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Research Article

Antibiotic Susceptibility Profile of Methicilin-Resistant *Staphylococcus aureus* Isolated from Cloaca and Stool Samples of Poultry Birds in Selected Farms in Malete, Kwara State

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ABSTRACT

The rise of methicillin-resistant *Staphylococcus aureus* (MRSA) infections in animals, especially poultry, represents a serious public health concern due to the possibility of zoonotic transmission. This study aimed to determine the antibiotic resistance profile and occurrence of *S. aureus* and MRSA that were isolated from the cloaca and faeces of chickens in a medium sized commercial farm in Malete in Kwara State. A total of 74 cloacal and stool samples were collected from layer and broiler birds across three medium-sized farms. *Staphylococcus aureus* was isolated and identified using standard microbiological and biochemical tests. Antibacterial susceptibility testing was performed using the Kirby-Bauer disk diffusion method against a panel of ten antibiotics. Methicillin resistance was phenotypically confirmed using cefoxitin (30µg) disk screen. The prevalence of *S. aureus* was 85.1% (63/74). Of these isolates, 28.6% (18/63) were confirmed as MRSA. The antibiotic susceptibility profile revealed high levels of multidrug resistance. The highest resistance rates were observed against cefuroxime (84.1%), cefotaxime (82.5%), and meropenem (76.2%). In contrast, the lowest resistance was noted against ofloxacin (6.4%) and augmentin (11.1%). Chi-square analysis indicated $p < 0.05$ between the isolation of *S. aureus* and resistance to beta-lactam antibiotics. This study demonstrates a high prevalence of multidrug-resistant *S. aureus* and MRSA in poultry from the study area. The findings highlight a critical reservoir of antimicrobial resistance and suggest that ofloxacin may be a more effective therapeutic option in this context, though its use should be controlled to prevent emergence of resistance.

Keywords: Antibacterial resistance; MRSA; Multidrug resistance; Nigeria; Poultry; Zoonosis

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INTRODUCTION

A bacterial pathogen, *Staphylococcus aureus,* can infect humans and animals causes with a variety of diseases (Lowy, 1998). Treatment regimens around the world have become extremely complex due to the advent of methicillin-resistant *Staphylococcus aureus* (MRSA), which is characterized by resistance to all beta-lactam medicines (Turner *et al.,* 2019). The MRSA has been found in community and livestock-associated reservoirs (LA-MRSA), despite its historical association with healthcare settings (Verkade and Kluytmans, 2014).

Due to the increased use of antimicrobial drugs for prophylaxis, growth promotion, and treatment brought about by the global expansion of animal husbandry, resistant bacteria are under selective pressure to evolve and spread (Van Boeckel *et al.,* 2015). One important industry in this dynamic is poultry production. The

gastrointestinal tract of birds harbour *S. aureus* and MRSA which may be isolated from cloacal and faecal samples (Akindolire *et al.,* 2018). The transmission of these resistant pathogens can occur through the food chain, direct contact with birds, or via contaminated environments, posing a direct threat to farm workers and, ultimately, consumers (Fetsch *et al.,* 2017).

In Nigeria, poultry farming is a major agricultural activity, yet comprehensive data on the prevalence and resistance profiles of MRSA in this sector, particularly in the Malete, remain scarce. Previous studies in other parts of Nigeria have reported alarming rates of antimicrobial resistance in poultry isolates (Odonkor and Addo, 2018), highlighting a pressing public health issue. This study therefore aimed to isolate, characterize, and determine the antibacterial susceptibility patterns of *S. aureus* and MRSA from poultry in selected farms in Malete, Nigeria. The findings will provide crucial baseline data to inform local antimicrobial use policies and mitigate the risk of zoonotic transmission.

MATERIALS AND METHODS

Study Area and Collection of Samples

This cross-sectional study was conducted in three (3) medium-sized commercial poultry farms in Malete, Nigeria. A total of seventy-four (74) samples, comprising cloacal swabs and fresh stool, were aseptically collected from both layer and broiler birds. Samples were placed in sterile containers, appropriately labeled, and transported in a cool box to the laboratory for immediate processing.

Isolation and Identification of *Staphylococcus aureus*

Samples were inoculated onto Mannitol Salt Agar (MSA) and incubated aerobically at 37°C for 24-48 hours. Presumptive *S. aureus* colonies (golden-yellow colonies) were sub-cultured on Blood Agar to obtain pure isolates. Conventional biochemical tests, including catalase, coagulase (tube and/or slide), and DNase tests, were performed for final confirmation (Cheesbrough, 2006).

Antibacterial Susceptibility Testing

Antimicrobial susceptibility testing was performed on confirmed *S. aureus* isolates using the Kirby-Bauer disk diffusion method on Mŭeller-Hinton Agar, as recommended by the Clinical and Laboratory Standards Institute (CLSI, 2020). The following antibiotic disks (Oxoid, UK) were used: Cefoxitin (30 µg), Cefuroxime (30 µg), Cefotaxime (30 µg), Meropenem (10 µg), Ceftazidime (30 µg), Ceftriaxone (30 µg), Ciprofloxacin (5 µg), Gentamicin (10 µg), Augmentin (Amoxicillin-clavulanate, 30 µg), and Ofloxacin (5 µg). *Staphylococcus aureus* ATCC 25923 was used as the quality control strain. Methicillin resistance was phenotypically defined as resistance to cefoxitin (Bale *et al.,* 2018).

Data Analysis

Data were entered into Microsoft Excel. The occurrence of *S. aureus* and MRSA was calculated as percentages. The resistance rates for each antibiotic were expressed as a percentage of the total isolates. A Chi-square test was performed using SPSS version 25.0 to assess the significance of the observed resistance patterns, with a p-value of ≤ 0.05 considered statistically significant.

RESULTS

A total of 63 out of 74 samples were confirmed positive for *Staphylococcus aureus*, yielding a prevalence of 85.1%. Phenotypic screening with cefoxitin disks identified 18 of the isolates as MRSA, resulting in an MRSA occurence of 28.6% among the *S. aureus* isolates. The antibacterial susceptibility profile of the 63 *S. aureus* isolates is presented in Figure 1. The isolates exhibited high levels of multidrug resistance. Resistance was most prevalent against cefuroxime (84.1%) and cefotaxime (82.5%), followed by meropenem (76.2%). In contrast, the lowest resistance was observed against ofloxacin (6.4%) and augmentin (11.1%).

Statistical analysis revealed a significant association between the source of isolation and resistance to multiple beta-lactam antibiotics ($p < 0.05$)

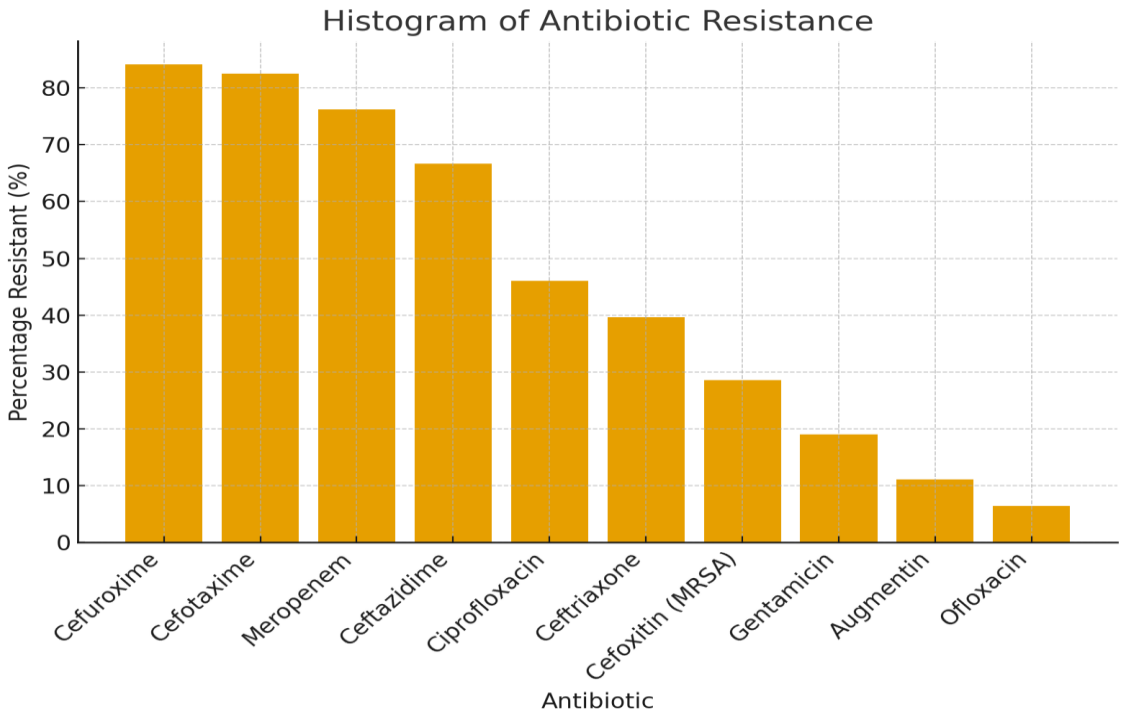

Figure 1. Antibiotics susceptibility profile of *Staphylococcus aureus* isolated from cloaca samples of poultry birds in Malete, Nigeria

DISCUSSION

This study an alarmingly high occurence of *S. aureus* (85.1%) and MRSA (28.6%) in poultry from the selected farms in Malete, Nigeria. The MRSA occurence highlights Nigeria as a possible hotspot for LA-MRSA and is in line with studies from other regions of the country, such as a study conducted in Ibadan that found a 26.5% MRSA occurrence in chicken (Adebowale *et al.,* 2019).

The significant degree of multidrug resistance, especially to beta-lactam antibiotics, is the most startling discovery. Since cefuroxime, cefotaxime, and meropenem are vital antibiotics for human medicine, the high rates of resistance to these drugs are quite worrisome (WHO, 2019). More genetic research is necessary since the resistance to the carbapenem and meropenem is especially worrisome and raises the possibility of extended-spectrum beta-lactamase (ESBL) producers or other intricate resistance mechanisms.

There is high resistance to many antibiotics but low resistance to ofloxacin (6.4%) and augmentin (11.1%). This pattern suggests that these antibiotics may not be commonly used in the studied farms, or that resistance has not yet been selected for. While this makes ofloxacin appear to be the most effective therapeutic option *in vitro*, its recommendation must be approached with extreme caution. Fluoroquinolones are classified as Highest Priority Critically Important Antimicrobials (HPCIAs) by the WHO (2019). Their indiscriminate use in veterinary practice could rapidly lead to resistance, rendering them ineffective for treating severe human infections.

The findings of this study are limited by its phenotypic approach. Molecular characterization of the MRSA isolates (e.g., detection of the *mecA* or *mecC* genes) and *SCCmec* typing would provide definitive confirmation and elucidate the genetic lineage and potential human transmissibility of these strains.

CONCLUSION

This study shows that multidrug-resistant *S. aureus* and MRSA are significantly found in poultry birds in Malete, Nigeria. A major threat to public health is the high occurrence of resistance to vital antibiotics. The chicken sector urgently needs to adopt and enforce antimicrobial stewardship programmes that encourage responsible drug use and investigate antibiotic.

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