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Original Article

Prevalence of Methicillin-Resistant *Staphylococcus aureus* and Their Antibiotic Susceptibility Test Among Diabetic Foot Infection Patients in Misurata City, Libya

Mohanned Al-Washaish[1]*, Nabil Elmahaishi[2], Asma Abu Zayan[1], Eman Al-Zawawi[1], Elham Al-Maliki[1]

[1]*Department of Biomedical Sciences, Faculty of Pharmacy, University of Misurata, Misurata, Libya*
[2]*Department of Pharmacology and Toxicology, Faculty of Pharmacy, University of Misurata, Misurata, Libya*
Corresponding Email. *mohanad.aloshesh@phar.misuratau.edu.ly*

Abstract

Diabetic foot infection (DFI) is a serious and common complication of diabetes mellitus. This study aimed to determine the prevalence of Methicillin-Resistant Staphylococcus aureus (MRSA) and assess the antibiotic resistance patterns of these isolates among DFIs. A total of 40 samples were collected, isolated, and identified using standard microbiological procedures and biochemical tests. Antibiotic susceptibility testing was carried out using the Disc Diffusion Method. The most common bacterial species detected was *Acinetobacter* spp. (30%), followed by *Staphylococcus aureus* (17.5%), *Pseudomonas aeruginosa* (17.5%), and *Klebsiella* spp. (15%), CONS (Coagulase-Negative Staphylococci) (15%), and *Proteus* spp. (5%). Out of the 7 isolates of *S. aureus*, 71.4% were MRSA, while 28.6% were MSSA (Methicillin-Sensitive Staphylococcus aureus). MRSA isolates showed a high rate of resistance to both AMP (Ampicillin) and AMC (Amoxicillin) (100%), followed by resistance to cefotaxime (80%) and ciprofloxacin (60%).

Keywords: Diabetic Foot Infection, Antibiotic Resistance, MRSA, MSSA.

Introduction

Diabetic foot infections (DFIs) are a frequent and serious complication of diabetes mellitus. These infections often begin as simple wounds but may progress into complex infections involving various microorganisms, especially when healing is delayed due to poor circulation or immune dysfunction. Clinical observations suggest that more than two-thirds of diabetic patients will experience such infections at some stage [1,2].

The nature of DFIs is typically polymicrobial, involving both aerobic and anaerobic organisms. Among these, *Staphylococcus aureus* is most commonly isolated and is particularly problematic due to its ability to adapt to host defenses and develop resistance to antibiotics. While mild infections are often caused by a single pathogen, more severe cases—especially those involving immunocompromised individuals—tend to present with multiple microbial species [3–5].

S. aureus has long been recognized for its dual behavior: it normally exists as part of the skin flora but can shift into a pathogenic form under the right conditions. Once it invades deeper tissues or the bloodstream, it can cause a wide spectrum of diseases, ranging from superficial skin infections to life-threatening systemic illnesses [6,7].

One of the major challenges in managing infections caused by *S. aureus* is its resistance to β-lactam antibiotics. These antibiotics target the bacterial cell wall, but the bacterium has developed mechanisms to neutralize them. In MRSA strains, for example, the mecA gene produces an altered penicillin-binding protein (PBP2a) that renders most β-lactam antibiotics ineffective [3,8,9]. The spread of MRSA outside hospital environments has complicated infection control efforts. Infections caused by MRSA are now observed in community settings as well, and strains have been linked to contact with livestock in certain cases [10]. These resistant strains are more difficult to treat, often requiring stronger antibiotics and prolonged treatment.

The increasing frequency of MRSA in DFIs is concerning, not only because of its resistance profile, but also due to the additional burden it places on healthcare systems. Poor antibiotic stewardship, prolonged hospital stays, and weakened host immunity all contribute to its spread and persistence [10,14]. This study aims to explore the microbial causes of diabetic foot infections, with particular emphasis on identifying the presence of MRSA and evaluating its resistance to commonly used antibiotics. Understanding these patterns is crucial for guiding effective clinical treatment and preventing further complications.

Methods

A total of 40 samples were collected from patients with DFI at Central Misurata Hospital, starting from December 15, 2023, to April 4, 2024. The samples were cultivated on blood and MacConkey agar for 24 hours at 37c°, then identified by macroscopic, microscopic and biochemical test like (coagulase, catalase, indole, triple sugar iron, and citrate test). To determine the ability of Staphylococcus aureus for methicillin resistance, a bacterial suspension was prepared in comparison with 0.5 MacFarland, then cultivated on Muller-Hinton agar and placed with cefoxitin discs for 24 hours at 37c° then the inhibition zone was measured and compared with CLSI 2023. The methicillin-resistant Staphylococcus aureus isolates

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determined their resistance to Ciprofloxacin, Ampicillin, Amoxicillin, and Ceftriaxone by the disc diffusion method. And statistical analysis was performed by SPSS 25.

Results

Distribution of gender among DFI.

(Figure 1) demonstrates a predominance of male patients, with 31 out of 40 cases (77.5%) compared to 9 female cases (22.5%), showing a statistically significant difference (p = 0.031).

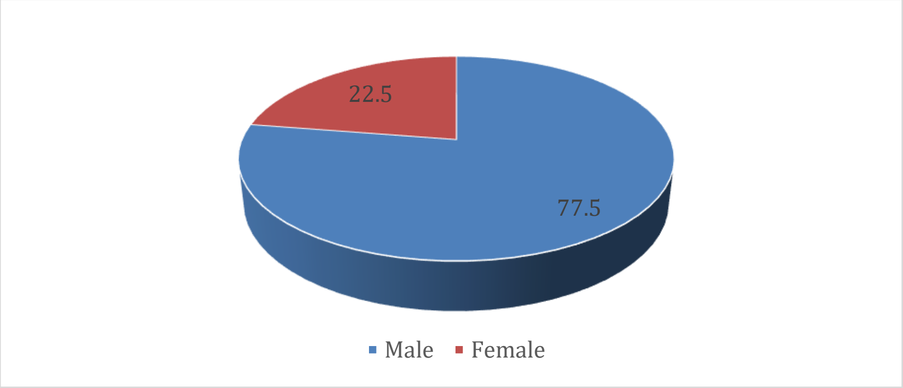

Figure 1. Distribution of gender among DFI.

Distribution of age groups among DFI

(Figure 2) illustrates that the most prevalent cases were observed in the 31–50 years age group, accounting for 57.5% of the total, while the least common cases were found in the 11-30 years range, representing 15%.

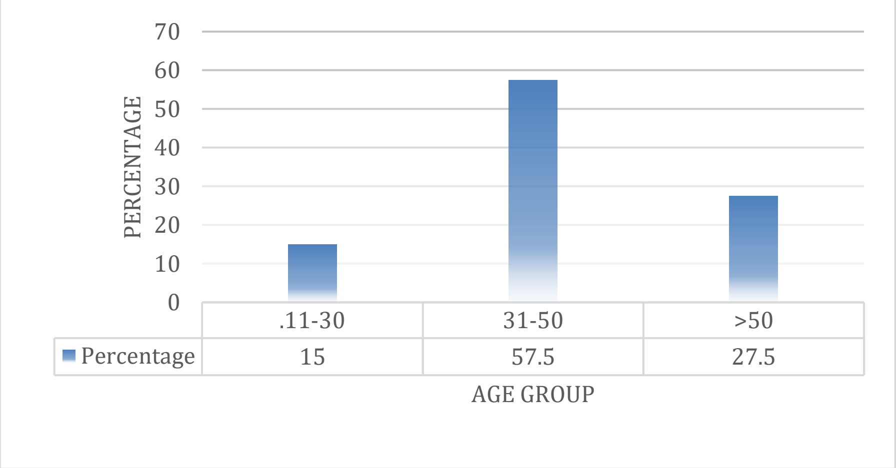

Figure 2. Distribution of age groups among DFI

Prevalence of bacterial Isolates among DFI patients.

Gram-negative bacterium, specifically *Acinetobacter* spp. (30%), *Pseudomonas aeruginosa* (17.5%), *Klebsiella* spp. (15%), and *Proteus* spp. (5%), were more frequently isolated than Gram-positive bacteria, which included *Staphylococcus aureus* (17.5%) and Coagulase-Negative Staphylococci (CONS) (15%). A statistically significant difference (p = 0.026) was observed in the isolation rates between Gram-positive and Gram-negative bacterial causes of infection, as shown in Table 1.

Table 1. Prevalence of bacterial Isolates among DFI patients.

Isolated Bacteria	**n**	**%**
Acinetobacter spp.	12	30%
Pseudomonas aeruginosa	7	17.5%
Staphylococcus aureus	7	17.5%
Klebsiella Spp.	6	15%
Coagulase-negative Staphylococcus	6	15%
Proteus Spp.	2	5%

Prevalence of MRSA among DFI patients

(Table 2) indicates that 71.4% of Diabetic Foot Infections (DFI) caused by Staphylococcus aureus were attributed to Methicillin-Resistant *Staphylococcus aureus* (MRSA), while the remaining 28.6% were due to

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Methicillin-Sensitive *Staphylococcus aureus* (MSSA).

Table 2. Prevalence of MRSA among DFIs patients.

***S. aureus* strain**	**n**	**%**
MRSA	5	71.4%
MSSA	2	28.6%

Antibiogram of MRSA.

(Table 3) demonstrates that 100% of the MRSA isolates exhibited resistance to Ampicillin (AMP) and Amoxicillin-clavulanic acid (AMC). Furthermore, 80% of the isolates were resistant to Cefotaxime (CTX), and 60% showed resistance to Ciprofloxacin (CIP).

Table 3. Antibiogram of MRSA

CIP		**CTX**		**AMC**		**AMP**	
R	**S**	**R**	**S**	**R**	**S**	**R**	**S**
3	2	4	1	5	-	5	-
60%	40%	80%	20%	100%	-	100%	-

" R = Resistance, S = Sensitive, CIP = Ciprofloxacin, CTX = Cefotaxime, AMC = Amoxicillin-clavulanic acid, AMP = Ampicillin"

Discussion

MRSA is considered a significant global health problem due to high resistance to a wide range of antibiotics. In our study, the findings revealed a higher prevalence of diabetic foot infections (DFIs) among male patients (77.5%) while (22.5%) female. This gender disparity is consistent with the study by Rajput *et al.*, (2023) [15], which also reported a predominance of male participants. Several biological and behavioral factors may contribute to this trend, including high rates of trauma, risk-related lifestyle habits such as smoking and alcohol consumption, and gender-specific physiological differences. Male skin tends to be thicker and may exhibit difficult healing, while hormonal and immunological variations could influence susceptibility to infection.

Age-group distribution showed that the majority of DFI cases (57.5%) occurred in the 31–50-year age group, aligning with the findings of Gebremeskel *et al.* (2022) [13]. Individuals within this age group are more likely to suffer from diseases such as diabetes, hypertension, and obesity, all known risk factors for impaired wound healing. Additionally, lifestyle pressures related to work, poor nutrition, stress, and reduced physical activity further predispose this group to infection. Also, the immune system, which begins to gradually decrease in this age group, may contribute to reduced host defense mechanisms and delayed tissue regeneration, thereby increasing infection susceptibility.

Microbiological analysis indicated a predominance of Gram-negative bacteria, particularly *Acinetobacter* spp. (30%), *Pseudomonas aeruginosa* (17.5%), *Klebsiella* spp. (15%), and *Proteus* spp. (5%). Gram-positive isolates included *Staphylococcus aureus* (17.5%) and coagulase-negative *Staphylococci* (15%). These results are in agreement with previous studies by Rajput *et al.* (2023), Tom *et al.* (2019), and Bessa *et al.* (2013) [15–16-17], which identified similar patterns in wound infections. The predominance of Gram-negative organisms may be related to the environmental conditions of the hospital, as these species are known for their resistance and ability to survive in moist and contaminated environments. In contrast, *Staphylococcus* spp. are part of the normal skin flora, making them common opportunistic pathogens in wound infections.

Regarding antibiotic resistance patterns, the present study showed that 71.4% of *S. aureus* isolates were methicillin-resistant (MRSA), while only 28.6% were methicillin-sensitive (MSSA). This distribution is supported by the results of Gurung *et al.* (2020) and Karki *et al.* (2021) [2-6], who similarly reported high MRSA prevalence in wound infections. The high proportion of MRSA highlights the growing concern of antibiotic resistance in healthcare settings and indicates the need for regular surveillance and targeted antimicrobial therapy.

The antibiotic susceptibility profile of MRSA isolates in this study revealed complete resistance (100%) to ampicillin and amoxicillin-clavulanic acid. Additionally, 80% were resistant to cefotaxime, and 60% to ciprofloxacin. These findings are consistent with those reported by Gurung *et al.* (2020) and Prajapati *et al.* (2018) [6-7], confirming the widespread resistance of MRSA to β-lactam antibiotics. This resistance is primarily attributed to the production of β-lactamase enzymes, which deactivate β-lactam antibiotics by breaking the β-lactam ring. Other contributing factors may include prior antibiotic exposure, self-medication, lack of infection control, and compromised host immunity, all of which may facilitate the emergence of multidrug-resistant organisms.

Conclusion

Diabetes and its associated complications, particularly Diabetic Foot Infections (DFIs), represent a substantial global health burden. These infections are frequently polymicrobial, with Staphylococcus aureus

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being identified as one of the most significant causative microorganisms. Phenotypic analysis revealed a higher prevalence of Methicillin-Resistant Staphylococcus aureus (MRSA) among isolates, demonstrating considerable resistance to various classes of antibiotics. This widespread antibiotic resistance poses a significant and escalating threat to public health.

Conflict of interest. Nil

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