Al-Mukhtar Journal of Basic Sciences 24 (2): 119-129, 2026
Doi: https://doi.org/10.54172/48rnqy36

Research Article
Open Access

Bacterial Pathogens in Diabetic Foot Infections at the Diabetes Center of Al-Bayda: Phenotypic Identification Using the Render MA 120 System

Latifa Issa [1], Nagah S.A Abubaker [2*]

[1] Department of Life Sciences, Microbiology, the Libyan Academy, Al Jabal Al-Akhdar, Libya.

***Corresponding author:** nagah.abubaker@omu.edu.ly
Department of Botany, Faculty of Science, Omar Al-Mukhtar University, Libya.

Received:
21 May 2026

Accepted:
21 July 2026

Publish online:
31 August 2026

Abstract

Diabetic foot infection (DFI) is a major clinical complication of diabetes mellitus, serving as a primary precursor to lower-limb amputation. This study aimed to identify and phenotypically characterize the bacterial pathogens associated with DFIs at the Diabetes Center in Al-Bayda, Libya. Samples were collected from infected wounds, tissues, and nails of 14 patients and analyzed using the Render MA 120 automated identification system. Results showed a high prevalence of Gram-negative bacteria (90%), with Klebsiella pneumoniae as the most frequent isolate (25.5%), followed by *Escherichia coli* (17.65%). Notably, rare opportunistic pathogens, including *Burkholderia pseudomallei* (1.96%) and *Raoultella ornithinolytica* (1.96%), were identified. These findings highlight a significant regional shift toward Gram-negative dominance and underscore the necessity for precise diagnostic tools and targeted empirical antibiotic protocols to prevent limb loss.

Keywords: Diabetic Foot, Libya, *Klebsiella Pneumoniae*, Gram-Negative Bacteria, Render MA 120, Opportunistic Pathogens.

INTRODUCTION

Diabetic foot infections (DFIs) represent a multifaceted global health challenge, and they are recognized as a leading cause of non-traumatic lower-extremity amputations, prolonged hospitalizations, and significantly increased mortality (Peters et al., 2020). The pathophysiology of DFIs involves a complex interplay between peripheral neuropathy, which results in a loss of protective sensation, and peripheral vascular disease, which impairs the delivery of immune cells and antibiotics to the site of injury (Lipsky et al., 2020),(Frykberg et al., 2000). If not identified and managed promptly, these infections can progress rapidly from superficial ulcerations to deep-seated tissue necrosis and osteomyelitis (Boyanova and Mitov, 2013 ; Thanganadar ,et al., 2021).

In Libya, the burden of diabetes mellitus is particularly acute. Epidemiological reports indicate that more than 70% of diabetic patients in the country are diagnosed with Type II diabetes, representing the highest prevalence in the North African and Arab regions (Eltobgi, 2009). This high prevalence directly translates to an increased incidence of DFIs, placing immense pressure on tertiary care facilities. Recent surveys in major Libyan cities such as Tripoli, Benghazi, and Misrata have documented a distinct shift in the microbial landscape of these infections. While Western European and North American cohorts are typically dominated by Gram-positive cocci like *S. aureus*, studies in warmer climates including Libya show a marked dominance of Gram-negative bacilli, which are found in approximately 70.9% of cases (Eldukali & Boshaalla, 2022; Mendes & Neves, 2012).

(Nuryastuti et al., 2018). In low-resource settings, the reliance on traditional manual biochemical tests can lead to the misidentification of fastidious or emerging pathogens, such as *R. ornithinolytca* or the *B. cepacia* complex (Paes Leme et al., 2022; Solak et al., 2011). Consequently, there is an urgent need for the integration of automated diagnostic systems that provide rapid and accurate species-level identification (Jones, et al., 2021).

Studies of bacteria isolated from diabetic foot in Libya are few and were mainly carried out in the major cities in Libya, including Tripoli and Misrata (Eshlak, et al., 2022), and Benghazi (Eldukali and Boshaalla, 2022), as well as Al-Bayda (Altabouli, et al., 2024).

Study Objectives

The primary objectives of this research were:

1. To identify the spectrum of bacterial pathogens associated with diabetic foot infections among patients at the Diabetes Center of Al-Bayda, Libya.
2. To phenotypically characterize the isolates using automated diagnostic technology (Render MA 120) to ensure high diagnostic precision.
3. To document the prevalence of rare or emerging opportunistic pathogens within the local diabetic population.

MATERIALS AND METHODS

Sample collection and site

This study was carried out between October and November 2024 in the surgical department at the Diabetes Center, which operates as the only outpatient facility for diabetes management in El-Beyda, with a sample population of 14 Libyan patients diagnosed with type 2 diabetes who presented with diabetic foot complications, consisting of 8 females and 6 males. The diabetes diagnosis was based on the criteria set by the World Health Organization (WHO). The study assessed key patient and clinical characteristics, including age, sex, duration of diabetes, prescribed medications, and the classification of the diabetic foot ulcer (DFU) (Eldukali and Boshaalla, 2022).

Wound site preparation and sample collection

Before commencing the procedure, it is essential to thoroughly wash hands and don sterile gloves. The next step involves cleaning the wound using sterile saline or an antiseptic solution, deliberately avoiding alcohol or hydrogen peroxide, as these agents can harm healthy tissue. Subsequently, necrotic tissue or foreign materials present in the wound should be debrided. This can be accomplished using sterile forceps or scissors, ensuring that samples are taken from viable tissue or areas susceptible to infection. A thorough inspection of the wound is necessary to identify any signs of infection, such as increased redness, warmth, swelling, discharge, or an unpleasant odor, prior to sample collection (Haesler, 2014).

For swab collection at the wound site, two methods were employed: the swab technique and tissue sampling, according to Daivasikamani (2015).

Nail clippings or scrapings should be taken from beneath the nail plate, or nail samples should be taken from the most proximal edge of the diseased nail (Chadwick and Podiatrist, 2013). The advancing edge is too deep to cut back; drilling a hole in the nail and curetting it out can yield better results (Sumikawa, *et al*., 2007). The nail is then placed in a sterile container before being promptly sent to the laboratory. The collected wound swabs and tissue samples, along with nail samples, should be cultured directly on nutrient agar for bacterial isolation (Chadwick and Podiatrist, 2013).

Specimen processing:
The samples (collected with the help of sterile swabs from the wound, tissue, and nails) for bacterial isolation were directly inoculated on nutrient agar and incubated at 35–37°C for 24–48 hours (Sanjith Saseedharan, *et al*., 2018).

Identification of Bacteria by Biochemical Tests and the Render Automated Microbiology System
The sample is inoculated onto nutrient agar and subsequently subcultured onto various agar plates, such as blood agar and MacConkey agar. These plates are then incubated at suitable temperatures, typically ranging from 35 to 37°C, for a duration of 24 to 48 hours. During this time, bacterial growth occurs, resulting in the formation of colonies. These colonies are analyzed based on their morphological characteristics (Kean et al., 2017).

Biochemical identification of bacteria
Once bacterial colonies have grown on culture media, further tests, including catalase, oxidase, and urease tests, are used to identify the species based on their biochemical properties. This is a crucial step in the identification process (Kean, *etal*., 2017). The isolated bacteria were then identified by the Render MA 120 system.

RESULTS

A total of 51 bacterial isolates were recovered from the 14 patients. The results showed an overwhelming dominance of Gram-negative bacteria (90%) compared to Gram-positive species, which were represented only by *Bacillus spp.* (10%). P. aeruginosa, B. cepacia, and B. gladioli exhibited positive results in the oxidase test, while Gram-negative isolates, including K. ornithinolytica, K. pneumoniae, E. coli, K. oxytoca, P. mirabilis, P. aeruginosa, S. maltophilia, A. baumanni, B. pseudomallei, and K. ozeanea, exhibited negative results in the oxidase test.

Most Gram-negative bacterial isolates, including K. pneumoniae, E. coli, K. oxytoca, P. mirabilis, P. aeruginosa, S. maltophilia, K. ozeanea, A. baumanni, and K. ornithinolytica, as well as the Gram-positive Bacillus spp., were positive for catalase. However, B. pseudomallei was negative for catalase (MacFaddin, 2000). The bacteria that demonstrated positive urease activity were K. pneumoniae, K. oxytoca, P. mirabilis, B. pseudomallei, K. ozeanea, and K. ornithinolytica, indicating their ability to hydrolyze urea into ammonia and carbon dioxide (Hadden et al., 2016). In contrast, B. cepacia, B. gladioli, P. aeruginosa, E. coli, A. baumannii, and S. maltophilia demonstrated negative urease activity. Additionally, urease activity remains unknown in Bacillus spp.

Taxonomic Distribution
The most prevalent pathogen was *K. pneumoniae* (25.5%), followed by *E. coli* (17.65%). Other members of the Enterobacteriaceae family and several non-fermenting Gram-negative bacilli were also frequently isolated. The distribution and biochemical profiles of these isolates are summarized in Table 1.

Table (1): Distribution and Clinical Profile of Bacterial Isolates

Taxonomic Group	Bacterial Species	Frequency (%)	No. of Isolates	Clinical/Pathogenic Role	Reference
Enterobacteriaceae	*K. pneumoniae*	25.5%	13/51	Primary opportunistic pathogen; biofilm producer.	(Sugumar, et al., 2014).
	E. coli	17.65%	9/51	Associated with environmental contamination.	(Johnson, 1991).
	K. oxytoca	9.80%	5/51	Emerging nosocomial pathogen. opportunistic pathogen	(Neog, et al., 2021)
	P. mirabilis	3.92%	2/51	Linked to deep tissue and bone involvement. Opportunistic pathogen	(Yuan, et al., 2021)
	K. ozaenae	1.96%	1/51	Rare respiratory-associated subspecies. Opportunistic pathogen	(Podschun and Ullmann, 1998(.
	R ornithinolytca	1.96%	1/51	Emerging histamine-producing pathogen. Opportunistic pathogen	(Solak, et al., 2011),
Non-Fermenting GNB	*A. baumannii*	9.80%	5/51	Highly resistant; risk factor for treatment failure. Opportunistic pathogen	(Lucidi, et al., 2024(.
	B. cepacia	7.85%	4/51	Rare in DFIs; associated with high mortality. Opportunistic pathogen	(Saeed, et al., 2024(.
	P. aeruginosa	3.92%	2/51	Common in warm climates; biofilm-rich. Opportunistic pathogen	(Ertugrul, et al., 2018).
	S. maltophilia	3.92%	2/51	Linked to a 4.76x increased amputation risk. Opportunistic pathogen	(Bhaumik, et al., 2024)
	B. pseudomallei	1.96%	1/51	Causative agent of melioidosis (rare). Opportunistic pathogen	(Ertugrul, et al., 2018).
	B. gladioli	1.96%	1/51	Opportunistic pathogen.	(Lee, et al., 2016)
Gram-Positive	*Bacillus spp.*	9.80%	5/51	Opportunistic environmental flora.	(Baindara and Aslam, 2023(.

DISCUSSION

The absolute dominance of Gram-negative bacteria (90.2%) in the Al-Bayda cohort is consistent with recent findings in Benghazi (70.9%) but represents a significantly higher frequency than global averages (Eldukali and Boshaalla, 2022). This pattern supports the hypothesis that warm climates and localized sanitation factors favor Gram-negative pathogens in chronic diabetic wounds (Mendes and Neves, 2012),(Jones *et al*., 2021).

Foot ulceration constitutes one of the most severe complications affecting individuals with diabetes and extends beyond superficial subcutaneous tissues. The development of diabetic foot ulcers (DFU) is primarily attributed to poor glycemic control and insufficient self-management behaviors (Mendes and Neves, 2012). Inadequate hygiene practices may contribute to the progression of necrotic wounds and gangrene, which can ultimately necessitate amputation. The likelihood of toe or lower-limb amputation is significantly elevated when ulcerations become complicated by bacterial or fungal infections (Heald, *et al*., 2001).

With an increasing diabetic population worldwide, there is a significant rise in the prevalence of foot infections. Al-Bayda also has an increased number of diabetic individuals, and this study characterized 14 patients with diabetic foot ulcers. All patients had type II diabetes mellitus, and 5 patients had previous toe amputations. Some of them exhibited superficial ulcers classified as Wagner grade 3. In contrast, more severe cases involved deep ulcers with complications like abscesses and bone involvement, classified as Wagner grades 2 and 3. These grades also had the highest number of isolates, consistent with findings from previous studies (González, *et al*., 2003). The results of this study indicated that the clinical manifestations of diabetic foot disease included delayed healing, delicate tissue, foul-smelling discharge, pain, and discomfort. Additionally, the presence of abnormal granulation tissue was observed, characterized by a color change from red to gray, along with increased fragility and bleeding upon contact. These results correlate with other studies (Cutting and Harding, 1994).

The higher incidence of superficial ulcers in these findings suggests that early intervention and effective diabetes management are essential in preventing the progression to more severe ulcer types. Nonetheless, the occurrence of deep ulcers with abscesses in several patients emphasizes the necessity for prompt surgical intervention and comprehensive treatment strategies to reduce the risk of amputation (Pickwell, *et al*., 2015). A microbiological study of foot ulcer samples from diabetic patients was performed using conventional microbiological and biochemical identification methods, and different bacterial species were identified. Chronic, non-healing foot ulcers frequently occur in diabetic patients as a result of underlying factors such as neuropathy and increased plantar pressure. These persistent ulcers were particularly vulnerable to infection, which exacerbates the impairment of the healing process. A wide variety of bacterial pathogens may contribute to these infections (Frykberg, *et al*., 2000).

The study results among the 14 patients diagnosed with type 2 diabetes, aged between 24 and 83 years, indicated that the majority of bacterial isolates from diabetic foot infections were Gram-negative, with eight isolates being Gram-positive, a finding similar to that reported (Turhan, *et al*, 2013). *K. pneumoniae* was predominantly isolated, followed by *E. coli* and *P. aeruginosa within the Enterobacteriaceae family* (Shaya, *et al*., 2006). Gram-negative bacteria were isolated more frequently than Gram-positive bacteria. Numerous prior studies have similarly identified members of the *Enterobacteriaceae* family as the predominant isolates in diabetic foot ulcers (DFU) (Thanganadar, *et al*., 2021). The results aligned with previous studies, indicating that Gram-negative bacteria were more frequently encountered than Gram-positive bacteria (Dwedar, *et al*., 2015). Similarly, E. coli, P. aeruginosa, K. pneumoniae, and Proteus species were among the most commonly isolated (Mamusavu, *et al*., 2022).

Other studies similar to this study reported diabetic foot infections (DFIs) resulting from Gram-negative *E. coli, K. pneumoniae and P. aeruginosa* (Slovenkai, 1998). Also, the main bacterial species identified were *Pseudomonas species and E. coli* (Atlaw, *et al*., 2022). In this study, a high incidence of E. coli isolated from dry superficial foot ulcers was observed, which aligns with the findings of Shaya et al. (2006).

Furthermore, the results revealed the isolation of *P. mirabilis* alongside other bacteria, including *K. pneumoniae, K. ozaenae, P. aeruginosa, K. oxytoca, Stenotrophomonas species*, and *E. coli*. Additionally, Gram-positive bacteria such as *Bacillus species were identified among the predominant microbial flora in the analyzed samples, which aligns with results from another study* (Thanganadar , *et al*., 2021). In contrast to recent studies that documented Gram-positive bacteria, S. aureus was identified as the predominant pathogen (Lipsky, *et al*., 2020).

This study found that diabetic foot infections are complex and often involve common bacteria such as K. pneumoniae, E. coli, and P. aeruginosa. In addition, studies have started to show that less common bacteria and fungi can also be important in these infections. Some of these bacteria include

B. pseudomallei, *S. maltophilia*, *B. cepacia*, *K. ozeanea*, *B. gladioli*, and *K. ornithinolytica*. In this study, Gram-negative bacteria were predominantly isolated compared to Gram-positive bacteria, a finding that aligns with results from other studies (Dwedar, *et al*., 2015). However, another study reported results contrary to the current findings, with Gram-positive organisms being the most common (Abdulrazak, *et al*., 2005).

In the present study, *R. ornithinolytica* was infrequently isolated, consistent with previous reports indicating that *R. ornithinolytica* was a rare pathogen in diabetic foot lesions (Pulian Morais, *et al*., 2009). In addition, B. cepacia complex (BCC) was infrequently isolated in this study. BCC comprises a group of widely distributed Gram-negative bacteria. Previous research has documented BCC species as common pathogens in patients with cystic fibrosis (CF), but they are less frequently reported in diabetic foot infections, consistent with the findings of the present study (Paes Leme, *et al*., 2022). Additionally, B. gladioli was less frequently isolated in this study; these results correlate with other studies, in which only a few patients with abscesses caused by B. gladioli infections have been reported (Wang , *et al*., 2022).

Previous studies consistent with these findings have documented the isolation of *S. maltophilia* in diabetic foot infections. Although reported infrequently, *S. maltophilia* may act as an opportunistic pathogen. These observations highlight the need to consider this bacterium as a significant factor in the management of complex infections in diabetic patients (Huchital, *et al*., 2020). In this study, a strain of B. pseudomallei was identified in a diabetic patient; this rare finding aligns with these studies (Nuryastuti, *et al*., 2018).

Significance of Emerging Pathogens

The identification of *R. ornithinolytica* (formerly *K. ornithinolytica*) highlights the utility of the Render system. This organism is an emerging pathogen in patients with comorbidities such as chronic kidney disease and diabetes (Solak ,*et al*., 2011). Its ability to produce histamine may complicate clinical symptoms with allergy-like rashes (Pulian Morais, *et al*., 2009). Similarly, the detection of S. maltophilia is clinically alarming. Recent literature indicates that its presence in limb-threatening ulcers is a significant predictor of major above-ankle amputation (OR: 4.76) (Hung, *et al*., 2023).

Rarity and Biosafety: *B. pseudomallei*

Based on the results obtained from the Render System, *B. pseudomallei accounted for 1.96%, which is a rare and critical clinical finding for Libya.* This agent causes melioidosis, a life-threatening disease typically endemic to Southeast Asia (Tin Som *et al*., 2024). Given its potential for laboratory-acquired infections through aerosolization, these isolates must be handled under Biosafety Level 3 (BSL-3) conditions, and plates should never be "sniffed" for identification (CDC, 2022).

CONCLUSION

This study demonstrates that diabetic foot infections in Al-Bayda are characterized by a complex, Gram-negative-dominated polymicrobial flora. The presence of rare pathogens like *B. pseudomallei* and *R. ornithinolytica* underscores the necessity of automated diagnostic platforms in Al-Bayda city to ensure accurate species differentiation. Bacterial infections in diabetic patients, if not detected and treated in time, can lead to fatal complications such as foot amputation. Adequate management of diabetic foot treatment is needed to reduce the risk of amputation.

LIMITATIONS

Several limitations should be acknowledged in this study. The relatively small sample size was mainly attributed to the limited recruitment period. Moreover, anaerobic pathogens were not assessed, and bacterial identification was not confirmed by molecular methods.

RECOMMENDATIONS:

1. Empirical Guidelines: Local clinical protocols should prioritize Gram-negative coverage for DFI management.
2. Susceptibility Testing: Routine AST is mandatory for every case due to the prevalence of inherently resistant species such as A. baumannii and *S. maltophilia*.

Biosafety Vigilance: Enhanced safety training is required for laboratory personnel to recognize and handle high-risk pathogens.

ACKNOWLEDGEMENT

The authors wish to express their gratitude to the first international laboratory for providing the necessary facilities to conduct this study.

ETHICS

Bioethics reference number: NBC:004.H.25.2**.**

Duality of interest: The authors declare that they have no duality of interest associated with this manuscript.

Author contributions: The authors contributed equally.

Funding: No specific funding was received for this work.

REFERENCES

Abu-Darwish, M. S., Al-Ramannneh, E. A. D. M., Kyslychenko, K. V. S., & Karpiuk, U. V. (2012). The antimicrobial activity of essential oils and extracts of some medicinal plants grown in the south of Jordan. *Pakistan Journal of Pharmaceutical Sciences, 25*(1), 239–246.

Abdulrazak, A., Bitar, Z. I., Al-Shamali, A. A., and Mobasher, L. A. (2005). Bacteriological study of diabetic foot infections. *Journal of Diabetes and its Complications*, *19*(3), 138-141.

Altabouli, S., Qowaider, S., Hamad, M., and Ali, M. (2024). Microbiological profile and antimicrobial susceptibility patterns in patients with tinea pedis at diabetic Center in El-Beyda City. *AlQalam Journal of Medical and Applied Sciences*, 50-55.

Atlaw, A., Kebede, H. B., Abdela, A. A., and Woldeamanuel, Y. (2022). Bacterial isolates from diabetic foot ulcers and their antimicrobial resistance profile from selected hospitals in Addis Ababa, Ethiopia. *Frontiers in Endocrinology*, *13,* 987487.

Baindara, P., and Aslam, B. (2023). Bacillus spp.-Transmission, pathogenesis, host-pathogen interaction, prevention and treatment. Frontiers in Microbiology, 14, 1307723.

Bhaumik, R., Aungkur, N. Z., and Anderson, G. G. (2024). A guide to Stenotrophomonas maltophilia virulence capabilities, as we currently understand them. Frontiers in Cellular and Infection Microbiology, 13, 1322853.

Boyanova, L., and Mitov, I. (2013). Antibiotic resistance rates in causative agents of infections in diabetic patients: rising concerns. *Expert Review of Anti-infective Therapy*, 11(4), 411-420.

Centers for Disease Control and Prevention (CDC). (2022). *Decoding Burkholderia Pseudomallei for the Laboratory*.

Chadwick, P., and Podiatrist, S. R. (2013). Fungal infection of the diabetic foot: the often ignored complication. *The Diabetic Foot Journal*, 16(3), 102-107.

Cutting, K. F., and Harding, K. G. (1994). Criteria for identifying wound infection. *Journal of Wound Care*, *3*(4), 198-201.

Daivasikamani, D. P. (2015). Comparing swab culture, tissue culture to identify the infecting organism in diabetic foot ulcers. *Proteus, 9*, 13.

Dwedar, R. A., Ismail, D. K., and Abdulbaky, A. (2015). Diabetic foot infection: microbiological causes with special reference to their antibiotic resistance pattern. *The Egyptian Journal of Medical Microbiology (EJMM), 24*(3).

Eldukali, M., and Boshaalla, A. (2022). Prevalence of bacterial resistance with diabetic foot patients in MMC. *Academic Research Journal*, 22, 36-42.

Eltobgi, A. (2009). Libya has the highest prevalence of diabetes mellitus type 2 in North Africa and in the Arab world. *Endocrine Abstracts*, 19.

Ertugrul, B. M., Oryasin, E., Lipsky, B. A., Willke, A., and Bozdogan, B. (2018). Virulence genes fliC, toxA and phzS are common among *Pseudomonas aeruginosa* isolates from diabetic foot infections. Infectious Diseases, 50(4), 273-279.

Eshlak, M., Elshagmani, E., and Alkesa, T. (2022). Prevalence of bacterial resistance with diabetic foot patients in MMC. *مجلة البحوث الأكاديمية*، *22*، 36-42.

Frykberg, R. G., Armstrong, D. G., Giurini, J., et al. (2000). Diabetic foot disorders: A clinical practice guideline. *Journal of Foot and Ankle Surgery*, 39(5), 1-60.

González, F. C., Alramadan, M., Matesanz, M., Diaz, A., Gonzalez-Romo, F., Candel, I., ... and Picazo, J. J. (2003). Infections in diabetic foot ulcers. *European journal of internal medicine*, *14*(5), 341-343

Hadden, J., Chedid, L., and Mullen, P. (Eds.). (2016). Advances in Immunopharmacology: Proceedings of the First International Conference on Immunopharmacology*, July 1980, Brighton, England*. Elsevier12(2) 309-314.

Haesler, E. (2014). *Prevention and Treatment of Pressure Ulcers: Quick Reference Guide*. National Pressure Ulcer Advisory Panel.

Heald, A. H., O'Halloran, D. J., Richards, K., Webb, F., Jenkins, S., Hollis, S., ... and Young, R. J. (2001). Fungal infection of the diabetic foot: two distinct syndromes. *Diabetic medicine*, *18*(7), 567-572.

Huchital, M. J., Kim, J., Mantzoukas, A., and Lucido, J. V. (2020). Insights for the Growth of Stenotrophomonas maltophilia in a Diabetic Patient with Long-Term Antibiotic Use: A Case Study. *Journal of the American Podiatric Medical Association*, *110*(5).

Hung, S. Y., et al. (2023). Microbiome of limb-threatening diabetic foot ulcers indicates the association of fastidious Stenotrophomonas and major amputation. *Journal of Microbiology, Immunology and Infection*, 57(1).

Johnson, J. R. (1991). Virulence factors in *Escherichia coli* urinary tract infection. Clinical microbiology reviews, 4(1), 80-128.

Jones, J., et al. (2021). The microbiology of diabetic foot infections: A meta-analysis. *BMC Infectious Diseases*, 21(1), 770.

Kean, R., Rajendran, R., Haggarty, J., Townsend, E. M., Short, B., Burgess, K. E., ... and Ramage, G. (2017). Candida albicans mycofilms support Staphylococcus aureus colonization and enhances miconazole resistance in dual-species interactions. *Frontiers in microbiology*, *8*, 258.

Lee, J., Park, J., Kim, S., Park, I., and Seo, Y. S. (2016). Differential regulation of toxoflavin production and its role in the enhanced virulence of Burkholderia gladioli. Molecular Plant Pathology, 17(1), 65-76

Lipsky, B. A., Senneville, É., Abbas, Z. G., et al. (2020). Guidelines on the diagnosis and treatment of foot infection in persons with diabetes (IWGDF 2019 update). *Diabetes/Metabolism Research and Reviews*, 36, e3280.

Lucidi, M., Visaggio, D., Migliaccio, A., Capecchi, G., Visca, P., Imperi, F., and Zarrilli, R. (2024). Pathogenicity and virulence of Acinetobacter baumannii: factors contributing to the fitness in healthcare settings and the infected host. *Virulence*, *15*(1), 2289769

MacFaddin, J. F. (2000). *Biochemical tests for identification of medical bacteria*. Williams and Wilkins.

Mamusavu, T. S., Rose, K., and Guyah, B. (2022). Phenotypic Profiling of Selected Bacterial Microbes Isolated from Wounds of Diabetic Patients at Jaramogi Oginga Odinga Teaching & Referral Hospital, Kenya.

Mendes, J. J., and Neves, J. (2012). Diabetic foot infections: Current management and treatment. *European Journal of Internal Medicine*, 14(5), 341-343.

Neog, N., Phukan, U., Puzari, M., Sharma, M., and Chetia, P. (2021). Klebsiella oxytoca and emerging nosocomial infections. Current Microbiology, 78(4), 1115-1123.

Nuryastuti, T., Umaroh, N., Asdie, R. H., Sari, I. P., and Musthafa, A. (2018). Pan-drug-resistant and biofilm-producing strain of Burkholderia pseudomallei: first report of melioidosis from a diabetic patient in Yogyakarta, Indonesia. *International medical case reports journal*, 319-323.

Paes Leme, R. C., Chaves, J. R. E., Gonçalves, L. C. S., Alvim, L. C., Almeida, J. R. C. D., and Renó, L. D. C. (2022). Diabetic foot infection caused by bacteria of the Burkholderia cepacia complex: report of an unusual case and a scoping literature review. *Revista do Instituto de Medicina Tropical de São Paulo, 64*, e36.

Peters, E. J. G., et al. (2020). Identification of the causative bacteria in DFIs. *Diabetic Medicine*, 37.

Pickwell, K., Siersma, V., Kars, M., Apelqvist, J., Bakker, K., Edmonds, M., ... and Schaper, N. (2015). Predictors of lower-extremity amputation in patients with an infected diabetic foot ulcer. *Diabetes care*, *38*(5), 852-857.

Podschun, R., and Ullmann, U. (1998). Klebsiella spp. as nosocomial pathogens: epidemiology, taxonomy, typing methods, and pathogenicity factors. Clinical microbiology reviews, 11(4), 589-603.

Pulian Morais, V., Daporta, M. T., Bao, A. F., et al. (2009). Enteric fever-like syndrome caused by Raoultella ornithinolytica. *Journal of Clinical Microbiology*, 47(3), 868-869.

Saeed, M., Rasheed, F., Jamil, I., Rasool, M. H., Saba, N., Ahmad, M., ... and Khurshid, M. (2024). Burkholderia Cepacia: Understanding Pathogenicity, Virulence Factors, and Therapeutic Strategies. Journal of Islamabad Medical & Dental College, 13(1), 167-173.

Sanjith Saseedharan, S. S., Manisa Sahu, M. S., Roonam Chaddha, R. C., Pathrose, E., Arun Bal, A. B., Pallavi Bhalekar, P. B., ... and Padma Krishnan, P. K. (2018). Epidemiology of diabetic foot infections in a reference tertiary hospital in India. ، *1*)1 ،(18-28.

Saseedharan, S., Sahu, M., Chaddha, R., et al. (2018). Epidemiology of diabetic foot infections in a reference tertiary hospital in India. *Journal of Medical Sciences*, 1(1), 18-28.

Shaya , R., AL-Salloum, S. M., and AL-Azawi, S. S. (2006). Microorganisms Isolated From Foot Ulcers Infection Of Diabetic Iraqi Patients. *Journal of the Faculty of Medicine Baghdad*, *48*(3), 297-301.

Slovenkai, M. P. (1998). Foot problems in diabetes. *Medical Clinics of North America*, *82*(4), 949-971.

Solak, Y., Gul, E. E., Atalay, H., et al. (2011). A rare human infection of Raoultella ornithinolytica in a diabetic foot lesion. *Annals of Saudi Medicine*, 31(1), 93-94.

Sugumar, M., Kumar, K. M., Manoharan, A., Anbarasu, A., and Ramaiah, S. (2014). Detection of OXA-1 β-lactamase gene of Klebsiella pneumoniae from blood stream infections (BSI) by conventional PCR and in-silico analysis to understand the mechanism of OXA mediated resistance. *PloS one*, *9*(3), e91800.

Sumikawa, M., Egawa, T., Honda, I., Yamamoto, Y., Sumikawa, Y., & Kubota, M. (2007). Effects of foot care intervention including nail drilling combined with topical antifungal application in diabetic patients with onychomycosis. *The Journal of dermatology*, *34*(7), 456-464

Thanganadar , S., Muniyan, A., Vasanthi Mohan, K., and Panchamoorthy, R. (2021). A study on isolation, characterization, and exploration of multiantibiotic-resistant bacteria in the wound site of diabetic foot ulcer patients. *The International Journal of Lower Extremity Wounds*, *20*(1), 6-14.

Tin Som, et al. (2024). The prevalence and distribution of Burkholderia pseudomallei. *Cambodian Journal of Public Health*, 437.

Turhan, V., Mutluoglu, M., Acar, A., Hatipoglu, M., Onem, Y., Uzun, G., ... and Gorenek, L. (2013). Increasing incidence of Gram-negative organisms in bacterial agents isolated from diabetic foot ulcers. *The Journal of Infection in Developing Countries*, *7*(10),

Wang, Y. T., Li, X. W., Xu, P. Y., Yang, C., and Xu, J. C. (2022). Multiple skin abscesses associated with bacteremia caused by Burkholderia gladioli: A case report. World Journal of Clinical Cases, 10(7), 2286.

Yuan, F., Huang, Z., Yang, T., Wang, G., Li, P., Yang, B., and Li, J. (2021). Pathogenesis of Proteus mirabilis in catheter-associated urinary tract infections. *Urologia internationalis*, *105*(5-6), 354-361.