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Bacterial Isolates and Antibiotic Sensitivity of Patients with Diabetic Foot Infections at Hayatabad Medical Complex of Peshawar, Pakistan

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Abstract: Diabetic foot ulcers (DFUs), often exacerbated by secondary bacterial infections, are a major complication of diabetes and a leading cause of morbidity. Understanding the spectrum of bacterial pathogens and their profiles of antibiotic resistance is essential for developing effective treatment strategies. This study aimed to identify bacterial isolates from the DFUs and evaluate their susceptibility to commonly used antibiotics. A total of 186 patients with the DFUs were examined at Hayatabad Medical Complex of Peshawar in Pakistan over a three-month period. Samples were collected from infected ulcer sites and cultured with standard microbiological techniques. Bacterial identification was performed with conventional methods, and antibiotic susceptibility testing was then conducted by using the Kirby-Bauer disk diffusion method. Gram-Negative bacteria were predominant, with *Pseudomonas aeruginosa*, *Proteus mirabilis*, *Acinetobacter spp.*, *Escherichia coli*, *Klebsiella spp.*, *Enterobacter spp.*, and *Streptococcus pyogenes* being the most commonly isolated organisms. Gram-Positive isolates including *Staphylococcus aureus* and *Staphylococcus epidermidis*, *P. aeruginosa*, and *Enterobacter spp.* showed high sensitivity to Gentamicin, Meropenem, and Imipenem. In contrast, *Acinetobacter spp.* and *Klebsiella spp.* exhibited significant resistance, particularly to carbapenems. *Staph. aureus* was generally sensitive to first-line antibiotics, such as Vancomycin and Rifampicin whereas *Staph. epidermidis* demonstrated multidrug resistance including pan-drug resistance in some cases. These findings highlighted the complex and resistant microbial profiles of diabetic foot infections, thus emphasizing the importance of the culture-guided antibiotic therapy. The emergence of carbapenem-resistant strains underlined the requisites for continuous surveillance, judicious antibiotic use, and improved infection control strategies to aid the recovery of patients.

Keywords: Diabetes mellitus; Diabetic foot infection; Antibiotic resistance; Gram-Negative; Gram-Positive; Antibiotic therapy; Antibiotic susceptibility

1. Introduction

Diabetes mellitus is a clinical syndrome characterized by reduced insulin secretion or action. It is a major global health concern, affecting a significant proportion of the population annually (Perim et al., 2015). Among diabetic individuals, approximately one-fourth develop foot ulcers during their lifetime (Lipsky et al., 2006). These individuals are prone to bacterial infections than non-diabetic individuals, with diabetic foot infections (DFIs) being particularly common. Such infections often involve antibiotic-resistant bacteria, including *Staphylococcus aureus*, which accounts for 10–15% of wound flora. The presence of resistant organisms contributes to prolonged hospital stays and increased healthcare burdens (Lavery et al., 2007).

The misuse and overuse of antibiotics, the frequent hospital admissions, and the chronic nature of diabetic foot wounds contribute to the emergence of multidrug-resistant (MDR) bacteria in these patients (Kandemir et al., 2007). The microbial profile of the DFIs varies significantly from one hospital to another and even from patient to patient (El-Tahawy, 2000), thus making standardized treatments more complicated.

In Pakistan, the DFIs are highly prevalent and typically arise from two primary complications of diabetes: peripheral neuropathy and ischemia. Neuropathy results from chronic hyperglycemia, which damages peripheral

nerves, hence reducing the patient's ability to perceive pain or trauma, especially in the lower extremities (Wukich et al., 2018). Ischemia, caused by poor vascular circulation, further compromises wound healing and increases the risk of infection. If untreated, the DFIs can progress to osteomyelitis, a severe bone infection (Senneville et al., 2006), and may ultimately require limb amputation (Hobizal & Wukich, 2012).

Antibiotic resistance poses a serious threat to the successful treatment of the DFIs. Bacterial species such as *Escherichia coli*, *Klebsiella spp.*, and *Staphylococcus aureus* have shown growing resistance to commonly prescribed antibiotics and so complicates treatment regimens. Resistance rates are significantly higher in diabetic patients than in non-diabetics, with approximately 70% of bacterial isolates from diabetic patients demonstrate antibiotic resistance. The global spread of antibiotic-resistant bacteria is a critical public health issue. According to the Centers for Disease Control and Prevention (CDC), more than 2 million individuals in the United States acquire antibiotic-resistant infections each year, resulting in approximately 23,000 deaths. Contributing factors include the inappropriate use of antibiotics in healthcare and agriculture, as well as the environmental dissemination of resistant strains through wastewater and other sources (Gentry, 1993).

Moreover, antibiotic resistance is a significant concern in the treatment of the DFIs. Many bacteria, such as *E. coli*, *Klebsiella*, and *Staphylococcus aureus*, have developed resistance to commonly used antibiotics, so it becomes harder to treat infections. Resistance rates are higher in diabetic patients compared to non-diabetic individuals, with approximately 70% of bacterial isolates from diabetic patients showing resistance. The spread of antibiotic-resistant bacteria is a growing global issue, with the CDC reporting that more than 2 million people in the USA develop antibiotic-resistant infections each year, resulting in 23,000 deaths. This problem is exacerbated by the overuse of antibiotics, both in healthcare and agriculture, and the spread of resistant strains through water treatment plants and other environmental sources (Larsson & Flach, 2022).

In response to these challenges, newer antibiotics have been developed for the management of the DFIs. Ertapenem, a broad-spectrum carbapenem, is effective against Methicillin-resistant *Staphylococcus aureus* (MRSA), Gram-Negative bacteria, and anaerobes, though it lacks efficacy against *Pseudomonas aeruginosa* (Tentolouris et al., 2006). Linezolid, an oxazolidinone antibiotic, demonstrates excellent activity against the MRSA and Vancomycin-resistant enterococci, with 100% oral bioavailability (Goldstein et al., 1996). Moxifloxacin, a fluoroquinolone, also shows promising activity against the MRSA and Gram-Negative organisms, although it may not be fully effective against the MRSA in clinical settings. Newer agents such as dalbavancin and ceftobiprole have also shown potential in the treatment of moderate to severe DFIs (Mougakou et al., 2023).

The aim of the present study is to isolate and identify bacterial pathogens from diabetic foot ulcers (DFUs) and to assess their antibiotic susceptibility profiles against commonly used antibiotics. The findings of this study assist in guiding effective and evidence-based antibiotic therapy, and help combat the growing threat of antimicrobial resistance in diabetic foot infections.

1.1 Recent Advancement in Diabetic Foot Infection Research and Their Relevance to This Study

Recent systematic reviews and primary studies have documented rising dominance of Gram-Negative bacteria, notably *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Escherichia coli*, and *Acinetobacter baumannii* in the DFIs, especially across the Asian countries. One meta-analysis of 73 studies indicated Gram-Negative organisms accounted for approximately 77% of the DFI pathogens, with polymicrobial infections in over 70% of cases (Ahmad et al., 2022). In Pakistan, regional hospital-based work similarly reported high rates of multidrug resistance (MDR), with *E. coli* and *Klebsiella* notably exhibiting resistance to Ampicillin (~72 %) and Ciprofloxacin (~55 %), while Carbapenems and Piperacillin-tazobactam retained relatively higher efficacy; the MRSA was isolated in over 60% of Gram-Positive samples (Ali et al., 2025).

Emerging evidence also highlights the prevalence of Carbapenemase-producing pathogens including *K. pneumoniae*, *A. baumannii*, *E. coli*, and *P. aeruginosa*, as many of which harbor bla_NDM and bla_KPC genes. These organisms frequently form robust biofilms, hence significantly hampering antibiotic effectiveness and complicating treatment strategies (Hammour et al., 2023).

Beyond traditional antibiotics, innovative therapeutic approaches are under exploration. Antimicrobial peptides (AMPs) and synthetic AMP-polymers, when used in combination with existing antibiotics such as Colistin or Tobramycin, have demonstrated potent synergistic effects against the MDR pathogens prevalent in the DFIs including *P. aeruginosa*, *Klebsiella*, *A. baumannii*, and *S. aureus*. Similarly, antimicrobial photodynamic therapy (aPDT), which utilizes photosensitizers activated by light to produce reactive oxygen species, shows promising activity against biofilm-associated and drug-resistant bacteria. aPDT demonstrates a low likelihood of resistance development and becomes a potential adjunct therapy in the DFU management (Li, 2023).

Finally, improved antibiotic stewardship strategies tailored for resource-limited settings have recently been developed. These include early gram-stain–guided empirical therapy aligned with the AWaRe framework of the World Health Organization to enable rapid decision-making and create cost-effective settings where full laboratory susceptibility profiling may be delayed or become unavailable (Monami et al., 2020).

2. Materials and Methods

2.1 Study Design and Setting

This cross-sectional study was conducted in the Hayatabad Medical Complex (HMC) of Peshawar, Pakistan, over a period of three months. The primary objective was to isolate bacterial pathogens from patients with diabetic foot ulcer (DFU) and evaluate their profiles of antibiotic susceptibility. The study included diabetic patients of all ages and sexes with clinically diagnosed foot ulcers. Patients with non-diabetic ulcers or systemic infections unrelated to diabetes were excluded from the study to guarantee specificity.

2.2 Sample Collection and Transportation

Proper aseptic techniques were strictly followed during sample collection to minimize contamination. Initially, the infected foot ulcer sites were examined carefully. The surface of the wound was cleansed using sterile normal saline to remove superficial debris and contaminants. Samples were then collected by scraping the wound base with a sterile curette or swabbing deep tissue from the ulcer margins. All samples were immediately placed in sterile containers, stored in a refrigerated environment at 2–8℃, and transported to the microbiology laboratory within 24–48 hours upon collection to ensure viability and prevent bacterial degradation.

2.3 Isolation and Identification of Gram-Negative Bacteria

Gram-Negative bacteria were isolated using MacConkey agar, a selective medium containing bile salts and crystal violet to inhibit the growth of Gram-Positive organisms. Plates were incubated aerobically at 37℃ for 18–24 hours. Following colony growth, Gram staining was performed to confirm Gram-Negative morphology. Further identification of isolates was carried out through standard biochemical tests, including oxidase, indole, catalase, urease, and nitrate reduction tests, according to the conventional microbiological protocols.

2.4 Isolation and Identification of Gram-Positive Bacteria

To isolate Gram-Positive bacteria, samples were cultured on blood agar and Phenylethyl Alcohol Agar (PEA), which selectively inhibit Gram-Negative bacteria. Plates were incubated at 37℃ for 18–24 hours under aerobic conditions. Gram staining was performed to confirm Gram-Positive morphology. Biochemical identification was conducted using catalase and coagulase tests for species differentiation, especially for *Staphylococcus aureus* and *Staphylococcus epidermidis*.

2.5 Antibiotic Susceptibility Testing

Antibiotic sensitivity of all confirmed bacterial isolates was determined using the Kirby-Bauer disk diffusion method on Mueller-Hinton agar, following the Clinical and Laboratory Standards Institute (CLSI) standards. The bacterial suspensions conform to the 0.5 McFarland turbidity standard and evenly inoculated onto the agar surface. Antibiotic discs were applied aseptically and plates were incubated at 37℃ for 18–24 hours. Meanwhile, zones of inhibition were measured in millimeters, and sensitivity or resistance was interpreted according to the CLSI standards.

The following antibiotics were used in the testing of susceptibility (showed in Table 1).

Table 1. Antibiotics used for testing susceptibility

Serial No.	Name of Antibiotic	Serial No.	Name of Antibiotic
1	Co-Trimaxazole	15	Fosfomycin
2	Gentamicin	16	Linezolid
3	Meropenem	17	Teicopianin
4	Imipenem	18	Fusidic acid
5	Co-Amoxiclav	19	Vancomycin
6	Ceftazidime	20	Rifampicin
7	Cefalexin	21	Penicillin
8	Cefotaxime	22	Doxycycline
9	Colistin	23	Clindamycin
10	Ciprofloxacin	24	Erythromycin
11	Tigecycline	25	Chloramphenicol
12	Polymxin-B	26	Nitrofurantoin
13	Amikacin	27	Cefoperazone/Sulbactam
14	Minocycline	28	Piperacillin+Tazobactam

2.6 Statistical Analysis

Both qualitative and quantitative data were compiled and analyzed using Microsoft Excel (version 2007). Descriptive statistics, including means and percentages, were calculated to summarize the distribution of bacterial isolates and their corresponding resistance patterns. No inferential statistics were applied in this study.

3. Results

The current study was conducted to obtain bacterial isolates and check their antibiotic sensitivity in patients suffering from the DFUs. The study was held in Hayatabad Medical Complex of Peshawar for a 3-month duration. During this period, 186 patients were examined irrespective of their genders and age groups; only diabetic patients with foot ulcers were included. The selected patients were carefully examined while samples were collected in a sterile environment. The collected samples were transported for further processing. Under the sterile environment and inside biosafety cabinets, the samples were cultured for Gram-Positive and Gram-Negative bacteria. From these 186 samples, different bacterial isolates were isolated, i.e., mono-cultured, di-cultured and mixed cultured. Each cultured isolate was confirmed via Gram staining and bio-chemical analysis. Among the 186 patients, 174 mono-microbial, 10 di-microbial and 7 mixed culture isolates were isolated; the bacterial isolates consist of both Gram-Positive and Negative bacteria as in Figure 1. Among the mono-microbial isolates, 34.23% were Gram-Positive bacteria whereas 62.01% were Gram-Negative bacteria as shown in Figure 2.

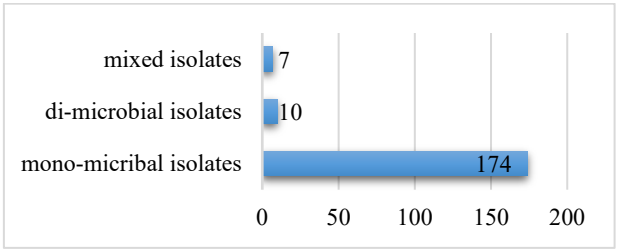

Figure 1. Mono-microbial, di-microbial and mixed isolates in patients infected with diabetic feet

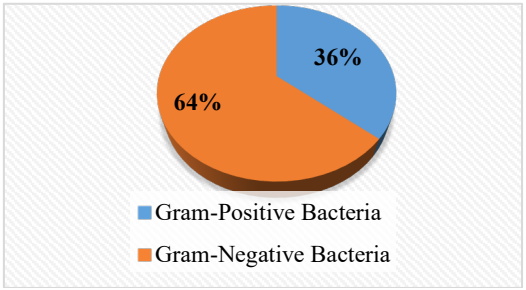

Figure 2. Pie chart representing the percentages of Gram-Positive and Gram-Negative bacteria

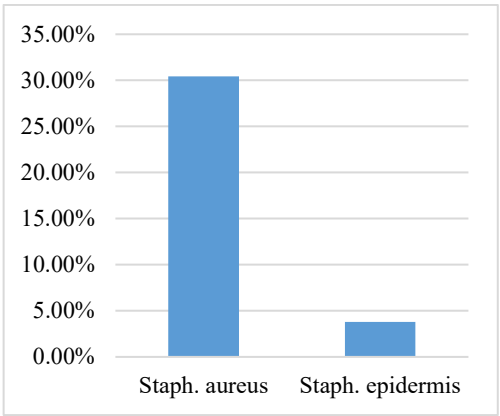

Figure 3. Gram-Positive bacteria present in diabetic foot infections

Gram-Positive bacteria include 30% *Staph. Aureus* out of the total isolates and 4% *Staph. epidermis* out of the 186 isolates as shown in Figure 3. Gram-Negative bacteria include *Prot. mirabilis*, *P. aeruginosa, E. coli, Acinetobacter spp., Klebsiella spp., Strep. pyogenes, Coliform spp.,* and *Enterobacter spp.* The percentage of isolated Gram-Negative bacteria are plotted in the line graph of Figure 4.

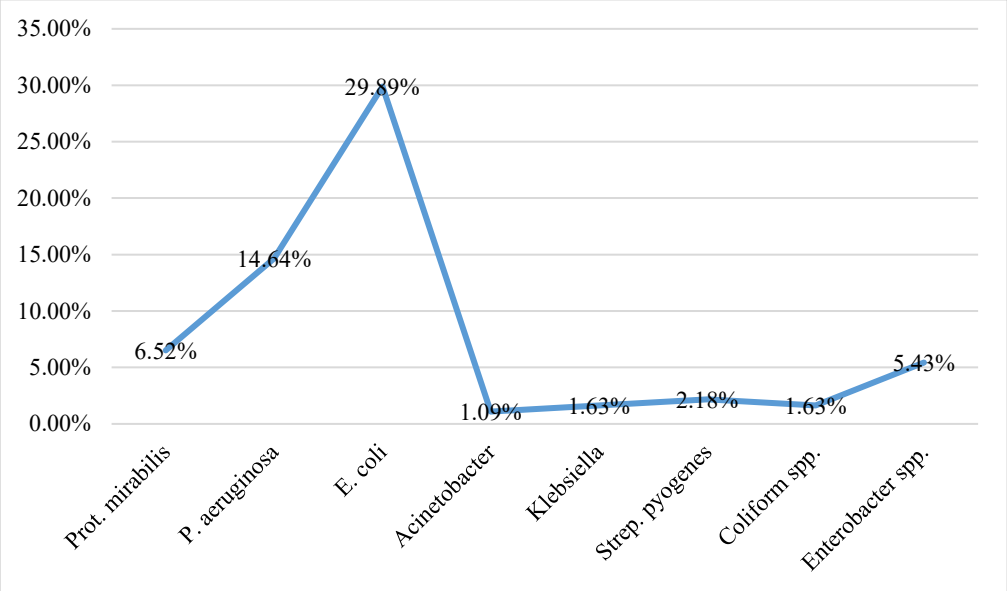

Figure 4. Gram-Negative bacteria present in diabetic foot infections

After identification of bacterial isolates, the sensitivity of both Gram-Positive and Gram-Negative bacteria were checked against commonly used antibiotics:

Gram-Negative bacteria included *Prot. mirabilis, P. aeruginosa, E. coli, Acinetobacter spp., Klebsiella spp., Strep. pyogenes, Coliform spp.,* and *Enterobacter spp.* Their sensitivity was checked against commonly used antibiotics in Hayatabad Medical Complex of Peshawar; they included Gentamicin, Meropenem, Imipenem, Co-Amoxiclav, Ceftazidime, Cefalexin, Cefotaxime, Colistin, Ciprofloxacin, Tigecycline, Polymxin-B, Amikacin, Minocycline, Fosfomycin, Linzolid, Teicopianin, Fusidic acid, Vancomycin, Rifampicin, Penicillin, and Doxycyclin. The antibiotic showed sensitivity, intermediate and resistant effect. ***Enterobacter spp.*** showed **sensitivity** to Gentamicin, Meropenem, Imipenem, Ciprofloxacin, Amikacin, efeperazone/sulbactam, Piperacillin+Tazobactam; **intermediate effect** to Colistin, Tigecycline, Polymxin-B, Minocycline; and **resistant** to Co-Amoxiclav, Ceftazidime, Cefalexin, Cefotaxime, Co-Trimaxazole. ***Prot. mirabilis*** showed **sensitivity to** Meropenem, Imipenem, Ciprofloxacin, Tigecycline, Amikacin, efeperazone/sulbactam, Piperacillin+Tazobactam; **intermediate effect** to Gentamicin and **resistant** to Co-Amoxiclav, Ceftazidime, Cefalexin, Cefotaxime, Colistin, Polymxin-B, Minocycline and Co-Trimaxazole. ***P. aeruginosa*** showed **sensitivity to** Gentamicin**,** Meropenem, Imipenem, Ceftazidime, Amikacin, efeperazone/sulbactam, Piperacillin+Tazobactam; **Intermediate effect** to Colistin, Polymxin-B and **resistant** to Co-Amoxiclav, Co-Trimaxazole, Tigecycline, Ciprofloxacin, Cefotaxime and Cefalexin. ***E. coli*** showed **sensitivity** to Gentamicin, Meropenem, Imipenem, Tigecycline, Amikacin, Amikacin, efeperazone/sulbactam, Piperacillin+Tazobactam; **intermediate effect** to Colistin and Polymxin-B and **resistant** to Co-Amoxiclav, Ceftazidime and Ciprofloxacin. ***Acinetobacter*** showed **sensitivity** to Amikacin, Fosfomycin and Nitrofurantoin; **intermediate effect** to Colistin and **resistant** to Co-Amoxiclav, Ceftazidime and Ciprofloxacin, Gentamicin, Meropenem, Imipenem, Tigecycline, efeperazone/sulbactam, Piperacillin+Tazobactam, Polymxin-B. ***Klebsiella*** showed **sensitivity** to Gentamicin, Meropenem, Imipenem, Ciprofloxacin, Amikacin, efeperazone/sulbactam, Piperacillin+Tazobactam and Co-Trimaxazole; **intermediate effect** to Colistin and Tigecycline and **resistant** to Co-Amoxiclav, Ceftazidime, Cefalexin, Cefotaxime and Polymxin-B. ***Strep. pyogenes*** showed **sensitivity** to Co-Amoxiclav, Co-Trimaxazole, Teicopianin, Fusidic acid, Vancomycin, Rifampicin, Penicillin, Doxycyclin**,** and **resistant to** Clindamycin, Chloramphenicol, Erythromycin, Ciprofloxacin and Polymxin-B. The last bacterial isolate among the Gram-Negative bacteria, ***Coliform spp.*** showed **sensitivity** to Meropenem, Imipenem, Ciprofloxacin, Tigecycline, Piperacillin+Tazobactam, Co-Trimaxazole, Amikacin and **resistant to** Gentamicin, Co-Amoxiclav, Cefalexin, Colistin, Cefotaxime, Polymxin-B, Co-Trimaxazole and Minocycline. The details of each bacterial isolate are given in Table 2.

Table 2. Profiles of Gram-Negative bacterial isolates and their antibiotic sensitivity

Name of Antibiotic	Name of Isolates							
	Enterobacter spp.	*Prot. mirabilis*	*P. aeruginosa*	*E. coli*	*Acinetobacter*	*Klebsiella*	*Strep. pyogenes*	*Coliform spp.*
Gentamicin	S	I	S	S	R	S	R	R
Meropenem	S	S	S	S	R	S	-	S
Imipenem	S	S	S	S	R	S	-	S
Co-Amoxiclav	R	R	R	R	R	R	S	R
Ceftazidime	R	R	S	R	R	R	-	R
Cefalexin	R	R	R	-	R	R	-	R
Cefotaxime	R	R	R	R	R	R	-	R
Colistin	I	R	I	I	I	I	-	R
Ciprofloxacin	S	S	R	R	R	S	R	S
Tigecycline	I	S	R	S	R	I	-	S
Polymxin-B	I	R	I	I	R	R	R	R
Amikacin	S	S	S	S	S	S	-	S
Minocycline	I	R	R	S	-	R	-	R
Fosfomycin	-	-	-	-	S	-	-	-
Linzolid	-	-	-	-	-	-	S	-
Teicopianin	-	-	-	-	-	-	S	-
Fusidic Acid	-	-	-	-	-	-	S	-
Vancomycin	-	-	-	-	-	-	S	-
Rifampicin	-	-	-	-	-	-	S	-
Penecillin	-	-	-	-	-	-	S	-
Doxycyclin	-	-	-	-	-	-	S	-
Clindamycin	-	-	-	-	-	-	R	-
Erythromycin	-	-	-	-	-	-	R	-
Chloramphenicol	-	-	-	-	-	-	R	-
Nitrofurantoin	-	-	-	-	S	-	-	-
Efeperazone / Sulbactam	S	S	S	S	R	S	-	R
Piperacillin + Tazobactam	S	S	S	S	R	S	-	S
Co-Trimaxazole	R	R	R	-	R	S	S	R

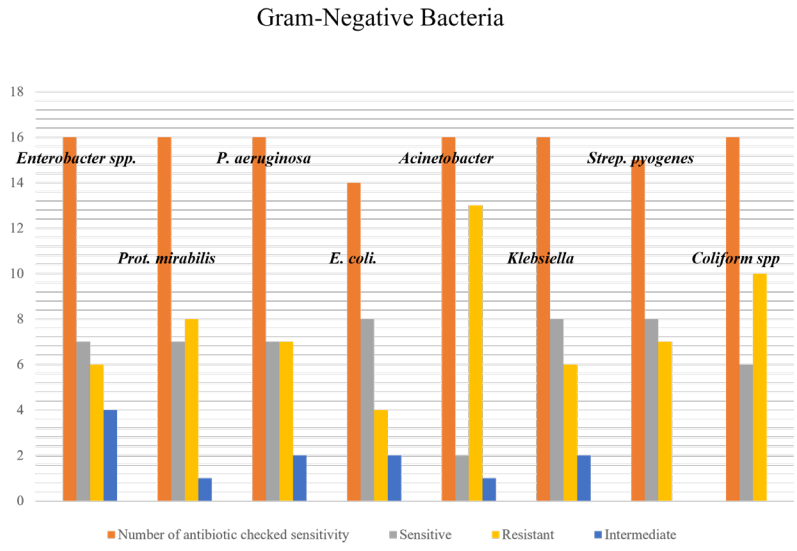

Figure 5. Gram-Negative bacteria and their antibiotic sensitivity

Each Gram-Negative isolate was checked against the specific antibiotics. *Enterobacter spp.* checked its sensitivity against 16 antibiotics and showed sensitivity to 43.75%, resistance to 37.5%, and intermediate effect to 18.75% of the antibiotics. *Prot. mirabilis* showed sensitivity to 43.75%, resistance to 50%, and intermediate effect to 6.25% of the antibiotics. *P. aeruginosa,* a prominent Gram-Negative bacteria, showed sensitivity to 43.75% of the antibiotics, 43.75% remained ineffective and showed an intermediate effect to 12.5% of the antibiotics. *E. coli,* a common isolate in urinary tract infection, was susceptible to 57.14% of the antibiotics and unsusceptible to

28.57% of the total antibiotics; it exhibited an intermediate effect to 14.28% of the antibiotics. *Acinetobacter* showed antibiotic sensitivity to 12.5% of the antibiotics, resistance to 81.57% and intermediate effect to 6.25%. *Klebsiella,* a bacterial isolate, exhibited sensitivity to half of the antibiotics, resistance to 37.5% and intermediate effect to 12.25% of the antibiotics. As regards *Strep. pyogenes,* the bacterial isolate was found sensitive to 53.33% antibiotics and resistant to 46.67% antibiotics. It did not show any intermediate effect to the antimicrobial drugs. *Coliform spp.* demonstrated antimicrobial susceptibility to 37.5% and resistance to 62.25% of the tested antibiotics, with no intermediate effect as shown in Figure 5.

Staph. aureus and *Staph. epidermidis* are the only two Gram-Positive bacteria isolated from patients suffering from diabetic foot infections. Their sensitivity was checked against commonly used antibiotics in Hayatabad Medical Complex of Peshawar, with the use of Kirby-Bauer disk diffusion method; the antibiotics included **Gentamicin, Co-Amoxiclav, Ciprofloxacin, Polymxin-B, Linzolid, Teicopianin, Fusidic acid, Vancomycin, Rifampicin, Penicillin, Doxycyclin, Clindamycin, Erythromycin, Chloramphenicol** and **Co-Trimaxazole**. *Staph. aureus* showed sensitivity to **Gentamicin, Co-Amoxiclav, Ciprofloxacin, Linzolid, Fusidic acid, Vancomycin, Rifampicin, Doxycyclin, Clindamycin, Erythromycin, Chloramphenicol,** and **Co-Trimaxazole**; resistance to **Polymxin-B** and **Penicillin** and intermediate effect to **Teicopianin**. *Staph. epidermidis* exhibited susceptibility to **Co-Amoxiclav, Fusidic acid, Rifampicin, and Doxycyclin**; resistance to **Gentamicin, Ciprofloxacin, Linzolid, Vancomycin, Penicillin, Clindamycin, Erythromycin, Chloramphenicol** and **Co-Trimaxazole** and intermediate effect to **Polymxin-B** and **Teicopianin.** The details are in Table 3.

Table 3. Profile of Gram-Positive bacteria and their antibiotic sensitivity

Name of Antibiotic	Name of Isolates	
	Staph. aureus	*Staph. epidermidis*
Gentamicin	S	R
Co-Amoxiclav	S	S
Ciprofloxacin	S	R
Polymxin-B	R	I
Linzolid	S	R
Teicopianin	I	I
Fusidic Acid	S	S
Vancomycin	S	-
Rifampicin	S	S
Penicillin	R	R
Doxycyclin	S	S
Clindamycin	S	R
Erythromycin	S	R
Chloramphenicol	S	R
Co-Trimaxazole	S	R

Staph. aureus demonstrated sensitivity to 80%, resistance to 13.33% and intermediate effect to 6.67% of the antibiotics. *Staph. epidermidis* exhibited antimicrobial susceptibility to 28.57%, showed resistance to 57.14% and demonstrated 14.28% intermediate effect of the antibiotics, showed in Figure 6.

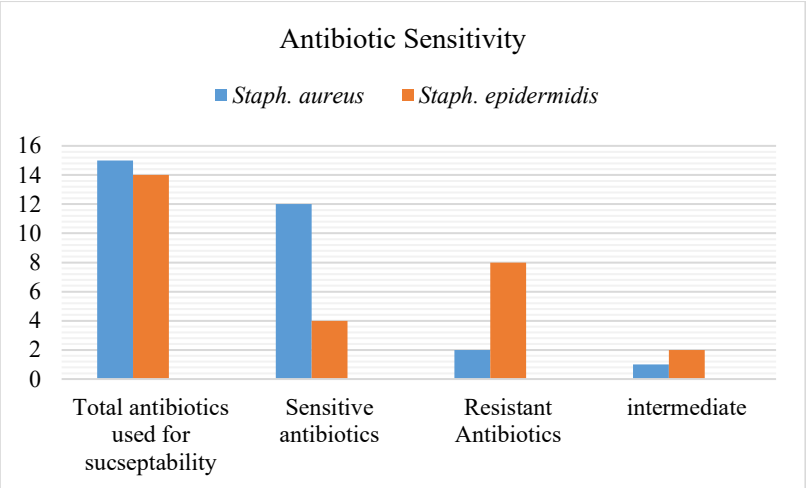

Figure 6. Antibiotic sensitivity of Gram-Positive bacteria**,** *Staph. aureus* and *Staph. Epidermidis*

4. Discussion

The current study provided insights into the usage pattern of antibiotics, culture isolates and their resistance to commonly used antibiotics. The results showed both Gram-Positive and Gram-Negative bacterial stains. It emphasized the complexity of diabetic foot infections and their appropriate antibiotic therapies. Moreover, it enhanced one's understanding of antibiotic sensitive profile that dwelt into appropriate therapeutic strategies.

The incidence of Gram-Negative bacteria persisted to be dominant at 62.01% whereas the Gram-Positive bacteria lagged behind at 34.23%. This report aligned with the previous reports to show a high prevalence of Gram-Negative in patients with the DFIs (Banashankari et al., 2012). The current study of Gram-Negative bacterial isolates included *Pseudomonas aeruginosa*, *Proteus mirabilis, Acinetobacter spp.*, *Streptococcus pyogenes*, *Coliform spp.*, *Klebsiella spp.*, *Enterobacter spp.,* and *Escherichia coli* whereas Gram-Positive included *Staphylococcus aureus* and *Staphylococcus epidermidis.* These isolates, involving mono-culture, di-culture and mixed, showed variations and a diverse nature of infections in patients with the DFU.

The predominant Gram-Negative microbes, especially *P. aeruginosa* and *E. coli*, naturally colonize feet and predominantly favour immune-compromised diabetic patients. It creates an environment for the progression of these pathogens (Sohail et al., 2022). *Klebsiella* and *Acinetobacter*, multi-drug resistant strains, are the organisms associated with prolonged hospitalization of the diabetic patients (Ajigbewu et al., 2025; Kyriakidis et al., 2021).

The clinical isolates and their sensitivity to the commonly used antibiotics were tested and this revealed critical data of their relative effectiveness. The result demonstrated that *P. aeruginosa* and *Enterobacter spp.* had the highest sensitivity to the commonly used antibiotics, Gentamicin, Meropenem, Imipenem, and Amikacin. The study aligned with previous reports, regardless of *P. aeruginosa,* a common multi-drug resistance strain, and showed sensitivity (Kwon & Armstrong, 2018). Conversely, *Acinetobacter spp.* was highly resistant to most of the common antibiotics; this aligned with the previous results. This bacterial isolate was known for its intrinsic property of resistance, thus posing notorious challenges to health.

The resistance showed by *Acinetobacter* and *Klebsiella* against commonly used antibiotics, like Meropenem and Imipenem, was worth concerning (Li et al., 2023; Nordmann et al., 2012). These major antibiotics were used as first line of treatment against major Gram-Negative infections. The result revealed increasingly world-wide issue of carbapenem-resistant strains, and challenged to manage and treat with alternative therapies (Nordmann et al., 2012).

Staphylococcus aureus, a Gram-Positive bacteria, showed sensitivity to several anti-microbial drugs. These included Ciprofloxacin, Vancomycin, and Gentamicin, and were used as first line of treatment. Resistance to Penicillin and Polymyxin-B aligned with previous reports, in which *Staphylococcus aureus* showed resistance to *β*-lactam drugs (Reveles et al., 2016). It was highly encouraging to discover that *Staphylococcus aureus* showed sensitivity to Vancomycin and Rifampicin, medicines to cure patients with the DFU.

Furthermore, *Staphylococcus epidermidis,* another Gram-Positive bacteria and pan-drug resistant strain, showed resistance to Gentamicin, Ciprofloxacin, and Penicillin. This report was parallel to previous studies that demonstrated *Staphylococcus epidermidis* as an opportunistic and highly resistant bacteria to commonly used antibiotics. Polymyxin-B and Teicoplanin showed intermediate effect to *Staphylococcus epidermidis*, hence suggesting its evasion from further antibiotics (Tong et al., 2015).

5. Conclusions

The current study highlighted the prevalence and antibiotic resistance of microbial isolates from patients suffering from the DFIs. On the one hand, the Gram-Negative bacteria, particularly *Acinetobacter spp.* and *Pseudomonas aeruginosa*, demonstrated high resistance to commonly used anti-microbial drugs. On the other hand, *Staph. aureus*, a Gram-Positive bacteria exhibited a high sensitivity pattern. These findings stressed personalized treatment plans and emphasized the necessity of continuous monitoring of antibiotic resistance trends to improve the outcomes of patients.

5.1 Clinical Implications and Recommendations

The findings from this study underscored the critical need for tailored antibiotic therapy in the DFIs. Given the high rates of antibiotic resistance, particularly among Gram-Negative organisms like *Acinetobacter* and *Klebsiella*, as well as the observed resistance in *Staphylococcus epidermidis*, empirical therapy based solely on clinical presentations may not be sufficient. Instead, the identification of bacterial species and their profiles of antibiotic susceptibility are crucial for effective treatment.

Clinicians should consider utilizing broad-spectrum antibiotics, followed by targeted therapy based on the specific resistance patterns of the isolates. In the case of resistant Gram-Negative pathogens, the use of combination therapy or alternative antibiotics may help achieve therapeutic success. Moreover, the findings highlighted the necessity for enhanced control measures against infections so as to prevent the spread of multi-

drug-resistant organisms in healthcare settings, where diabetic patients are at a higher risk of developing severe infections.

Data Availability

The datasets used and analyzed during this study are not publicly available due to restrictions from the corresponding author but can be provided upon reasonable request.

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Conflicts of Interest

The authors declare no conflicts of interest.

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