Govindaswamy et al | Article 9184
J Pure Appl Microbiol. 2025. doi: 10.22207/JPAM.19.3.21
Received: 12 December 2023 | Accepted: 23 June 2025

P-ISSN: 0973-7510; E-ISSN: 2581-690X

RESEARCH ARTICLE OPEN ACCESS

Bacteriological Profile and Antimicrobial Susceptibility Patterns in Patients with Diabetic Foot Infections in a Tertiary Care Hospital in South India

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Abstract

Patients with diabetes mellitus frequently develop foot infections, which can progress to gangrene and ultimately limb amputation. This study aimed to determine the microbiological profile and antibiotic susceptibility patterns of the causative agents of infections in diabetic feet. This six-month prospective observational study included 78 Type 2 diabetic patients with Wagner's grade 1 or higher. The microbiological profile and patterns of antibiotic susceptibility of the agents responsible for the infections in diabetic feet were determined. Culture tests were carried out on the tissue or pus that had been removed from the ulcer bases. An analysis of antibiotic sensitivity was performed following the identification of the organisms. Gram-negative pathogens (88%) were more prevalent than Gram-positive pathogens (12%). It was found that 41% of the patients had polymicrobial disease. *Escherichia coli* (17%), *Klebsiella pneumoniae* (17%), *Staphylococcus aureus* (13.8%), and *Pseudomonas aeruginosa* (12.7%) appeared to be the most frequent pathogens found in isolations. Ciprofloxacin was more resistant to *Escherichia coli*, whereas cotrimoxazole was more resistant to *K. pneumoniae.* In addition, *Acinetobacter baumannii*, a multidrug-resistant bacterium, was identified. The presence of multidrug-resistant organisms in diabetic foot infection treatment plan is important. The results of the present study further highlight the necessity of choosing antimicrobial treatments based on antimicrobial sensitivity patterns displayed by isolates and culture outcomes.

Keywords: Antimicrobial Susceptibility, Diabetic Foot Infections, Gram-negative Bacteria, Multidrug-resistance, Bacteriological Profile

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Citation: Govindaswamy S, Subbarayan E, Jaishwanth VM, Kumar SSA, Santhosh S, Renji SA. Bacteriological Profile and Antimicrobial Susceptibility Patterns in Patients with Diabetic Foot Infections in a Tertiary Care Hospital in South India. *J Pure Appl Microbiol.* Published online 13 August 2025. doi: 10.22207/JPAM.19.3.21

INTRODUCTION

According to the International Diabetes Federation (IDF), an estimated 588.7 million adults (aged 20-79 years) worldwide were living with diabetes as of 2024. This number is projected to increase to 852.5 million by 2050, highlighting a significant global rise in diabetes prevalence.[1] It is projected that by the year 2025, the number of individuals with diabetes residing in India will reach 57 million.[2] The prevalence of skin wounds, particularly chronic ulcers, is higher among individuals with diabetes as a result of neuropathy, vascular disease, or trauma.

Peripheral neuropathy and peripheral artery disease are common complications observed in diabetic individuals, resulting in foot nerve damage. The immune systems of individuals with diabetes exhibit certain impairments that remain incompletely comprehended, thereby posing challenges in the prevention and management of illnesses.[3] Diabetes patients are at a high risk of developing foot ulcers, the most common cause of hospitalization,[4] with a lifetime incidence rate ranging from 12% to 25%, posing a significant public health concern.[2,5]

Diabetes patients with infected ulcers have a higher rate of limb amputation compared to those without diabetes, with a 40:1 ratio.[6,7] Co-occurrence of diabetic foot infection and foot ischemia also increases the likelihood of amputation.[8]

The aetiology of foot infections in individuals with diabetes is multifaceted, with host-related abnormalities, including immunopathies, neuropathies, and arteriopathies, playing a predominant role in the incidence and severity of infections. Pathogen-related factors, such as virulence, antibiotic resistance, and microbial load, are of secondary importance.[9,10] The optimal selection of antibiotic therapy requires the identification of the specific causative infections, as a diabetic foot infection (DFI) can arise from a variety of different organisms, either independently or in conjunction.

Medical practitioners should avoid prescribing unnecessary antibiotics, as this can lead to adverse effects, high costs, and antibiotic resistance.[11,12] Preventing foot problems requires precise patient evaluation, prompt identification, and efficient infection control measures. A comprehensive understanding of bacterial isolates associated with diabetic foot infections is essential for effective treatment strategies, mitigating resistance patterns, and minimizing healthcare costs. This comprehensive understanding is crucial for effective management of foot problems.

The objective of the present prospective observational study was to evaluate the diverse microorganisms that cause infection in DFI and analyse their antibiotic susceptibility profiles.

MATERIALS AND METHODS

This prospective observational study included 78 patients with type 2 diabetes and diabetic foot infections admitted to a Quaternary care hospital in South India between June and December 2022. This study was approved by the institutional ethical committee with reference number EC/AP/946/07/2022 dated 13/07/2022. The sample size was calculated using the single population proportion formula with a prevalence of 6% in the South Indian diabetic scenario.[13] Inclusion criteria were patients aged >18 years, of any sex, irrespective of antibiotic use, and culture reports within 5 days of admission or during admission. We excluded patients with foot ulcers and foot infections without diabetes mellitus

Table 1. The baseline demographic and clinical characteristics of the patients

Variables	Observations (N = 78)
Mean Age (years)	60.82 ± 9.8
Mean HbA1c (%)	9.775 ± 2.26
Male	58 (74.4%)
Female	20 (25.6%)
Average duration of diabetes (years)	8 ± 3.2
Co-morbid conditions	
Hypertension	23 (29.5%)
Coronary Artery Disease	15 (19.2%)
Sepsis	17 (21.8%)
Cerebrovascular Disease	3 (3.8%)
Thyroid	4 (5.1%)
Peripheral Vascular Disease	6 (7.7%)
Others	10 (12.9%)

Table 2. Grade-wise distribution of Diabetic foot ulcer and number of organisms

Ulcer Grade	Diabetic foot ulcer N = 78 (%)	Monomicrobial	Polymicrobial
Grade I	12 (15.4%)	10 (83%)	2 (17%)
Grade II	19 (24.4%)	13 (68%)	6 (32%)
Grade III	28 (35.9%)	15 (54%)	13 (46%)
Grade IV	15 (19.2%)	7 (47%)	8 (53%)
Grade V	4 (5.1%)	1 (25%)	3 (75%)
Total	78 (100%)	46 (59%)	32 (41%)

Table 3. Gram-negative and Gram-Positive distribution of isolated microorganisms

Microorganisms	Number of Microbes	Percentage of micro-organisms (%)
Gram-negative isolates	69	88
Gram-positive isolates	25	32

and diabetic foot infections with negative culture reports from the study.

We used the Wagner Classification System to grade the foot lesions. The VITEK 2 fully automated ID/AST MIC system was used to perform the antibiotic susceptibility test. Data were recorded according to the patient's age, gender, antimicrobial therapy, nature of the clinical specimen, species of the isolated pathogen, and antibiotic susceptibility of the clinical isolates. Statistical analyses were performed using Microsoft Excel software.

RESULTS

The study included 78 patients diagnosed with diabetic foot infections, of which 58 (74.4%) were male. The mean age of the entire patient population was 60.82 years with a standard deviation of 9.8 years. The present study revealed that hypertension (29.5%) and sepsis (21.8%) were the two most commonly occurring co-morbidities in the examined cohort. The demographic and clinical baselines of the patients are presented in Table 1. Grade III diabetic foot ulcers were observed in the greatest proportion (35.9%), whereas Grade V ulcers were observed in the smallest proportion (5.1%) among the 78 patients who participated, as illustrated in Table 2. A total of 94 culture specimens were acquired, with 45 (47.9%) pus samples and 49 (52.1%) tissue samples. The results of our study indicate an average isolation value of 1.2.

Among the cohort of 78 patients under investigation, 59% (n = 46) and 41% (n = 32) presented monomicrobial and polymicrobial pathogens, respectively.

The prevalence of ulcers with polymicrobial aetiology increased in proportion to the severity of the ulcer. Specifically, 75% of gangrenous ulcers were found to have a polymicrobial aetiology (Table 2). *Escherichia coli* (17%), *Klebsiella pneumoniae* (17%), *Staphylococcus aureus* (*S. aureus*) (13.8%), and *Pseudomonas aeruginosa* (12.7%) were the six most prevalent microbial isolates.

Streptococcus agalactiae (*S. agalactiae*) and *Streptococcus pyogenes* (*S. pyogenes*) had a prevalence of 6.38% each. The prevalence of gram-negative pathogens was found to be 88%, as indicated in Table 3 and 4 displays the findings of antibiotic susceptibility. *E. coli* exhibited greater susceptibility towards the antibiotics Cefuroxime, Levofloxacin, Linezolid, Meropenem, and Penicillin, while displaying resistance towards Ciprofloxacin. *K. pneumoniae* exhibited greater susceptibility to Ertapenem, Minocycline, and Tigecycline, while displaying resistance to Cotrimoxazole. *S. aureus* exhibits greater susceptibility to Teicoplanin, while demonstrating resistance to Erythromycin. *P. aeruginosa* exhibits greater susceptibility to Amikacin and Doripenem, while displaying resistance to the combination of Ticarcillin and Clavulanate. *Streptococcus agalactiae* exhibits greater susceptibility to ceftriaxone and resistance to amoxicillin/clavulanate combination.

Acinetobacter baumannii (*A. baumannii*), a multidrug-resistant organism, was identified in patients with Grade V diabetic foot infections and

Table 4. Antibiotic Sensitivity profile of isolated microorganisms

Antibiotics	*E. coli* (%)	*K. pneumoniae* (%)	*S. aureus* (%)	*P. aeruginosa* (%)	*S. agalactiae* (%)	*S. pyogenes* (%)
Amikacin	88	88	-	100	-	-
Amoxicillin/Clavulanate	19	69	-	-	20	-
Ampicillin	80	-	-	-	-	83
Cefaperazone/Sulbactam	75	88	-	-	-	-
Cefipime	44	68	-	67	-	-
Cefotaxime	-	-	-	-	-	83
Ceftriaxone	25	75	-	-	80	83
Cefuroxime	100	75	-	-	-	-
Chloramphenicol	-	88	-	-	40	-
Ciprofloxacin	16	56	-	75	-	-
Clindamycin	80	-	25	-	-	-
Colistin	31	88	-	-	-	-
Cotrimoxazale	44	25	77	-	60	-
Doripenem	-	-	-	100	-	-
Doxycline	50	75	-	-	-	-
Ertapenem	88	94	-	-	-	-
Erythromycin	-	-	23	-	40	-
Fosfomycin	100	81	-	-	-	-
Gentamicin	50	75	77	-	-	-
Imipenem	-	88	-	77	-	-
Levofloxacin	100	-	-	75	-	83
Linezolid	100	-	85	-	-	100
Meropenem	100	88	-	92	-	-
Minocycline	63	94	-	-	-	-
Oxacillin	-	-	46	-	-	-
Penicillin	100	-	-	-	-	83
Piperacillin/Tazobactam	75	75	-	-	-	-
Teicoplanin	-	-	92	-	-	-
Tetracycline	40	-	77	-	40	83
Ticarcilin/Clavulanate	-	-	-	58	-	-
Tigecycline	50	94	-	-	-	-
Vancomycin	-	-	90	-	-	100

was susceptible only to colistin and tigecycline. *S. pyogenes* exhibited susceptibility to Linezolid and Vancomycin but exhibited a small degree of resistance to antibiotic treatment.

DISCUSSION

The mean age of the study participants was 60.82 ± 9.8 years. Jain and Barman reported a similar observation in the northeastern region of India in 2017.[14] In developing countries, diabetic foot ulcers (DFUs) are most commonly observed in individuals aged 45 to 64 years. For instance, a recent study from rural Central India reported that the majority of DFU cases occurred in the 45-54 years (27.6%) and 55-64 years (27.6%) age groups. Our study shows a similar age distribution, reinforcing that DFUs predominantly affect middle-aged to older adults in these settings.[15] Within this particular age bracket, there is a higher prevalence of comorbidities such as sepsis, neuropathy, hypertension, and peripheral vascular disease, which may potentially serve as the aetiology for the issue at hand. Of the 78 patients diagnosed with DFI, 58 (74.4%) were male. Jain and Barman's study conducted in north-eastern India revealed that diabetic foot infections are more prevalent among men than women.[14] One possible explanation for this phenomenon is that males tend to participate in greater amounts of outdoor physical activity, often in hot and humid conditions, and may not prioritise proper foot care. The research conducted

revealed that a significant proportion of patients with diabetic foot infections (DFI) self-reported an advanced stage of infection, specifically Wagner Grade III and higher. The prevalence of Grade III diabetic foot ulcer was highest at 35.9% in this study. The lack of adequate foot care among the general public and healthcare practitioners is commonly associated with this issue.[16] The present study observed a predominance of monomicrobial culture growth, accounting for 59% of the samples analysed. Polymicrobial growth was detected in 41% of the patients.

The results of our study are consistent with the research conducted by Shah *et al*. in the year 2021.[17] In 2020, Goh *et al.* reported that a majority of patients, specifically 85%, who had diabetic foot infections were diagnosed with polymicrobial infections.[18] The observed discrepancy may be attributed to the characteristics of the sampled population. In our study, we observed a corresponding increase in the percentage of ulcers with a polymicrobial aetiology as the grade of the ulcer deteriorated. In particular, the proportion of gangrenous ulcers with polymicrobial aetiology reached 75%.

The microbiological examination of diabetic foot infections (DFI) in this study revealed a predominance of gram-negative organisms (88%) over gram-positive organisms (22%), consistent with previous research findings.[14,19,20] The average isolation value was 1.2 in this study.

Comparable findings were reported in Malaysia (1.5),[21] as well as in diverse areas of India, such as Maharashtra (1.8),[22] Chandigarh (1.5),[23] and New Delhi (2.3).[24] The research conducted by Zahid *et al.* in underdeveloped nations has shown that *E. coli* is the most prevalent gram-negative bacteria.[25] *E. coli, P. aeruginosa,* and *K. pneumoniae* were identified as the prevailing gram-negative microorganisms. The results of the susceptibility testing revealed that *E. coli* exhibited resistance to ciprofloxacin, ceftriaxone, colistin, and cotrimoxazole. The organism was highly sensitive to cefuroxime, levofloxacin, Linezolid, Meropenem, penicillin, and amikacin. The substantial escalation of this infection could be attributed to prolonged hospitalization and inappropriate administration of antibiotics. Ertapenem, minocycline, and tigecycline demonstrated greater efficacy against *K. pneumoniae*, whereas cotrimoxazole exhibited lower efficacy.

The results indicated that *P. aeruginosa* exhibited resistance to cefepime, ticarcillin/clavulanate, and levofloxacin, whereas amikacin, doripenem, and imipenem demonstrated complete sensitivity, as determined by the study. Three gram-positive bacterial strains, *S. aureus, S. agalactiae,* and *S. pyogenes*, were found to be the most commonly isolated strains. *S. aureus* exhibited resistance to erythromycin *S. agalactiae* exhibits greater susceptibility to ceftriaxone and resistance to the amoxicillin and clavulanate combination. The susceptibility of the pathogen was found to be noteworthy towards teicoplanin, vancomycin, and gentamycin.

The results of the susceptibility testing indicated that *S. pyogenes* exhibited susceptibility to Linezolid and Vancomycin, while displaying minimal resistance to the antibiotics tested. *A. baumannii*, a pathogen that is resistant to multiple drugs, was identified in patients with Grade V diabetic foot infections and exhibited susceptibility solely to tigecycline and colistin.

A similar discovery was made in previous studies.[26-31] Possible factors contributing to the emergence and spread of multidrug-resistant (MDR) bacteria include recurrent hospital admissions, recent utilisation of broad-spectrum antibiotics, inadequate implementation of surgical source control measures, persistent wounds, inappropriate antibiotic prescribing practices, and dissemination of resistance genes through various modes of transmission.

Healthcare professionals should exercise discretion when administering antibiotics and ensure timely and appropriate dosage. In addition, relevant governing bodies should regularly monitor antibiotic consumption to enhance patient outcomes and decrease the incidence of amputation.[32]

On the basis of the results of culture reports, it is recommended that clinicians consider utilising more targeted spectrum therapy. Adequate and timely surgical intervention is essential for mitigating potential sources of infection.

As a result of these developments, the use of antibiotics in an untargeted and disproportionate manner is expected to decrease.

CONCLUSION

This study reveals that gram-negative microorganisms are the predominant type of microorganism found in diabetic foot infections, with *E. coli* and *K. pneumoniae* being the most common. The prevalence of monomicrobial infections is higher than polymicrobial infections. The presence of multidrug-resistant microorganisms is a concern. The present study emphasizes the importance of selecting antibiotic therapy based on culture outcomes and antimicrobial susceptibility profiles. Factors such as overall health, glycemic control, illness severity, drug allergies, prior antibiotic usage, antibiotic activity, excretion, and toxicity influence antibiotic therapy selection. A thorough diagnosis, appropriate treatment, and effective management of foot infections are crucial for optimal outcomes.

Limitations

This research exhibits a potential constraint in terms of a limited sample size, which may impede the generalizability of the findings. In addition, the study was conducted for a brief duration. The extended duration of the study could facilitate the examination of pathogens in diabetic foot infections and enable the evaluation of antibiotic susceptibility and resistance patterns.

ACKNOWLEDGMENTS

None.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

ES and SG conceptualized and designed the study. VMJ, SSAK, SS and SAR performed execution, acquisition of data, analysis, and interpretation. SG wrote, reviewed and edited the manuscript. All authors read and approved the final manuscript for publication.

FUNDING

None.

DATA AVAILABILITY

The datasets generated and/or analysed during the current study are available from the corresponding author on reasonable request.

ETHICS STATEMENT

This study was approved by the Institutional Ethical Committee, Koval Medical Center and Hospital Limited, Coimbatore, India, with reference number EC/AP/946/07/2022 dated 13/07/2022.

INFORMED CONSENT

Written informed consent was obtained from the participants before enrolling in the study.

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