5	1	AMP, C, CIP, CAZ, CRO, FOX, NA, OX, S, TE
6	3	AMP, AZM, C, CN, CAZ, CIP, CRO, FOX, NA, OX, S, TE

Nalidixic acid (NA, 30µg), Ceftazidime (CAZ, 30µg), Gentamicin (CN, 30µg), Chloramphenicol (C, 30µg), Streptomycin (S, 10µg), Ciprofloxacin (CIP, 5µg), Ampicillin (AMP, 10µg), Tetracycline (TE, 30µg), Azithromycin (AZM, 15µg), Cefoxitin (FOX, 30µg), Oxacillin (OX, 1µg), Ceftriaxone (CRO, 30µg)

The antimicrobial susceptibility profile of the *S. aureus* isolates is a critical indicator of the effectiveness of various antibiotics against this pathogenic bacterium. This data highlights the concerning issue of antibiotic resistance, a global health challenge. The most striking finding in this study is the high level of resistance displayed by *S. aureus* isolates to several commonly used antibiotics. Ampicillin, ceftazidime, and oxacillin showed a 100% resistance rate. Compared to other studies, high resistance rates were also recorded for ampicillin (Alembo & Torka, 2023), ceftazidime (Omoshaba *et al*., 2018), and oxacillin (Omoshaba *et al*., 2018; Sayed-Ahmed *et al*., 2020; Esonu *et al*., 2021). These antibiotics are crucial for treating a wide range of bacterial infections, and their ineffectiveness against *S. aureus* is alarming. This high resistance might be attributed to the overuse or misuse of these antibiotics in clinical settings or agriculture, contributing to the development of resistant strains. The organisms (91.7%)

exhibited resistance to cefoxitin, a third-generation cephalosporin, indicating that even newer and more potent antibiotics are facing challenges when it comes to *S. aureus* infections. Comparably, high levels of resistance to cefoxitin were also noticed in *S. aureus* isolated from some dairy products in previous studies (Aliyu *et al*., 2019; Alembo & Torka, 2023).Tetracycline also showed a significant level of resistance at 83.3% similar to other studies (Okpo *et al*., 2017; Omoshaba *et al*., 2018; Esonu *et al*., 2021), emphasizing the need for alternative treatment options. Ceftriaxone also showed a high level of resistance (83.3%) to the isolated *S. aureus*. However, other studies have found lower or no levels of resistance to this antibiotic (Daka *et al*., 2012; Tibebu *et al*., 2021), a reason why it is often used to treat *S. aureus* infections (Alsowaida *et al*., 2022).

Interestingly, some antibiotics such as ciprofloxacin, gentamicin, and azithromycin demonstrated lower resistance levels, with only 25.0% of *S. aureus* being resistant. This suggests that these antibiotics could still be effective in treating *S. aureus* infections in the studied population. However, it is crucial to note that antibiotic susceptibility can vary regionally and over time due to different factors, including local antibiotic usage patterns and genetic mutations in bacterial populations. The study's finding that three-quarters of the isolates (75.0%) were susceptible to gentamicin is a glimmer of hope. Previous studies have detected low resistance levels to gentamicin (Elias *et al*., 2020; Gentilini *et al*., 2000; Li *et al*., 2015; Okpo *et al*., 2017; Tibebu *et al*., 2021; Anueyiagu *et al*., 2022). Gentamicin is a valuable antibiotic for treating severe infections, and its continued effectiveness is encouraging.

The resistance patterns observed in the isolated *S. aureus* strains provide valuable insights into the complexity of antibiotic resistance in this pathogen. This data underscores the pressing issue of multidrug-resistant *S. aureus* strains, which pose significant challenges for clinical management and public health. One of the most concerning findings is the presence of six distinct resistance patterns among the *S. aureus* isolates. These patterns reflect the diversity of resistance mechanisms at play in the studied population. In particular, the existence of isolates resistant to all 12 tested antibiotics is alarming. Such extensively drug-resistant (XDR) strains are especially difficult to treat and are associated with poor clinical outcomes. Other studies have also identified multidrug-resistant *S. aureus* strains from milk and other dairy products and this portends a serious risk to animal and human health (Oloso *et al*., 2018; Oludairo *et al*., 2020; Sayed-Ahmed *et al*., 2020; Tibebu *et al*., 2021; Badawy *et al*., 2022; Murray *et al*., 2022; Alembo & Torka, 2023; Gaddafi *et al*., 2023). The multidrug resistance patterns expressed by the *S. aureus* isolates suggest a broader resistance profile, encompassing multiple antibiotic classes, which further complicates treatment options. The universal resistance to ampicillin, ceftazidime, and oxacillin across all isolates is a significant cause for concern. These antibiotics are commonly used in clinical practice, and the widespread resistance to them limits treatment options for *S. aureus* infections.

Our results not only underscore the urgency of addressing antibiotic resistance but also highlight the need for prudent antibiotic use, robust surveillance, and the development of alternative treatment strategies. Strategies such as antibiotic stewardship programs, increased public awareness of antibiotic resistance, and research into new antibiotics or treatment modalities are essential to combat the growing threat of AMR.

CONCLUSION

This study showed that there is a risk of spread of multidrug-resistant *S. aureus* among consumers of nono milk marketed in Ilorin, suggested to be a result of vendors' hygiene as at the time of milking cattle, indiscriminate use of antibiotics without the supervision of veterinarians, lack of proper screening procedures for cows to be milked for sale, and environmental condition at the point of sale in the market. This study has shown that *S. aureus* isolated was resistance to the majority of antimicrobial agents used especially third-generation antimicrobials which are to be resorted to in the management of conditions when common or first-generation agents lack effectiveness, hence, there is a need for regulation and monitoring of the use of drugs in the animal sector and enforcement of strict adherence to withdrawal period after usage. Further study on the molecular characterization of the types and subtypes of *S. aureus* isolated from the milk, as well as possible toxins that may be produced by the isolates is required.

REFERENCES

Alembo, E.A. & Torka, T.T. (2023). Prevalence, contamination level, and associated factors of methicillin-resistant *Staphylococcus aureus* in raw cow milk at selected districts of Gamo Zone, Southern Ethiopia. *Veterinary Medicine International*, Article ID 6238754.

Aliyu, Y., Abdullahi, I.O., Whong, C.M.Z. & Olayinka, B.O. (2019). Antibiotic resistant phenotypes of *Staphylococcus aureus* isolated from fresh and fermented milk in parts of Nasarawa State, Nigeria. *African Journal of Microbiology Research*, 13(26), 446–456.

Aliyu, Y., Reuben, R.C., Abdullahi, I.O., Olayinka, B.O. & Abdullahi, M.S. (2022). A systematic review on the prevalence of multidrug-resistant *Staphylococcus aureus* from milk and milk products in Nigeria. *Pan African Medical Journal One Health*, 7, 15.

Alsowaida, Y.S., Benitez, G., Bin Saleh, K., Almangour, T.A., Shehadeh, F. & Mylonakis, E. (2022). Effectiveness and safety of ceftriaxone compared to

standard of care for treatment of bloodstream infections due to methicillin-susceptible *Staphylococcus aureus*: A systematic review and meta-Analysis. *Antibiotics*, 11(3), 375.

Anueyiagu, K.N., Benson, N.P., Daniel, J., Nze, C. & Esilonu, J.T. (2022). Prevalence of methicillin-resistant *Staphylococcus aureus* in bovine subclinical mastitis in Jos South Local Government Area of Plateau State, Nigeria. *European Journal of Veterinary Medicine*, 2(6), 7–11.

Badawy, B., Elafify, M., Farag, A.M.M., Moustafa, S.M., Sayed-Ahmed, M.Z., Moawad, A. A., Algammal, A.M., Ramadan, H. & Eltholth, M. (2022). Ecological distribution of virulent multidrug-resistant *Staphylococcus aureus* in livestock, environment, and dairy products. *Antibiotics*, 11(11), 1651.

Bitrus, D.E. (2021). *Developing geo-database for housing and facilities management in Sobi Barracks Ilorin, Kwara State, Nigeria*. *June 2020*.

Cha, J.O., Lee, J.K., Jung, Y.H., Yoo, J.I., Park, Y.K., Kim, B.S. & Lee, Y.S. (2006). Molecular analysis of *Staphylococcus aureus* isolates associated with staphylococcal food poisoning in South Korea. *Journal of Applied Microbiology*, 101(4), 864–871.

Clinical and Laboratory Standards Institute (CLSI). (2022). *Performance standards for antimicrobial susceptibility testing* (32nd ed.).

Daka, D., Gsilassie, S. & Yihdego, D. (2012). Antibiotic-resistance *Staphylococcus aureus* isolated from cow's milk in the Hawassa area, South Ethiopia. *Annals of Clinical Microbiology and Antimicrobials*, 11, 26

Elias, L., Balasubramanyam, A.S., Ayshpur, O.Y., Mushtuk, I.U., Sheremet, N.O., Gumeniuk, V.V., Musser, J.M.B. & Rogovskyy, A.S. (2020). Antimicrobial susceptibility of *Staphylococcus aureus*, *Streptococcus agalactiae*, and *Escherichia coli* isolated from mastitic dairy cattle in Ukraine. *Antibiotics*, 9(8), 469.

Esonu, D., Ismail, S., Ajala, A., Yusuf, S. & Otolorin, R. (2021). Occurrence and antimicrobial susceptibility patterns of *Staphylococcus aureus* and Salmonella species in fresh milk and milk products sold in Zaria and environs, Kaduna State, Nigeria. *Sahel Journal of Veterinary Science,* 18(2), 1-8.

Gaddafi, M.S., Yakubu, Y., Junaidu, A.U., Bello, M.B., Bitrus, A.A., Musawa, A.I., Garba, B. & Lawal, H. (2023). Occurrence of methicillin-resistant *Staphylococcus aureus* (MRSA) from dairy cows in Kebbi, Nigeria. *Iranian Journal of Veterinary Medicine*, 17(1), 19–26.

Gentilini, E., Denamiel, G., Llorente, P., Godaly, S., Rebuelto, M. & DeGregorio, O. (2000). Antimicrobial susceptibility of *Staphylococcus aureus* isolated from bovine mastitis in Argentina. *Journal of Dairy Science*, 83(6), 1224–1227.

Ghali-Mohammed, I., Odetokun, I.A., Raufu, I.A. & Adetunji, V.O. (2022). Handling practices and contamination of raw milk sold for consumption in markets of Kwara State, Nigeria. *Sokoto Journal of Veterinary Sciences*, 20(1), 50–58.

Ghali-Mohammed, I., Odetokun, I.A., Raufu, I.A., Alhaji, N.B. & Adetunji, V.O. (2023). Prevalence of *Escherichia coli* O157 isolated from marketed raw cow milk in Kwara State, Nigeria. *Scientific African*, 19, e01469.

Hassan, R., Aliyu, S., Adam, A., Mienda, B. & Muhammad, A. (2021). Prevalence and antibiotic susceptibility patterns of *Staphylococcus aureus* in locally pasteurised cow-milk sold at Dutse metropolis, Jigawa state, Nigeria. *International Journal of Biology Sciences*, 3(1), 29–33.

Kadariya, J., Smith, T.C. & Thapaliya, D. (2014). *Staphylococcus aureus* and staphylococcal food-borne disease: An ongoing challenge in public health. *BioMed Research International,* Article ID 827965.

Kou, X., Cai, H., Huang, S., Ni, Y., Luo, B., Qian, H., Ji, H. & Wang, X. (2021). Prevalence and characteristics of *Staphylococcus aureus* isolated from retail raw milk in Northern Xinjiang, China. *Frontiers in Microbiology*, 12, 705947.

Lozano, C., Gharsa, H., Ben Slama, K., Zarazaga, M. & Torres, C. (2016). *Staphylococcus aureus* in animals and food: Methicillin resistance, prevalence and population structure. a review in the African continent. *Microorganisms*, 4(1), 12.

Murray, C.J., Ikuta, K.S., Sharara, F., Swetschinski, L., Robles Aguilar, G., Gray, A., Han, C., Bisignano, C., Rao, P., Wool, E., Johnson, S.C., Browne, A.J., Chipeta, M.G., Fell, F., Hackett, S., Haines-Woodhouse, G., Kashef Hamadani, B.H., Kumaran, E.A.P., McManigal, B., … Naghavi, M. (2022). Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. *The Lancet*, 399(10325), 629–655.

Odetokun, I.A., Adetona, M.A., Ade-Yusuf, R.O., Adewoye, A.O., Ahmed, A.N., Ghali-Mohammed, I., Al-Mustapha, A.I. & Fetsch, A. (2023). *Staphylococcus aureus* contamination of animal-derived foods in Nigeria: a systematic review, 2002—2022. *Food Safety and Risk*, 10, 6.

Odetokun, I.A., Afolaranmi, Z.M., Nuhu, A.A., Borokinni, B.O., Ghali-Mohammed, I., Cisse, H. & Alhaji, N.B. (2022). Knowledge and self-reported food safety practices among meat consumers in Ilorin, Nigeria. *Dialogues in Health*, 1, 100039.

Odetokun, I.A., Ballhausen, B., Adetunji, V.O., Ghali-Mohammed, I., Adelowo, M.T., Adetunji, S.A. & Fetsch, A. (2018). *Staphylococcus aureus* in two municipal abattoirs in Nigeria: Risk perception, spread and public health implications. *Veterinary Microbiology*, 216, 52–59.

Odetokun, I.A., Borokinni, B.O., Bakare, S.D., Ghali-Mohammed, I. & Alhaji, N.B. (2021). A cross-sectional survey of consumers' risk perception and hygiene of retail meat: A nigerian study. *Food Protection Trends*, 41(3), 274-283.

Odetokun, I.A., Maurischat, S., Adetunji, V.O. & Fetsch, A. (2022). Methicillin-resistant *Staphylococcus aureus* from municipal abattoirs in Nigeria: Showing highly similar clones and possible transmission from

slaughter animals to humans. *Foodborne Pathogens and Disease*, 19(1), 56-61

Okpo, N.O., Abdullahi, I.O., Whong, C.M.Z. & Ameh, J.B. (2017). Occurrence and antibiogram of *Staphylococcus aureus* in dairy products consumed in parts of Kaduna State, Nigeria. *Bayero Journal of Pure and Applied Sciences*, 9(2), 225-229.

Oloso, N.O., Fagbo, S., Garbati, M., Olonitola, S.O., Awosanya, E.J., Aworh, M.K., Adamu, H., Odetokun, I.A. & Fasina, F.O. (2018). Antimicrobial resistance in food animals and the environment in Nigeria: A review. *International Journal of Environmental Research and Public Health*, 15(6), 1284.

Oludairo, O.O., Olatoye, O.I., Awoniyi, O.E., Adejumobi, O.A. & Aiyedun, J.O. (2020). Prevalence of methicillin resistant *Staphylococcus aureus* (MRSA) in cattle milk from dairy herds in Oyo State, Nigeria. *Journal of Veterinary and Biomedical Sciences*, 2(2), 72–83.

Olufemi, F., Akinduti, P., Keinde, O. & Odunfa, O. (2018). Prevalence and antibiogram of methicilin-susceptible *Staphylococcus aureus* (MSSA) isolated from raw milk of asymptomatic cows in Abeokuta, Nigeria. *Alexandria Journal of Veterinary Sciences*, 57(2), 34-40.

Omoshaba, E.O., Ojo, O.E., Sofela, O. & Onifade, O.I. (2018). Prevalence and antibiotic resistance patterns of methicillin-resistant *Staphylococcus aureus* in raw milk and soft cheese (wara) sold in Abeokuta, Nigeria. *Sokoto Journal of Veterinary Sciences*, 16(1), 1-8.

Park, J.Y. & Seo K.S. (2019). *Staphylococcus aureus*. In *Food Microbiology: Fundamentals and Frontiers* (pp. 555–584).

Rainard, P., Foucras, G., Fitzgerald, J.R., Watts, J. L., Koop, G. & Middleton, J.R. (2018). Knowledge gaps and research priorities in *Staphylococcus aureus* mastitis control. *Transboundary and Emerging Diseases*, 65, 149–165.

Ranzani, O.T., Motos, A., Chiurazzi, C., Ceccato, A., Rinaudo, M., Li Bassi, G., Ferrer, M. & Torres, A. (2020). Diagnostic accuracy of Gram staining when predicting staphylococcal hospital-acquired pneumonia and ventilator-associated pneumonia: a systematic review and meta-analysis. *Clinical Microbiology and Infection*, 26(11), 1456–1463.

Reiner, K. (2013). *Catalase Test Protocol*. *November 2010*, 1–9.

Sayed-Ahmed, Z.A.M., Ahmed, A.A., Amer, A.A. & Abdelshahi, Y.S.Y. (2020). Foodborne methicillin-resistant *Staphylococcus aureus* (FB-MRSA) in skimmed dairy products in Egypt: Prevalence, molecular characterization, and antimicrobial susceptibility. *Alexandria Journal of Veterinary Sciences*, 64(2), 78–86.

Sperber, W.H. & Tatini, S.R. (1975). Interpretation of the tube coagulase test for identification of *Staphylococcus aureus*. *Applied Microbiology*, 29(4), 502–505.

Tibebu, L., Belete, Y., Tigabu, E. & Tsegaye, W. (2021). Prevalence of *Staphylococcus aureus*, methicillin-resistant *Staphylococcus aureus* and potential risk factors in selected dairy farms at the interface of animal and human in Bishoftu, Ethiopia. *Veterinary Medicine: Research and Reports*, 12, 241–251.

Tong, S.Y.C., Davis, J.S., Eichenberger, E., Holland, T.L. & Fowler, V.G. (2015). *Staphylococcus aureus* infections: Epidemiology, pathophysiology, clinical manifestations, and management. *Clinical Microbiology Reviews*, 28(3), 603–661.

World Health Organization (WHO). (2017). *WHO publishes list of bacteria for which new antibiotics are urgently needed*. https://www.who.int/news/item/27-02-2017-who-publishes-list-of-bacteria-for-which-new-antibiotics-are-urgently-needed

World Population Review (WPR). (2023). *Ilorin Population 2023*. Ilorin Population. https://worldpopulationreview.com/world-cities/ilorin-population

Wu, W., Zhang, A., van Klinken, R.D., Schrobback, P. & Muller, J.M. (2021). Consumer trust in food and the food system: A critical review. *Foods*, 10(10), 2490.