ORIGINAL ARTICLE

A Retrospective Study of Clinical, Microbiological, and Ulcer Severity Patterns of Diabetic Foot Presenting to a Tertiary Care Hospital

Asif Dabeer Jafri[1*], Srikant Kumar Dhar[2], Muktesh Singh[3], Ratender Kumar Singh[4]

Received: 25 May 2026; *Accepted:* 17 July 2026

Abstract

Background: Diabetic foot ulcers (DFUs) are a major complication of diabetes and are frequently associated with infection, antimicrobial resistance, and limb amputation. Regional Indian data on microbiological patterns and antimicrobial resistance remain limited.

Methods: This retrospective observational study evaluated 40 patients with DFUs presenting to a tertiary care hospital over a 12-month period. Clinical and laboratory parameters, including glycated hemoglobin (HbA1c), C-reactive protein (CRP), total leukocyte count (TLC), and serum albumin, were analyzed. Ulcer severity was graded using the Wagner classification, and wound cultures with antibiotic susceptibility testing were performed.

Results: Most patients were male (70%) and aged 40–60 years. Mean HbA1c was significantly higher in male than in female patients (13.99 ± 1.40 vs 10.45 ± 1.41; $p < 0.05$). Monomicrobial infections predominated, with Gram-negative organisms accounting for 70.3% of isolates, followed by Gram-positive bacteria (22.2%) and fungi (7.4%). Polymicrobial infections occurred in 12.5% of cases. Gram-negative isolates showed universal sensitivity to colistin, whereas *Acinetobacter* spp. demonstrated high resistance to other antibiotics. Higher Wagner grades were significantly associated with elevated HbA1c and CRP levels. All amputations occurred in male patients with polymicrobial infections.

Conclusion: Gram-negative pathogens predominated in DFUs in this setting, contrasting with the Gram-positive predominance reported in Western literature. Poor glycemic control and elevated inflammatory markers were associated with increasing ulcer severity and amputation risk.

Keywords: Amputation; Diabetes mellitus complications, Diabetic foot, Diabetic foot infections, Foot ulcer, Glycated hemoglobin.

Journal of The Association of Physicians of India (2026): 10.59556/japi.74.1675

Introduction

Diabetic foot ulcers (DFUs) are among the most common long-term complications of poorly controlled diabetes and are frequently associated with inadequate glycemic control, peripheral vascular disease, neuropathy, poor foot care, and trauma. In diabetic people, the lifetime chance of getting a DFU is estimated to be 12 to 25%.[1,2] Diabetes mellitus (DM) is now the most common cause of nontraumatic amputations in the world, and approximately 1% require amputation. Patients with DFU are approximately two to three times more likely to die than patients who do not have DFU.[3,4] Therapy targeting recognized causal organisms can considerably improve outcomes and reduce infection-related morbidities.

The microbiological profile of Diabetic foot infections (DFIs) varies considerably across geographical regions. Studies from Western countries commonly report Gram-positive organisms, particularly Staphylococcus aureus and Streptococci, as the predominant pathogens.[5] In contrast, several studies from India and other developing countries have demonstrated a predominance of Gram-negative bacteria along with an increasing burden of multidrug-resistant (MDR) organisms. The choice of empirical antibiotics frequently relies on information from Western nations, which may not be suitable for application in these regions.[6]

Hyperglycemia impairs both innate and adaptive immune responses, increasing susceptibility to bacterial infections in patients with diabetes. DFIs are further promoted by peripheral neuropathy, vasculopathy, impaired circulation, and immunocompromised states.[7,8] Inflammatory biomarkers such as C-reactive protein (CRP), procalcitonin (PCT), total leukocyte count (TLC), and interleukin-6 (IL-6) have been associated with infection severity, lower-limb amputation, soft tissue infection, osteomyelitis, and sepsis in diabetic patients. Several clinical factors, including age, male gender, duration of diabetes, poor glycemic control, peripheral vascular disease, neuropathy, renal dysfunction, osteomyelitis, gangrene, and elevated leukocyte count, have also been identified as predictors of amputation in patients with diabetic foot ulcers.[9,10]

Despite the substantial burden of DFIs in India, region-specific data integrating microbiological spectrum, antimicrobial susceptibility patterns, ulcer severity, and biochemical markers remain limited. Therefore, the present study was undertaken to evaluate the microbiological profile and antimicrobial resistance patterns of diabetic foot infections and to assess their association with glycemic status, inflammatory markers, ulcer severity, and clinical outcomes, including amputation.

Materials and Methods

Study Design and Ethical Considerations

This retrospective observational study was conducted in the Department of General Medicine at SUM Hospital, Bhubaneswar, India, over a 12-month period from 1 January 2025 to 1 January 2026. A total of 40 patients with diabetic foot ulcers were included in the analysis. Ethical approval for the study was obtained from the Institutional Ethics Committee-Clinical Research and Studies (IEC No. ECR/1604/Inst/OD/2021/03/01/2026). Owing to the retrospective nature of the study and the use of anonymized clinical data, the requirement for informed consent was waived by the Ethics Committee.

[1]Senior Registrar, Department of Emergency Medicine, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh; [2]Professor, Department of General Medicine, IMS and SUM Hospital, Bhubaneswar, Odisha; [3]Assistant Professor, Department of Anesthesiology, MLN Medical College, Prayagraj; [4]Professor and Head, Department of Emergency Medicine, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India; *Corresponding Author

How to cite this article: Jafri AD, Dhar SK, Singh M, *et al.* A Retrospective Study of Clinical, Microbiological, and Ulcer Severity Patterns of Diabetic Foot Presenting to a Tertiary Care Hospital. J Assoc Physicians India 2026;74(8):52–56.

Inclusion and Exclusion Criteria

Adults aged 20–80 years with diabetic foot ulcers were included in the study. Patients with nondiabetic foot ulcers, immunocompromised states, chronic inflammatory diseases, hemoglobinopathies, pregnancy, or chronic liver disease were excluded.

Clinical and Laboratory Assessment

Demographic details, duration of diabetes, associated comorbidities, and clinical findings were recorded for all patients. Ulcer severity was graded using the Wagner classification system. Peripheral neuropathy was assessed clinically using a 10-g Semmes–Weinstein monofilament, vibration perception testing with a 128 Hz tuning fork, ankle reflex testing, and temperature sensation assessment. Peripheral vascular disease was assessed clinically by pedal pulse examination.

Laboratory investigations included complete blood count (CBC), glycated hemoglobin (HbA1c), CRP, liver function tests, kidney function tests, serum albumin, and serum creatinine.

Microbiological Analysis

Wound exudate and pus samples were collected under sterile precautions for microbiological evaluation. Specimens were cultured using standard microbiological techniques, and isolates were identified according to Clinical and Laboratory Standards Institute (CLSI) guidelines (2023). Antibiotic susceptibility testing was performed using the modified Kirby–Bauer disk diffusion method and interpreted according to CLSI criteria. Methicillin resistance in *Staphylococcus aureus* was assessed using the cefoxitin disk diffusion test. Colistin susceptibility was confirmed by broth microdilution.

Statistical Analysis

Descriptive statistics were used to summarize demographic and clinical data. Continuous variables were expressed as mean ± standard deviation (SD), while categorical variables were presented as frequencies and percentages. The chi-square test was used for categorical variables, and the independent *t*-test was applied for comparison of continuous variables between groups. One-way ANOVA was used to compare clinical parameters across age groups. Correlation analysis was performed using Pearson's correlation coefficient. A *p*-value < 0.05 was considered statistically significant.

Results

A total of 40 patients with DFUs were included in the study, comprising 28 males (70%) and 12 females (30%). The majority of patients belonged to the 40–60 years of age group. Mean HbA1c levels were higher in males than females across all age groups and were significantly elevated in the 40–60 years age group ($p < 0.001$), as shown in Table 1.

The mean HbA1c level of the study population was 12.93%, indicating poor glycemic control. Mean CRP and TLC values were elevated, suggesting active inflammation and infection, while lower hemoglobin and serum albumin levels reflected poor nutritional status (Fig. 1).

Male patients demonstrated significantly higher HbA1c levels than females, while hypertension, neuropathy, chronic kidney disease, and peripheral vascular disease were more frequent among males (Table 2).

Microbiological analysis revealed monomicrobial growth in 67.5% ($n = 27$) of cultures, polymicrobial growth in 12.5% ($n = 5$), and no growth in 20% ($n = 8$). Gram-negative organisms predominated (70.3%), followed by Gram-positive organisms (22.2%) and fungi (7.4%). The most common isolates were *Pseudomonas* spp., *Escherichia coli*, *Klebsiella* spp., and *Acinetobacter* spp., while methicillin-resistant *Staphylococcus aureus* (MRSA) was the predominant Gram-positive organism. Polymicrobial infections were more frequent in males (Table 3).

Gram-negative isolates demonstrated high resistance to β-lactams, cephalosporins, and fluoroquinolones, whereas colistin retained activity against most isolates. *Acinetobacter* spp. exhibited the highest multidrug resistance. Among Gram-positive organisms, vancomycin and linezolid showed good activity. *Candida albicans* was isolated in 5% of cases, with resistance to fluconazole observed in all isolates (Table 4).

Higher Wagner grades showed significant associations with HbA1c, CRP, and albumin levels (Table 5).

Debridement was the most commonly performed procedure across Wagner grades. Amputations were observed only in male patients with grade III and grade V ulcers

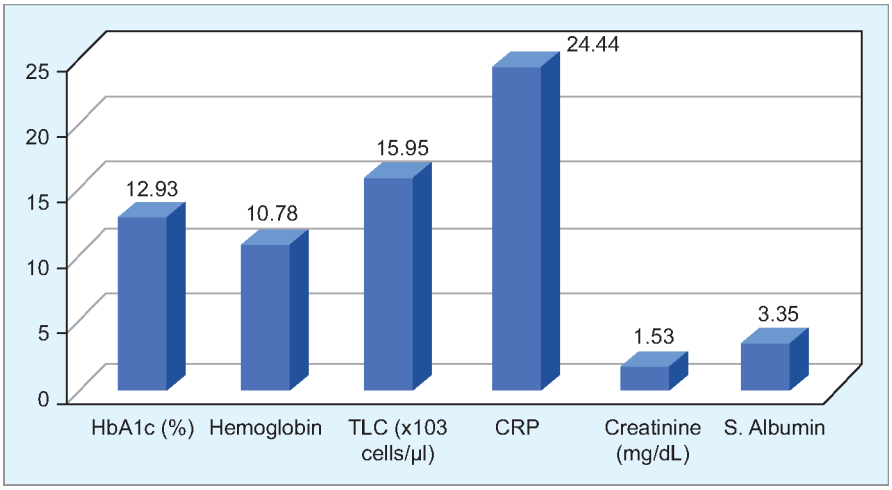

Fig. 1: Mean values of the laboratory parameters

Table 1: HbA1c levels across various age groups for both genders

Age in years	*Male (n = 28)*	*Female (n = 12)*	*p-value*	*HbA1c male*	*HbA1c female*	*p-value*
<40	7 (25%)	3 (25%)	0.99	13.98 ± 1.55	10.53 ± 1.35	0.010
40–60	15 (53.5%)	8 (66.7%)	0.48	13.80 ± 1.42	10.31 ± 1.58	<0.001
>60	6 (21.4%)	1 (8.4%)	0.652	14.48 ± 1.31	11.30	0.075

Table 2: Comparison of clinical variables in males and females

Variables	*Male (n = 28)*	*Female (n = 12)*	*p-value*
HbA1c (mean)	13.99 ± 1.40	10.45 ± 1.41	<0.001
Hypertension	16 (57.2%)	5 (41.7%)	0.369
Neuropathy	13 (46.5%)	5 (41.7%)	0.781
Chronic kidney disease	7 (25%)	1 (8.4%)	0.462
Peripheral vascular disease	4 (14.3%)	0 (0%)	0.168

and were associated with poor glycemic control and polymicrobial infections (Fig. 2).

Discussion

The microbiological profile and antibiotic sensitivity of DFIs vary significantly across regions, largely due to differences in antibiotic use and healthcare practices. Male sex and poor glycemic control have consistently been identified as important risk factors for infected and non-healing DFUs.[11] In our study, male patients demonstrated poorer glycemic control and a higher frequency of severe infections, which is consistent with previous reports. Lacopi et al. also reported that male patients with diabetic foot disease tend to have more severe clinical presentations, higher rates of comorbidities, and increased risks of minor and major amputations.[12] Similar findings have been reported in European DFI studies, where male predominance and uncontrolled diabetes were commonly observed among patients with severe DFIs.[11,13,14]

Bacteriological patterns of DFIs demonstrate considerable regional variation. While Western studies frequently report Gram-positive predominance,[15,16] several Indian studies have documented a higher prevalence of Gram-negative organisms.[17–19] Our findings are consistent with these observations, with Gram-negative bacteria accounting for the majority of isolates. Previous antibiotic exposure and chronic ulceration may contribute to the predominance of Gram-negative organisms in severe DFIs. *Acinetobacter* and *Klebsiella* species, which were commonly isolated in our study, have also been associated with increased risk of major amputations.[20] In addition, severe DFIs in our cohort were predominantly polymicrobial, whereas mild-to-moderate infections were mainly monomicrobial, similar to findings from earlier studies.[21,22] Gram-negative infections, particularly *Pseudomonas aeruginosa*, are associated with greater severity, antibiotic resistance, tissue damage, and higher amputation risk.[23,24]

The emergence of multidrug-resistant organisms (MDROs) in DFIs represents a major therapeutic challenge. Factors such as ischemia, ulcer severity, osteomyelitis, previous antibiotic exposure, and repeated hospitalizations contribute to the development of antimicrobial resistance.[25] In the present study, Gram-negative organisms exhibited high resistance to β-lactams, cephalosporins, and fluoroquinolones, whereas colistin retained good activity against most isolates. Among Gram-positive organisms, vancomycin and linezolid remained highly effective. *Candida* isolates were also

Table 3: Culture results of diabetic foot ulcers (DFUs) ($n = 40$)

Type of growth	*Organism*	*Frequency (%)*	*Male (n = 28)*	*Female (n = 12)*
Monomicrobial	*Acinetobacter*	3 (7.5)	2	1
	Klebsiella pneumoniae	3 (7.5)	2	1
	Pseudomonas	5 (12.5)	3	2
	Citrobacter	2 (5)	1	1
	Candida	2 (5)	2	0
	Enterococcus	2 (5)	2	0
	MRSA	3 (7.5)	2	1
	E. coli	4 (10)	3	1
	MSSA	1 (2.5)	1	0
	Proteus mirabilis	2 (5)	2	0
Polymicrobial	*Acinetobacter* + *Klebsiella*	1 (2.5)	1	0
	Klebsiella spp. + Proteus	1 (2.5)	1	0
	Pseudomonas + *Acinetobacter*	1 (2.5)	1	0
	E. coli + *Pseudomonas*	1 (2.5)	1	0
	E. coli + *Klebsiella*	1 (2.5)	0	1
No growth	–	8 (20)	6	2

Table 4: Major antimicrobial resistance patterns among isolates from diabetic foot infections

Organism	*Predominant Resistance Pattern*	*Antibiotics with Highest Sensitivity*
Acinetobacter spp. ($n = 5$)	High resistance to cephalosporins, fluoroquinolones, and β-lactams	Colistin
Klebsiella spp. ($n = 6$)	Resistant to third-generation cephalosporins and β-lactams	Colistin, meropenem
Proteus spp. ($n = 3$)	High resistance to cephalosporins and β-lactams	Meropenem, colistin
Pseudomonas aeruginosa ($n = 7$)	Resistant to cephalosporins and fluoroquinolones	Colistin
Escherichia coli ($n = 6$)	High resistance to cephalosporins and β-lactams	Colistin, meropenem
Citrobacter spp. ($n = 2$)	Multidrug resistance to β-lactams and cephalosporins	Colistin
MRSA ($n = 3$)	Resistant to oxacillin, levofloxacin, and clindamycin	Vancomycin, linezolid
Enterococcus spp. ($n = 2$)	Resistant to amoxiclav and doxycycline	Vancomycin, linezolid
Candida albicans ($n = 2$)	Resistant to fluconazole	Caspofungin, micafungin, amphotericin B

Table 5: Association between Wagner ulcer grading and selected biochemical and hematological parameters. Values are expressed as mean ± standard deviation (SD) for each Wagner ulcer grade (I–V). *p*-values represent the statistical significance of differences across Wagner grades

Wagner grading	*I*	*II*	*III*	*IV*	*V*	*p-value*
HbA1c (mean ± SD)	10.22 ± 1.12	12.90 ± 0.53	14.27 ± 0.37	15.20 ± 0.07	15.95 ± 0.43	0.001
CRP (mg/L)	16.83 ± 7.25	23.16 ± 17.03	16.68 ± 9.66	14.60 ± 30.92	44.50 ± 31.9	0.026
TLC ($\times 10^3/\mu L$)	10.92 ± 4.40	17.00 ± 10.78	14.71 ± 3.14	21.60 ± 12.06	23.00 ± 8.83	0.057
Albumin (gm/dL)	3.73 ± 0.38	3.21 ± 0.71	3.42 ± 0.73	3.28 ± 0.76	2.57 ± 0.67	0.045
Hemoglobin (gm/dL)	11.42 ± 1.35	10.68 ± 1.85	11.27 ± 1.59	9.92 ± 1.50	9.40 ± 1.90	0.174
Creatinine (mg/dL)	0.96 ± 0.71	1.86 ± 1.09	1.45 ± 0.71	1.64 ± 0.74	2.30 ± 1.51	0.092

HbA1c, glycated hemoglobin; CRP, C-reactive protein; TLC, total leukocyte count; SD, standard deviation

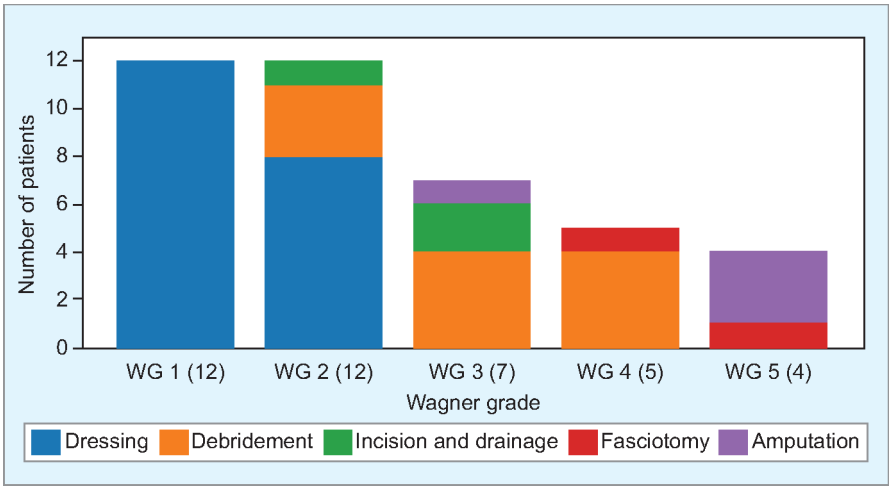

Fig. 2: Treatment modalities according to Wagner grade in patients with diabetic foot ulcers. The stacked bar chart illustrates the distribution of treatment modalities across Wagner grades I-V. Dressing was the predominant treatment in grade I ulcers, whereas debridement was most commonly performed in grades II–IV. Amputations were observed only in grades III and V, reflecting the association between advanced ulcer severity and limb loss

identified, highlighting the possibility of fungal superinfection in chronic DFIs.

Inflammatory and biochemical markers are important indicators of infection severity and prognosis in DFUs. Elevated CRP, TLC, and HbA1c levels in our study correlated with increasing Wagner grades and severity of infection, consistent with previous meta-analyses.[26,27] Poor glycemic control was strongly associated with severe infection and amputation risk. In addition, reduced serum albumin and hemoglobin levels were associated with advanced ulcer severity, reflecting the impact of nutritional status on wound healing and clinical outcomes. Similar associations between hypoalbuminemia, elevated inflammatory markers, and increased amputation risk have been reported previously.[28]

Overall, our findings highlight the predominance of Gram-negative and multidrug-resistant organisms in DFIs in this region and emphasize the importance of early microbiological evaluation, appropriate antimicrobial selection, and optimal glycemic control in reducing ulcer progression and amputation risk.

Limitations

The small sample size and single-center retrospective design limit the generalizability of the findings. Wound swab cultures were used instead of deep tissue biopsy, which may not reliably distinguish colonization from true infection. Anaerobic cultures and prior antibiotic exposure data were unavailable. The limited number of severe cases and amputations also precluded multivariate analysis. Therefore, these findings should be considered preliminary and require validation in larger prospective multicenter studies.

Conclusion

This study demonstrates a predominance of Gram-negative organisms in DFIs, with a notable burden of multidrug-resistant pathogens. Poor glycemic control and abnormal inflammatory and nutritional markers were associated with increasing ulcer severity and adverse outcomes. These findings emphasize the importance of early microbiological evaluation and consideration of broader Gram-negative coverage when initiating empirical therapy in similar clinical settings.

Conflict of Interest

None.

Source of Funding

None.

Authors' Contributions

Conceptualization: ADJ, SKD, MS, RKS; Methodology: ADJ; Software: ADJ; Validation: ADJ, SKD, MS, RKS; Formal analysis: ADJ, SKD; Investigation: ADJ, SKD; Resources: ADJ; Data curation: ADJ, SKD; Writing—original draft preparation: ADJ; Writing—review and editing: ADJ, SKD, MS, RKS; Visualization: ADJ, SKD, MS, RKS; Supervision: ADJ, SKD; Project administration: SKD.

Orcid

Asif Dabeer Jafri https://orcid.org/0009-0008-4498-2583
Srikant Kumar Dhar https://orcid.org/0000-0001-5259-8081
Muktesh Singh https://orcid.org/0009-0003-6439-3771
Ratender Kumar Singh https://orcid.org/0000-0002-0811-5254

References

1. Ramsey SD, Newton K, Blough D, et al. Incidence, outcomes, and cost of foot ulcers in patients with diabetes. Diabetes Care 1999;22(3):382–387.
2. Ledoux WR, Shofer JB, Smith DG, et al. Relationship between foot type, foot deformity, and ulcer occurrence in the high-risk diabetic foot. J Rehabil Res Dev 2005;42(5):665–672.
3. Moulik PK, Mtonga R, Gill GV. Amputation and mortality in new-onset diabetic foot ulcers stratified by etiology. Diabetes Care 2003;26(2):491–494.
4. Walsh JW, Hoffstad OJ, Sullivan MO, et al. Association of diabetic foot ulcer and death in a population-based cohort from the United Kingdom. Diabet Med 2016;33(11):1493–1498.
5. Dang CN, Prasad YD, Boulton AJ, et al. Methicillin-resistant Staphylococcus aureus in the diabetic foot clinic: a worsening problem. Diabet Med 2003;20(2):159–161.
6. Nelson SB. Management of diabetic foot infections in an era of increasing microbial resistance. Curr Infect Dis Rep 2009;11(5):375–382.
7. Frydrych LM, Fattahi F, He K, et al. Diabetes and sepsis: risk, recurrence, and ruination. Front Endocrinol (Lausanne) 2017;8:271.
8. Chastain CA, Klopfenstein N, Serezani CH, et al. A clinical review of diabetic foot infections. Clin Podiatr Med Surg 2019;36(3):381–395.
9. Dmitriyeva M, Kozhakhmetova Z, Urazova S, et al. Inflammatory biomarkers as predictors of infected diabetic foot ulcer. Curr Diabetes Rev 2022;18(6):e280921196867.
10. Lin C, Liu J, Sun H. Risk factors for lower extremity amputation in patients with diabetic foot ulcers: a meta-analysis. PLoS One 2020;15(9):e0239236.
11. Shakil S, Khan AU. Infected foot ulcers in male and female diabetic patients: a clinico-bioinformative study. Ann Clin Microbiol Antimicrob 2010;9:2.
12. Iacopi E, Pieruzzi L, Riitano N, et al. The weakness of the strong sex: differences between men and women affected by diabetic foot disease. Int J Low Extrem Wounds 2023;22(1):19–26.
13. Prompers L, Schaper N, Apelqvist J, et al. Prediction of outcome in individuals with diabetic foot ulcers: focus on the differences between individuals with and without peripheral arterial disease. The EURODIALE Study. Diabetologia 2008;51(5):747–755.
14. Sivanmaliappan TS, Sevanan M. Antimicrobial susceptibility patterns of Pseudomonas aeruginosa from diabetes patients with foot ulcers. Int J Microbiol 2011;2011:605195.
15. Mendes JJ, Marques-Costa A, Vilela C, et al. Clinical and bacteriological survey of diabetic foot infections in Lisbon. Diabetes Res Clin Pract 2012;95(1):153–161.
16. Citron DM, Goldstein EJ, Merriam CV, et al. Bacteriology of moderate-to-severe diabetic foot infections and in vitro activity of antimicrobial agents. J Clin Microbiol 2007;45(9):2819–2828.
17. Elamurugan TP, Jagdish S, Kate V, et al. Role of bone biopsy specimen culture in the management of diabetic foot osteomyelitis. Int J Surg 2011;9(3):214–216.
18. Gadepalli R, Dhawan B, Sreenivas V, et al. A clinico-microbiological study of diabetic foot ulcers in an Indian tertiary care hospital. Diabetes Care 2006;29(8):1727–1732.
19. Bansal E, Garg A, Bhatia S, et al. Spectrum of microbial flora in diabetic foot ulcers. Indian J Pathol Microbiol 2008;51(2):204–208.

20. Cardoso NA, Cisneiros LDL, Machado CJ, et al. Bacterial genus is a risk factor for major amputation in patients with diabetic foot. Rev Col Bras Cir 2017;44(2):147–153.
21. Goldstein EJ, Citron DM, Nesbit CA. Diabetic foot infections: bacteriology and activity of 10 oral antimicrobial agents against bacteria isolated from consecutive cases. Diabetes Care 1996;19(6):638–641.
22. Ramakant P, Verma A, Misra R, et al. Changing microbiological profile of pathogenic bacteria in diabetic foot infections: time for a rethink on which empirical therapy to choose? Diabetologia 2011;54(1):58–64.
23. Sugandhi P, Prasanth DA. Microbiological profile of bacterial pathogens from diabetic foot infections in tertiary care hospitals, Salem. Diabetes Metab Syndr 2014;8(3):129–132.
24. Roberts W, Roessler C, Francis PJ, et al. Post-surgical gangrene with Pseudomonas luteola resulting in limb amputation: a case review. Cureus 2018;10(10):e3441.
25. Dai J, Jiang C, Chen H, et al. Assessment of the risk factors of multidrug-resistant organism infection in adults with type 1 or type 2 diabetes and diabetic foot ulcer. Can J Diabetes 2020;44(4):342–349.
26. Kishore S, Upadhyay AD, Jyotsna VP. Awareness of foot care among patients with diabetes attending a tertiary care hospital. Natl Med J India 2015;28(3):122–125.
27. Wang YT, Zhang LX, Li Y, et al. Inflammatory markers in diabetic foot infection: a meta-analysis. Wounds 2023;35(12):425–432.
28. Sun JH, Tsai JS, Huang CH, et al. Risk factors for lower extremity amputation in diabetic foot disease categorized by Wagner classification. Diabetes Res Clin Pract 2012;95(3):358–363.