

## Article

# Spectral and Antibiotic Susceptibility of Pathogens Isolated from Saudi Patients with Diabetic Foot Infections

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**Abstract:** Diabetic foot infections (DFIs) are a significant health issue and a common complication among patients with diabetes. To develop antibiotic therapy for these high-risk patients, the current study evaluates the scope of DFIs and identifies the causing microbes. It also measures spectrum and antibiotic susceptibility of the pathogens isolated from adults with DFIs in Saudi Arabia. To achieve the study objectives, a cross-sectional study was implemented and the baseline characteristics for 44 patients with DFIs were defined. Optimal aerobic and anaerobic microbiological techniques were utilized to culture specimens isolated from infected foot ulcers. The standard microbiological methods were employed to identify the bacterial isolates and antibiotic susceptibility testing was conducted following the procedures of the Clinical and Laboratory Standards Institute (CLSI). *Results* showed that 12 microorganisms were isolated from the participants' diabetic foot ulcers. *Staphylococcus Aureus* was ranked first because it appeared in 29 (65.9%) cases. *Streptococcus Agalactiae* was ranked second and multi-microbial infections were also found. Most of the organisms were susceptible to Vancomycin, Ciprofloxacin, and Cefalexin, but they were resistant to Methicillin, Gentamicin, and Ampicillin antibiotics. *Staphylococcus Aureus* was most sensitive to Ciprofloxacin, while it was resistant to Methicillin. About 10% of the isolates were multidrug-resistant. The study concludes that while Vancomycin should be used empirically for Gram-positive isolates, Ciprofloxacin can be taken into consideration for most of the Gram-negatives aerobes. Based on including various microorganisms and the advent of multidrug-resistant strains, proper culture and sensitivity testing are necessary prior to the empirical therapy.

**Keywords:** diabetic foot infection; diabetic neuropathy; antimicrobial susceptibility; antibiotic resistance; multidrug-resistant organisms



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## 1. Introduction

Diabetic complications are a serious health concern that affect patients' quality of life because they have to carry a great fiscal burden. Thus, interventions aimed at improving the quality of healthcare or outcomes should be supported. This perquisite a frequent observation of the examination of patients with foot ulcers and isolation of the causative organisms. Further, culture and sensitivity play a role in the selection of the empirical treatment and improvement of the outcome.

There is a high rate of diabetes prevalence in Saudi Arabia, as 13.4% (14.8% males and 11.7% females) of Saudi adults have diabetes, and it increases with aging by 7.8% among adults aged 25–34 years, while it reaches 50.4% among 65+ years old Saudis [1].

According to the International Diabetes Federation, the number of diabetic patients will increase from 240 million in 2007 to 380 million in 2025. About 15% of patients with diabetes suffer from foot ulcers, and they are highly vulnerable to diabetic foot infections (DFIs) [2].

In most cases, early intervention includes a first-hand antimicrobial treatment depending on the local epidemiological data of the antimicrobial susceptibility. Hence, identifying the causative microbe of infection helps prescribe the adequate antibiotic therapy [3].

Patients with diabetes commonly suffer from foot ulcers that have a high rate of cumulative life incidence up to 25%. It infects soft tissue and can cause lower-limb amputation. However, the provision of proper healthcare may prevent diabetic foot ulcer [4,5]. Therefore, early diagnosis of local diabetic foot ulcers and systemic treatment for diabetes are vital [6].

Due to the increasing incidence of diabetes mellitus worldwide, more diabetic patients suffer from its complications, i.e., 15% of them are prone to have foot ulcers with a 2–3% incidence annually. Accordingly, it is important to identify the risk factors making diabetic patients prone to foot ulcer infection in order to help them avoid amputation [7–10].

The infection management requires administering an antibiotic susceptibility test, since it helps provide suitable therapeutic choices and thus improves the outcome. In addition, antibiotic susceptibility patterns illustrate diverse ethnic and geographical subsets of the patients [11–13].

The present study aims to identify the microorganisms causing DFIs and their antibiotic sensitivity among Saudi patients with type 2 diabetes.

## 2. Materials and Methods

The study sample covered 44 Saudi patients with type 2 diabetes and DFIs admitted at King Fahad Medical City in Saudi Arabia, Riyadh.

Following the standards of the WHO, diabetes mellitus was defined as “fasting plasma glucose  $\geq 7.0$  mmol/L (126 mg/dL) or 2-h plasma glucose  $\geq 11.1$  mmol/L (200 mg/dL)” [14].

Before conducting the study, an ethical approval from the institutional review board and a written consent from the participants were obtained after explaining the study objectives for them. Accordingly, the participants’ demographic and clinical features, including sex, age, height, weight BMI, type of diabetes, and wound size were taken in addition to the history of other diabetic complications such as nephropathy retinopathy, CVD, MI, stroke, hypertension, and dyslipidemia, HbA1C, lipid profile, and kidney profile as well as the foot ulcer duration record.

### 2.1. Isolation, Culture, and Sensitivity of the Microorganisms

#### 2.1.1. Bacterial Isolation

Two swabs per patient were taken for the culture of the causative organisms. Adopting the deep swab technique, the wound was debrided, and deep swabs were obtained. After transferring the swabs into sterile tubes, they were immediately transported to the microbiology laboratory for further culture and sensitivity. The isolates were identified according to the standard methods.

#### 2.1.2. Antibiotic Sensitivity

Susceptibility of the microorganisms obtained from the isolates from diabetic patients with foot ulcers was subjected to an extremely wide range of different antibiotics exposure using disc diffusion methods according to the CLSI [15].

Antibiotics tested were Trimethoprim, Rifampicin, Mupirocin, Amikacin, Gentamicin, Imipenem, Ceftriaxone, Ceftizoxime, Fusidic Acid, Nitrofurantoin, Fosfomycin, Tigecycline, Tetracycline, Minocycline, Doxycycline, Vancomycin, Teicoplanin, Linezolid, Clindamycin, Erythromycin, Roxithromycin, Clarithromycin, Azithromycin, Sparfloxacin, Pefloxacin, Ofloxacin, Norfloxacin, Moxifloxacin, Levofloxacin, Ciprofloxacin, Tobramycin, Ceftazidime, Cefotaxime, Cefoperazone, Cefixime, Cefdinir, Cefoxitin, Cefuroxime, Cefamandole, Cefradine, Cefalexin, Cefadroxil, Cefaclor, Oxacillin, Methicillin, Flucloxacillin, Piperacillin/Tazobactam, Piperacillin, Ticarcillin/Clavulanic Acid, Amoxicillin/Clavulanic Acid, mpicillin, Amoxicillin, Benzylpenicillin, and Flomoxef.

The author inoculated pus samples on blood agar, chocolate agar, Sabouraud Dextrose agar, and Maconkey agar plates and incubated them at 37 °C overnight [15]. The growth was identified according to colony morphology, Gram stain, and appropriate biochemical

tests. Antibiotic susceptibility tests were implemented utilizing Kirby Bauer disk diffusion method on Mueller Hinton agar plates [16].

## 2.2. Statistical Analysis

Utilizing SPSS version 17, data were analyzed and presented as mean  $\pm$  SD for continuous variables and frequency with percentages for categorical variables.

## 3. Results

Table 1 shows the participants' demographic characteristics: age was  $59.4 \pm 7.6$ , and there were 23 (52.2%) male and 21 (47.7%) females. The duration of the diabetes was  $14.6 \pm 3$  years.

**Table 1.** Baseline characteristics of the study population.

Variables	Mean $\pm$ SD
Males	23 (52.2%)
Females	21 (47.7%)
Age (y)	$59.4 \pm 7.6$
HT (cm)	$169.5 \pm 9.3$
WT (kg)	$85.7 \pm 27$
BMI (kg/m <sup>2</sup> )	$29.7 \pm 7.7$
BP_SY (mmHg)	$143.5 \pm 20.9$
BP_DIAS (mmHg)	$92.8 \pm 23.9$
Duration Of D.M	$14.6 \pm 3$ y
Hypertensive	26 (59.1%)
Duration of foot infection	$33 \pm 6$ days
H/O Foot Ulcer in Past	16 (40.9%)
Empirical therapy received	12 (27.2%)
Family history of foot infection	9 (20.4%)

According to Table 2, more than 75 % of patients (16 males and 18 females) were obese (BMI  $\geq$  25 kg/m<sup>2</sup>). The co-morbidities were CVD 36 (81.8%), retinopathy 6 (13.6%), and hypertension 26 (59.1%). Further, 10 patients (22.7%) had a foot ulcer history.

**Table 2.** Baseline comorbidities in the study population.

Comorbidities	No. %
HTN	26 (59.2%)
Neuropathy	23 (52.2%)
Retinopathy	6 (13.6%)
Nephropathy	12 (27.2%)
Dyslipidemia	24 (54.5%)
CVD	36 (81.8%)
MI	1 (2.27%)
CVA	1 (2.27%)
CRF	7 (15.9%)

The culture illustrated polymicrobial growth in six (13.6%) out of the 44 specimens of the diabetic foot lesions. Moreover, the prevalence of Gram-negative organisms 28 (63.6%) was higher than Gram-positive organisms 16(36.3%). Most of the organisms were susceptible to Vancomycin, Ciprofloxacin, and Cefalexin, but they were resistant to Methicillin, Gentamicin, and Ampicillin antibiotics. Staphylococcus Aureus was most sensitive to Ciprofloxacin, while it was resistant to Methicillin. About 10 % of the isolates were multidrug-resistant. Staphylococcus aureus was the most frequent pathogen 29 (65.9%). No bacterial growth was found in the specimens of six patients (13.6%).

#### 4. Discussion

Diabetic foot ulceration is a common complication that occurs due to uncontrolled diabetes. Reports reveal that there is a high rate of diabetes prevalence in Saudi Arabia, which increases with age, i.e., 50.4% in persons aged 65+ years [1].

The present study revealed that males are at a higher risk for infected foot ulcers. This result matches the findings of other studies [3].

In the present study, polymicrobial infection was common, which is consistent with other studies asserting that DFIs are mostly Polymicrobial [3,17].

The study found out that *S. aureus* was a common bacterial pathogen isolated from 29 foot ulcers of diabetic patient specimens. Other studies also reported a more frequent infection by *S. aureus* [18].

Contrary to the present study, some studies found out that the most frequent pathogens isolated from diabetic foot were *E. coli* and *P. aeruginosa* [19–21].

Globally, antimicrobial resistance has become an increasing setback while treating the diabetic foot [22].

The results of culture and antimicrobial susceptibility can be utilized by providing an adequate management of DFIs and thus decreasing the rates of morbidity and amputation and improving the outcome [23].

The present study reveals the prevalence of Gram-positive isolates among Saudis with DFIs. This result agrees with the findings of other studies [24].

Matching other studies, the present study showed that all Gram-positive isolates were sensitive to Vancomycin [25,26].

Moreover, the study showed that *S. aureus* isolated was susceptible to Ciprofloxacin, while it was resistant to Methicillin. These antibiotics are highly effective against Gram-positive Cocci isolated in the current study, and they fit as an initial therapy for DFIs.

The findings showed that vancomycin is the gold standard for the majority of DFIs.

#### 5. Conclusions

Diabetic complications and comorbidities are quite prevalent in the Saudi population. The study's findings illustrated that DFIs are most frequently affected by *S. aureus*. DFIs are often polymicrobial in most cases associated with *S. aureus*, *E. coli*, and *Enterococcus* spp. Microbial profile diversity and resistance to antibiotics highlight the need to have culture specimens from infected ulcers to carry out microbial assessment and antibiotic susceptibility testing.

To improve the outcome, strict glycemic control and frequent surveillance for early detection of diabetic complications and infected feet ulcers are highly recommended.

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**Data Availability Statement:** The data presented in this study available on request from the corresponding author.

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**Conflicts of Interest:** The author declares no conflict of interest.

#### References

1. Saudi Arabia, Ministry of Health. 2015. Available online: <https://www.moh.gov.sa/en/HealthAwareness/healthDay/2015/Pages/HealthDay-2015-11-14.aspx> (accessed on 14 November 2020).
2. Bansal, E.; Garg, A.; Bhatia, S.; Attri, A.K.; Chander, J. Spectrum of microbial flora in diabetic foot ulcers. *Indian J. Pathol. Microbiol.* **2008**, *51*, 204–208. [PubMed]
3. Gadepalli, R.; Dhawan, B.; Sreenivas, V.; Kapil, A.; Ammini, A.C.; Chaudhry, R. A Clinico-microbiological Study of Diabetic Foot Ulcers in an Indian Tertiary Care Hospital. *Diabetes Care* **2006**, *29*, 1727–1732. [CrossRef] [PubMed]

4. Singh, N.; Armstrong, D.G.; Lipsky, B.A. Preventing Foot Ulcers in Patients with Diabetes. *JAMA* **2005**, *293*, 217–228. [[CrossRef](#)] [[PubMed](#)]
5. Richard, J.-L.; Sotto, A.; Lavigne, J.-P. New insights in diabetic foot infection. *World J. Diabetes* **2011**, *2*, 24–32. [[CrossRef](#)] [[PubMed](#)]
6. Lavery, L.A.; Armstrong, D.G.; Wunderlich, R.P.; Mohler, M.J.; Wendel, C.S.; Lipsky, B.A. Risk Factors for Foot Infections in Individuals with Diabetes. *Diabetes Care* **2006**, *29*, 1288–1293. [[CrossRef](#)] [[PubMed](#)]
7. Vileikyte, L. Diabetic foot ulcers: A quality of life issue. *Diabetes Metab. Res. Rev.* **2001**, *17*, 246–249. [[CrossRef](#)]
8. Shailesh, K.; Ashok, K.; Surya, K.; Sanjeev, K. Prevalence of diabetic foot ulcer and associated risk factors in diabetic patients from north India. *JDFC* **2012**, *4*, 83–91.
9. Snyder, R.J.; Hanft, J.R. Diabetic foot ulcers—Effects on QOL, costs, and mortality and the role of standard wound care and advanced-care therapies. *Ostomy Wound Manag.* **2009**, *55*, 28–38.
10. Won, S.H.; Chung, C.Y.; Park, M.S.; Lee, T.; Sung, K.H.; Lee, S.Y.; Kim, T.G.; Lee, K.M. Risk Factors Associated with Amputation-Free Survival in Patient with Diabetic Foot Ulcers. *Yonsei Med. J.* **2014**, *55*, 1373–1378. [[CrossRef](#)]
11. Ramakant, P.; Verma, A.K.; Misra, R.; Prasad, K.N.; Chand, G.; Mishra, A.; Agarwal, G.; Mishra, S.K. Changing microbiological profile of pathogenic bacteria in diabetic foot infections: Time for a rethink on which empirical therapy to choose? *Diabetologia* **2010**, *54*, 58–64. [[CrossRef](#)]
12. Mathangi, T.; Prabhakaran, P. Prevalence of bacteria isolated from type 2 diabetic foot ulcers and the antibiotic susceptibility pattern. *Int. J. Curr. Microbiol. Appl. Sci.* **2013**, *2*, 329–337.
13. Turhan, V.; Mutluoglu, M.; Acar, A.; Hatipoglu, M.; Onem, Y.; Uzun, G.; Ay, H.; Oncul, O.; Gorenek, L. Increasing incidence of Gram-negative organisms in bacterial agents isolated from diabetic foot ulcers. *J. Infect. Dev. Ctries.* **2013**, *7*, 707–712. [[CrossRef](#)] [[PubMed](#)]
14. WHO; IDF. *Definition and Diagnosis of Diabetes Mellitus and Intermediate Hyperglycaemia*; Report of WHO/IDF Consultation; WHO Press: Geneva, Switzerland, 2006.
15. Raja, N.S. Microbiology of diabetic foot infections in a teaching hospital in Malaysia: A retrospective study of 194 cases. *J. Microbiol. Immunol. Infect.* **2007**, *40*, 39–44. [[PubMed](#)]
16. Saeed, N.K.; Kambal, A.M.; El-Khizzi, N.A. Antimicrobial-resistant bacteria in a general intensive care unit in Saudi Arabia. *Saudi Med. J.* **2010**, *31*, 1341–1349. [[PubMed](#)]
17. Tiwari, S.; Pratyush, D.D.; Dwivedi, A.; Gupta, S.K.; Rai, M.; Singh, S.K. Microbiological and clinical characteristics of diabetic foot infections in northern India. *J. Infect. Dev. Ctries.* **2012**, *6*, 329–332. [[CrossRef](#)]
18. El-Tahawy, A.T. Bacteriology of diabetic foot. *Saudi Med. J.* **2000**, *21*, 344–347.
19. Shankar, E.; Mohan, V.; Premalatha, G.; Srinivasan, R.; Usha, A. Bacterial etiology of diabetic foot infections in South India. *Eur. J. Intern. Med.* **2005**, *16*, 567–570. [[CrossRef](#)]
20. Hadadi, A.; Ghiasi, H.O.; Hajiabdolbaghi, M.; Zandekarimi, M.; Hamidian, R. Diabetic Foot: Infections and Outcomes in Iranian Admitted Patients. *Jundishapur J. Microbiol.* **2014**, *7*, e11680. [[CrossRef](#)]
21. Cabete, J.; De Carvalho, F.M.; Moniz, L.; Pinto, M.; Neves, J.; Alves, C.P. Microbiological profile and antibiotic susceptibility patterns of organisms isolated from diabetic foot ulcers in a Portuguese hospital. *Rev. Port. Cir Cardiorac. Vasc.* **2011**, *18*, 53–60.
22. Wang, S.-H.; Sun, Z.-L.; Guo, Y.-J.; Yang, B.-Q.; Yuan, Y.; Wei, Q.; Ye, K.-P. Meticillin-resistant *Staphylococcus aureus* isolated from foot ulcers in diabetic patients in a Chinese care hospital: Risk factors for infection and prevalence. *J. Med. Microbiol.* **2010**, *59*, 1219–1224. [[CrossRef](#)]
23. Shanmugam, P.; Jeya, M.; Susan, L.S. The Bacteriology of Diabetic Foot Ulcers, with a Special Reference to Multidrug Resistant Strains. *J. Clin. Diagn. Res.* **2013**, *7*, 441–445. [[CrossRef](#)] [[PubMed](#)]
24. Tascini, C.; Piaggese, A.; Tagliaferri, E.; Iacopi, E.; Fondelli, S.; Tedeschi, A.; Rizzo, L.; Leonildi, A.; Menichetti, F. Microbiology at first visit of moderate-to-severe diabetic foot infection with antimicrobial activity and a survey of quinolone monotherapy. *Diabetes Res. Clin. Pract.* **2011**, *94*, 133–139. [[CrossRef](#)] [[PubMed](#)]
25. Rani, K.L.; Madhumathi, B.S.; Ramana, B.V. Bacteriological profile of diabetic foot ulcer. *IJPRBS* **2013**, *2*, 36–45.
26. Sugandhi, P.; Prasanth, D.A. Microbiological profile of bacterial pathogens from diabetic foot infections in tertiary care hospitals, Salem. *Diabetes Metab. Syndr. Clin. Res. Rev.* **2014**, *8*, 129–132. [[CrossRef](#)] [[PubMed](#)]