

# Study on bacteriological profile and antibiotic susceptibility pattern in patients with diabetic foot ulcers in a tertiary care teaching hospital

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## Abstract

**Introduction:** Diabetic foot ulcer and infections are one of the major complications in diabetic patients leading to frequent hospitalization and increased mortality. Knowledge about the microbes that cause infections will be helpful for providing appropriate antimicrobial therapy.

**Aim:** To evaluate the bacteriological profile of patients with diabetic foot ulcers and their antibiotic susceptibility pattern.

**Methodology:** A cross-sectional study was carried out for a period of eight months in the Department of surgery in patients with diabetic foot ulcer at a tertiary care teaching hospital. Patient data relevant to the study were collected using a standard data collection form designed as per the need of the study. Details of the organisms isolated and susceptibility pattern were collected from microbiology department.

**Results:** A total of 122 pathogens were identified from 71 patients with male (63.38%) predominance over females (36.61%). Out of the 71 patients, 38 (53.52%) patients had monomicrobial infections and 33 (46.47%) patients had polymicrobial infections. Of the total 122 organisms, 79(64.75%) organisms were found to be gram negative organisms and 43(35.24%) were gram positive. *Pseudomonas aeruginosa* found in 22 (18.03%) patients was the predominant pathogen isolated followed by *Klebsiella pneumonia* found in 18 (14.75%) patients. The gram-positive organisms isolated showed maximum susceptibility towards antibiotics Teicoplanin and Linezolid while the gram-negative organisms showed susceptibility to Imipenem, Meropenem, and Piperacillin/Tazobactam combination.

**Conclusion:** The study showed a preponderance of gram-negative bacilli among the isolates from the diabetic foot ulcers. It is recommended that antimicrobial sensitivity testing is necessary for initiating appropriate antibiotic regimen which will help to reduce the drug resistance and minimize the healthcare costs.

**Keywords:** Diabetic foot ulcer, antibiotic susceptibility, bacterial isolates

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## INTRODUCTION

Diabetes mellitus (DM) is one of the leading prevalent chronic diseases affecting a large number of population and also a major public health problem increasing globally at an alarming rate over the past couple of decades.<sup>[1]</sup>

Long-term uncontrolled blood glucose will lead to microvascular and macrovascular complications such as ischemic heart disease, nephropathy, retinopathy, neuropathy, and non-healing ulcers (diabetic foot ulcer).<sup>[2]</sup> Foot infections account for 20% of hospitalization of patients with DM annually. Infection worsens the wound infection, delays the healing mechanism and if interventions are not taken in time, it can progress to systemic infection, septicemia, amputation or even death. The vital components involved in treating diabetic foot infections are blood sugar control, treating co-morbidities, broad-spectrum antibiotic therapy, surgical treatment, proper dressing and wound care, personal hygiene, and prevention of recurrence.<sup>[3,4]</sup>

The selection of appropriate antibiotic regimen has become the need of the hour for the proper management of diabetic foot ulcers. Adequate knowledge about the microbes that cause infection is very important and helps in determining appropriate antibiotic therapy and proper management of these infections. Hence, the study was carried out to evaluate the microbiological characteristics of diabetic foot ulcers and their susceptibility pattern to various antimicrobials.

## METHODOLOGY

This was a cross-sectional study carried out in the Department of Surgery of a tertiary care teaching hospital for a period of eight months from August 2016 to March 2017. The study was approved by the Institutional Research and Ethics Committee. All the hospitalized patients of either gender aged 18 years and above, diagnosed with diabetic foot ulcer were included in the study and patients attending the outpatient clinic as well as patients whose foot is at risk but do not have diabetic foot ulcer complications were excluded from the study. Patient case sheets of the eligible subjects who met the study criteria were reviewed by the pharmacist and their data were collected using standard data collection form designed as per the need of the study. The clinical history of the patient was elicited with regards to the age of the patient, duration of diabetes, the type of treatment which was received, the presence of other systemic illnesses, size of ulcer, and duration of the ulcer. The bacterial culture reports of patient's pus samples were obtained from laboratory investigation data of the patient. Subsequently, the antimicrobial susceptibility test

was performed as per the Clinical and Laboratory Standards Institute guidelines. The microbial isolates from the samples and their susceptibility to different antimicrobials were analyzed and tabulated. Once the antibiotic susceptibility pattern was studied, the drug therapy was reviewed for their appropriateness by assessing the choice of the drug, their dosage, frequency, side effects and safety according to patients' co-morbid conditions. Data were represented as mean  $\pm$  SD, frequency, and percentage and were analyzed using SPSS (Version 16.0).

## Antibiotic sensitivity testing

Antibiotic susceptibility testing was performed using the Kirby–Bauer disk diffusion method according to the clinical and laboratory standards institute (CLSI) guidelines. The antibiotics tested for gram positive bacteria were benzyl penicillin (10 units/disc), gentamicin (120 mcg/disc), ciprofloxacin (5 mcg/disc), levofloxacin (5 mcg/disc), linezolid (30 mcg/disc), tigecycline (15 mcg/disc), vancomycin (30 mcg/disc), tetracycline (30 mcg/disc), trimethoprim/sulfamethoxazole (25 mcg/disc), piperacillin/tazobactam (100/10 mcg), imipenem 10 mcg/disc, meropenem (10 mcg/disc), ertapenem (10 mcg/disc), while the antibiotics tested for gram-negative bacteria were ampicillin (10 mcg), amoxicillin/clavulanic acid (30 mcg/disc), cefuroxime (30 mcg/disc), gentamicin (120 mcg/disc), ciprofloxacin (5 mcg/disc), cefoperazone/sulbactam (75 + 30 mcg), amikacin (30 mcg/disc), meropenem (10 mcg/disc), doripenem (10 mcg/disc), ticarcillin/clavulanic acid (100 + 10 mcg).

## RESULTS

### Age and gender-wise distribution of the study subjects

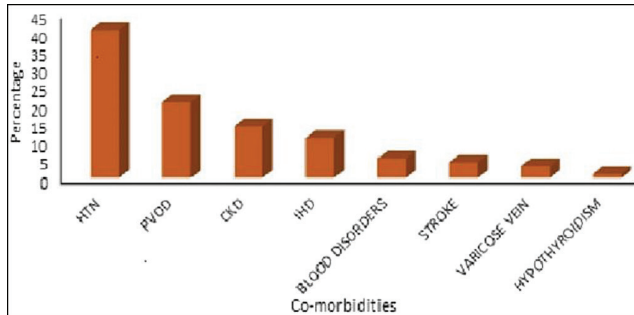
A total number of 100 patients with diabetic foot ulcer cases were reviewed during the study period and 71 patients who met the study criteria completed the study. During the study period, males [45 (63.38%)] predominance was noted over females [26 (36.61%)]. The mean age of the study populations was found to be 58.78 years  $\pm$  10.09 S.D (range: 33–85 years). According to the age wise distribution of the study subjects, the majority of the patients (28 patients) belonged to the age group of 60–69 years followed by 19 patients in the age group of 50–59 years. The age and gender-wise distribution of the study subjects is shown in the following Table 1.

### Co-morbidities among the study populations

Considering the co-morbidities in the study populations, a total of 92 co-morbidities were identified from 71 patients.

**Table 1: Age and gender-wise distribution of the study subjects**

Sl.no	Characteristics	Frequency	Percentage
1	<b>Gender</b>		
	Male	45	63.38
	Female	26	36.61
2	<b>Age-wise distribution</b>		
	30 – 39	2	2.82
	40 – 49	13	18.30
	50 – 59	19	26.76
	60 – 69	28	39.43
	70 – 79	8	11.26
	≥ 80	1	1.40

**Figure 1:** Pattern of co-morbidity among patients with diabetic foot ulcer

Hypertension was the most common co-morbidity identified during the study in 37 (40.21%) patients followed by peripheral vascular occlusive disease 19 (20.65%) and kidney diseases 13 (14.13%). The mean duration of DM in subjects was found to be  $9.24 \pm 6.778$  S.D. years (range: 0.5–32 years). It is well known that long-term uncontrolled blood glucose levels lead to macrovascular or microvascular complications including diabetic infections. In this study, the mean glycated hemoglobin value was found to be  $9\% \pm 1.894$  S.D. (range: 5–13%), which suggests poor blood sugar control. The co-morbidities of the study populations were shown in the following Figure 1.

#### Distribution of subjects according to their ulcer size, severity pattern and ulcer recurrence

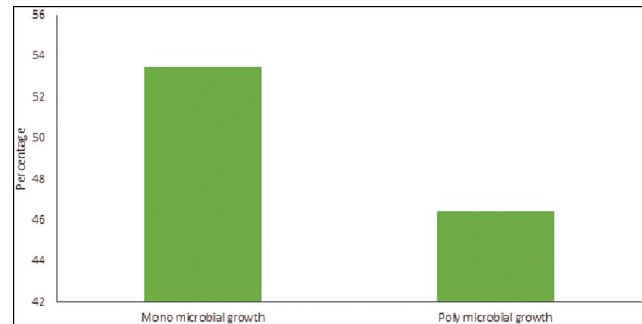
Among the total study populations, 43 (60.56%) patients were found to develop foot ulcers of size less than or equal to 4 cm and 28 (39.43%) subjects had ulcers greater than 4 cm. Out of 71 patients, 26 (36.61%) subjects developed necrotic lesion while 45 (63.38%) had non-necrotic lesion. Among the study subjects, 8 were diagnosed with gangrene, 2 with osteomyelitis and 5 cases of abscess and cellulitis. Ulcer recurrence was observed in 23 (32.39%) patients. The distribution of subjects according to their ulcer size, severity pattern, and ulcer recurrence is shown in the following Table 2.

#### Distribution of bacteria isolated from diabetic foot ulcers

In this study, monomicrobial growth was seen predominantly in 38 (53.52%) while 33 (46.47%) subjects developed

**Table 2: Size, frequency, and recurrence of ulcer in study subjects**

Sl.no	Size	Frequency & Percentage
1	Less than or equal to 4 cm <sup>2</sup>	43 (60.56)
	Greater than 4 cm <sup>2</sup>	28 (39.43)
2	<b>Number of patients</b>	<b>Frequency</b>
	Recurrence present	23 (32.39)
3	No recurrence	48 (67.60)
	<b>Severity pattern</b>	<b>Frequency</b>
	Non – necrotic	45 (63.38)
	Necrotic	26 (36.61)

**Figure 2:** Nature of microbial growth in patient with diabetic foot ulcer**Table 3: Bacteria isolated from the diabetic foot ulcer of the study subjects**

Sl no	Bacteria	Frequency (n = 122)	Percentage
1	<b>Gram-positive organisms</b>		
	Coagulase negative <i>S.aureus</i>	15	12.29
	<i>Staphylococcus aureus</i>	15	12.29
	<i>Enterococcus</i> spp.	13	10.65
	<b>Total</b>	<b>43</b>	<b>35.24</b>
2	<b>Gram-negative organisms</b>		
	<i>Acinetobacter baumannii</i>	10	8.19
	<i>Pseudomonas aeruginosa</i>	22	18.03
	<i>Klebsiella pneumonia</i>	18	14.75
	<i>E.coli</i>	12	9.83
	<i>Citrobacter koseri</i>	2	1.63
	<i>Proteus</i> spp	12	9.83
	<i>Morganella morganii</i>	3	2.45
	<b>Total</b>	<b>79</b>	<b>64.75</b>

polymicrobial growth. The nature of microbial growth in diabetic foot ulcers is shown in Figure 2.

#### Bacteria isolated from DFUs of the study subjects

Microbiological evaluation of the ulcers revealed that the prevalence of gram-negative organisms 79 (64.75%) were found to be more than gram-positive organisms 43 (35.24%). A total of 122 organisms were isolated from 71 subjects with an average of 1.71 organisms per patient. *Pseudomonas aeruginosa* was the most frequent pathogen isolated from 22 (18.03%) subjects followed by *Klebsiella pneumoniae* isolated from 18 (14.75%) subjects. The different types of gram negative and gram positive bacteria isolated from diabetic foot ulcers are summarized in Table 3.

### Prevalence of gram positive and gram-negative bacteria

Considering the bacterial growth in patients with a diabetic foot ulcer, it was found that majority of the bacterial growth were gram-negative organisms 79(64.75%) followed by 43(35.24%) gram positive. The prevalence of bacterial growth in patients with diabetic foot ulcer is shown in the following Figure 3.

### Antibiotic susceptibility pattern of gram positive and gram-negative organisms

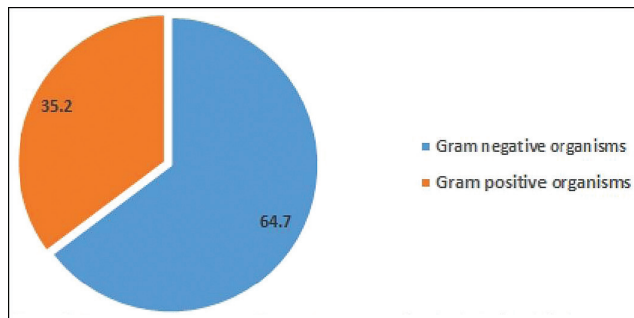
The common antibiotics are tested to determine the sensitivity pattern of the organism isolated. In this study, Enterococcus species shows maximum sensitivity to Linezolid (69.23%) followed by Teicoplanin (61.53%) and Benzyl Penicillin (61.53%). Maximum susceptibility of coagulase negative *Staphylococcus aureus* was observed to be towards Vancomycin (73.34%), Teicoplanin (73.34%), and

Linezolid (73.34%). *Staphylococcus aureus* was found to be susceptible to Vancomycin (86.67%), Teicoplanin (86.67%), and Linezolid (86.67%). The maximum susceptibility of *Staphylococcus* was towards Tetracycline (93.34%).

Among gram-negative organisms, *Acinetobacter baumannii* was found to be more susceptible to antibiotics like Tigecycline, Trimethoprim/Sulfamethoxazole and Cefoperazone/Sulbactam with 70% sensitivity each. In the case of *Klebsiella* species, maximum sensitivity was found towards Imipenem (77.78%) and Meropenem (77.78%). *Proteus* species showed 100% sensitivity towards Piperacillin/Tazobactam combination while *E.coli* shows 100% susceptibility towards Amikacin. *Morganella* species showed sensitivity towards almost all the antibiotics.

Therefore, it can be summarized that the gram-positive organisms isolated were most commonly sensitive to antibiotics Teicoplanin and Linezolid while the gram-negative organisms isolated were found to be sensitive to antibiotics like Imipenem, Meropenem, and Piperacillin/Tazobactam combination. Table 4 and 5 show antibiotic susceptibility pattern of the organisms isolated.

The resistance pattern of the pathogens isolated in patients with diabetic foot ulcers towards the common antibiotics is highlighted in Table 6 and 7.



**Figure 3:** Prevalence of gram positive and gram negative bacteria identified from the bacterial isolates of patients with diabetic foot ulcer

**Table 4:** Antibiotic susceptibility pattern of gram-positive organisms

Antibiotics	Enterococcus spp (n = 13)	Coagulase negative <i>S.aureus</i> (n = 15)	<i>Staphylococcus aureus</i> (n = 15)
Benzyl Penicillin (10 units/disc)	8 (61.53%)	1 (6.67%)	2 (13.34%)
Penicillin (10 units/disc)	-	1 (6.67%)	-
Gentamicin (120 mcg/disc)	6 (46.15%)	8 (53.34%)	12 (80%)
Ciprofloxacin (5 mcg/disc)	6 (46.15%)	4 (26.67%)	3 (20%)
Levofloxacin (5 mcg/disc)	4 (30.76%)	4 (26.67%)	11 (73.34%)
Erythromycin (15 mcg/disc)	5 (38.46%)	5 (33.34%)	5 (33.34%)
Clindamycin (2 mcg/disc)	-	5 (33.34%)	9 (60%)
Teicoplanin (30 mcg/disc)	8 (61.53%)	11 (73.34%)	13 (86.67%)
Linezolid (30 mcg/disc)	9 (69.23%)	11 (73.34%)	13 (86.67%)
Tigecycline (15 mcg/disc)	7 (53.84%)	5 (33.34%)	13 (86.67%)
Vancomycin (30 mcg/disc)	7 (53.84%)	11 (73.34%)	13 (86.67%)
Tetracycline (30 mcg/disc)	2 (15.38%)	8 (53.34%)	14 (93.34%)
Trimethoprim-Sulfamethoxazole (25 mcg/disc)	2 (15.38%)	7 (46.67%)	7 (46.67%)
Piperacillin-Tazobactam (100/10 mcg)	2 (15.38%)	-	-
Cefuroxime (30 mcg/disc)	1 (7.69%)	-	-
Cefuroxime axetil (30 mcg/disc)	1 (7.69%)	-	-
Cefoperazone-sulbactam (75 + 30 mcg)	2 (15.38%)	-	-
Cefepime (30 mcg/disc)	2 (15.38%)	-	-
Imipenem (10 mcg/disc)	1 (7.69%)	-	-
Meropenem (10 mcg/disc)	2 (15.38%)	-	-
Amikacin (30 mcg/disc)	3 (23.07%)	-	-
Colistin (10 mcg/disc)	2 (15.38%)	-	-
Ceftriaxone (30 mcg/disc)	2 (15.38%)	1 (6.67%)	-
Ertapenem (10 mcg/disc)	2 (15.38%)	-	-
Oxacillin (1 mcg/disc)	-	2 (13.34%)	8 (53.34%)



**Table 5: Antibiotic susceptibility pattern of gram-negative organisms**

Antibiotics	<i>A.baumannii</i> (n = 10)	<i>Klebsiella</i> Species (n = 18)	<i>P.aeruginosa</i> (n = 22)	<i>E.coli</i> (n = 12)	<i>C.koseri</i> (n = 2)	<i>Proteus spp</i> (n = 12)	<i>Morganella</i> spp (n = 3)
Gentamicin (120 mcg/disc)	6 (60%)	10 (55.56%)	13 (59.09%)	11 (91.67%)	2 (100%)	8 (66.67%)	3 (100%)
Ciprofloxacin (5 mcg/disc)	6 (60%)	10 (55.56%)	11 (50%)	6 (27.27%)	-	6 (50%)	2 (66.6%)
Levofloxacin (5 mcg/disc)	5 (50%)	-	10 (45.45%)	-	-	-	1 (33.3%)
Tigecycline (15 mcg/disc)	7 (70%)	8 (44.45%)	4 (18.18%)	4 (18.18%)	2 (100%)	1 (8.33%)	-
Trimethoprim/Sulfamethoxazole (25 mcg/disc)	7 (70%)	6 (33.34%)	1 (4.54%)	9 (40.90%)	1 (50%)	6 (50%)	3 (100%)
Ampicillin (10 mcg)	-	-	-	1 (8.33%)	-	6 (50%)	-
Amoxicillin-Clavulanic acid (30 mcg/disc)	-	9 (50%)	-	-	1 (50%)	-	-
Piperacillin-Tazobactam (100 + 10 mcg)	6 (60%)	11 (61.12%)	8 (36.36%)	10 (83.34%)	1 (50%)	12 (100%)	3 (100%)
Cefuroxime (30mcg/disc)	-	5 (27.78%)	-	3 (25%)	-	9 (75%)	-
Cefuroxime axetil (30 mcg/disc)	-	5 (27.78%)	-	3 (25%)	-	9 (75%)	-
Cefepime (30 mcg)	5 (50%)	11 (61.12%)	14 (63.63%)	9 (75%)	2 (100%)	10 (83.34%)	3 (100%)
Cefoperazone-sulbactam (75 + 30 mcg)	7 (70%)	11 (61.12%)	13 (59.09%)	11 (91.66%)	2 (100%)	11 (91.67%)	3 (100%)
Imipenem (10 mcg/disc)	6 (60%)	14 (77.78%)	19 (86.36%)	11 (91.66%)	2 (100%)	-	-
Meropenem (10 mcg/disc)	6 (60%)	14 (77.78%)	17 (77.27%)	11 (91.66%)	2 (100%)	11 (91.67%)	3 (100%)
Amikacin (30 mcg/disc)	6 (60%)	15 (83.34%)	16 (72.72%)	12 (100%)	2 (100%)	10 (83.34%)	3 (100%)
Colistin (10 mcg/disc)	6 (60%)	10 (55.56%)	16 (72.72%)	3 (25%)	1 (50%)	-	-
Ceftriaxone (30 mcg/disc)	-	6 (33.34%)	-	3 (25%)	1 (50%)	8 (66.67%)	2 (66.6%)
Ertapenem (10 mcg/disc)	-	13 (72.22%)	-	9 (75%)	2 (100%)	-	2 (66.6%)
Doripenem (10 mcg/disc)	-	-	10 (45.45%)	1 (8.34%)	-	-	-
Ticarcillin- Clavulanic acid (100 + 10 mcg)	-	-	8 (36.36%)	-	-	-	1 (33.3%)

**Table 6: Antibiotic resistance pattern of gram positive organisms**

Antibiotics	<i>Enterococcus spp</i> (n = 13)	<i>Coagulase negative S.aureus</i> (n = 15)	<i>Staphylococcus aureus</i> (n = 15)
Benzyl Penicillin (10 units/disc)	1 (7.69%)	13 (86.6%)	12 (80%)
Penicillin (10 units/disc)	-	1 (6.67%)	1 (6.67%)
Gentamicin (120 mcg/disc)	1 (7.69%)	6 (40%)	2 (13.3%)
Ciprofloxacin (5 mcg/disc)	5 (38.46%)	10 (66.6%)	10 (66.6%)
Levofloxacin (5 mcg/disc)	3 (23.07%)	8 (53.3%)	-
Erythromycin (15 mcg/disc)	3 (23.07%)	9 (60%)	9 (60%)
Clindamycin (2 mcg/disc)	3 (23.07%)	9 (60%)	5 (33.3%)
Teicoplanin (30 mcg/disc)	-	2 (13.3%)	1 (6.6%)
Linezolid (30 mcg/disc)	-	2 (13.3%)	2 (13.3%)
Tigecycline (15 mcg/disc)	-	1 (6.6%)	-
Vancomycin (30 mcg/disc)	-	3 (20%)	1 (6.6%)
Tetracycline (30 mcg/disc)	7 (53.8%)	3 (20%)	1 (6.6%)
Trimethoprim-Sulfamethoxazole (25 mcg/disc)	-	2 (13.3%)	3 (20%)
Piperacillin-Tazobactam (100 + 10 mcg)	1 (7.69%)	-	-
Cefuroxime (30 mcg/disc)	1 (7.69%)	-	-
Cefuroxime axetil (30 mcg/disc)	1 (7.69%)	-	-
Cefoperazone-sulbactam (75 + 30 mcg)	2 (15.38%)	-	-
Cefepime (30 mcg/disc)	1 (7.69%)	-	-
Ceftriaxone (30 mcg/disc)	2 (15.38%)	1 (6.67%)	-

## DISCUSSION

Diabetic foot ulcer is an important complication of DM. Untreated diabetic foot ulcers will become infected leading to various other consequences such as gangrene or amputation of the limb. Surgical intervention and treatment with antibiotic regimen are the options used for the management of DFUs.<sup>[5]</sup> The study was carried out to determine the predominant organisms isolated from DFUs and evaluate their sensitivity pattern to different antimicrobials which are vital for prescribing appropriate antibiotic regimen.

In the present study, male predominance was noted over females. Previous studies have shown that the susceptibility to foot infections is greater in male patients than in female patients.<sup>[6,7]</sup> This may be due to the fact that males tend to be more active in the outdoor activities leading to injuries and prone to development of ulcers. In the current study, we found that elderly patients with age range 60–69 years constituted the majority with foot infections. The mean age of patients in the present study is  $58.78 \pm 10.09$  years which is on the line of study by Sundresh NJ *et al.*<sup>[8]</sup> and Halpati

**Table 7: Antibiotic resistance pattern of gram-negative organisms**

Antibiotics	<i>A.baumannii</i> (n = 10)	<i>Klebsiella</i> Species (n = 18)	<i>P.aeruginosa</i> (n = 22)	<i>E.coli</i> (n = 12)	<i>C.koseri</i> (n = 2)	<i>Proteus spp</i> (n = 12)	<i>Morganella spp</i> (n = 3)
Gentamicin (120 mcg/disc)	3 (30%)	3 (16.6%)	9 (41%)	1 (8.3%)	-	3 (25%)	-
Ciprofloxacin (5 mcg/disc)	3 (30%)	6 (33.3%)	10 (45.45%)	5 (41.6%)	1 (50%)	5 (41.6%)	-
Levofloxacin (5 mcg/disc)	1 (10%)	-	10 (45.45%)	-	-	1 (8.3%)	-
Tigecycline (15 mcg/disc)	1 (10%)	-	11 (50%)	-	-	4 (33.3%)	1 (33.3%)
Trimethoprim/Sulfamethoxazole (25 mcg/disc)	1 (10%)	9 (50%)	11 (50%)	2 (16.6%)	1 (50%)	4 (33.3%)	-
Ampicillin (10 mcg/disc)	3 (30%)	15 (83.3%)	1 (4.6%)	10 (83.34%)	-	5 (41.67%)	2 (66.6%)
Amoxicillin-Clavulanic acid (30 mcg/disc)	3 (30%)	7 (38.9%)	1 (4.6%)	2 (16.6%)	1 (50%)	1 (8.3%)	2 (66.6%)
Piperacillin-Tazobactam (100 + 10 mcg)	4 (40%)	6 (33.3%)	11 (50%)	1 (8.3%)	-	1 (8.3%)	-
Cefuroxime (30 mcg/disc)	3 (30%)	11 (61.72%)	1 (4.6%)	8 (66.6%)	2 (100%)	2 (16.6%)	2 (66.6%)
Cefuroxime axetil (30 mcg/disc)	3 (30%)	11 (61.72%)	1 (4.6%)	8 (66.6%)	2 (100%)	2 (16.6%)	2 (66.6%)
Cefoperazone-sulbactam (75 + 30 mcg/disc)	2 (20%)	5 (27.8%)	9 (41%)	1 (8.3%)	-	1 (8.3%)	-
Cefepime (30 mcg/disc)	4 (40%)	6 (33.3%)	6 (50%)	2 (16.6%)	-	2 (16.6%)	-
Imipenem (10 mcg/disc)	3 (30%)	3 (16.67%)	3 (13.7%)	1 (8.3%)	-	-	-
Meropenem (10 mcg/disc)	3 (30%)	3 (16.67%)	4 (18.18%)	1 (8.3%)	-	1 (8.3%)	-

A *et al.*<sup>[9]</sup> which showed an incidence of 58.3 years and 59.5 years, respectively.

Among the co-morbidities, hypertension was the most prevalent co-morbidity seen among the study population followed by peripheral vascular occlusive disease, kidney diseases, ischemic heart disease, and blood disorders. These findings are consistent with the earlier published literatures.<sup>[10,11]</sup> It is well known that hypertension is a common co-morbidity in patients with DM with a prevalence of up to two-thirds of the population and it may be present by the time patients are diagnosed to have DM or even before the onset of hyperglycemia. Hypertension enhances the risk for cardiovascular diseases in patients with DM by increasing the risk of developing microvascular and macrovascular complications.

Our study revealed that 43 out of 71 patients developed ulcers with size less than or equal to 4cm<sup>2</sup> and showed recovered quickly while 28 subjects had ulcers of size greater than 4cm<sup>2</sup> where recovery was slower compared to patients with smaller ulcer size. A study carried out by Oyibo *et al.*<sup>[12]</sup> showed that greater the area of the ulcer more would be the healing time.

The current study categorized the ulcers into necrotic (36.61%) and non-necrotic (63.38%) where the frequency of subjects who presented with non-necrotic ulcers was higher. Previous studies have shown that there is an association between bacterial growth and severity pattern of ulcers.<sup>[13]</sup> In our study, monomicrobial growth (53.52%) was predominant than polymicrobial infections (46.47%) which could be the reason for the increased prevalence of non-necrotic ulcers. Monomicrobial nature of diabetic foot ulceration has been reported in several studies conducted in

this region and elsewhere.<sup>[14,15]</sup> But few studies from India have reported a higher prevalence of necrotic cases and polymicrobial infections.<sup>[16-18]</sup>

Microbiological evaluation of diabetic foot ulcer infections showed that the prevalence of gram-negative organisms was found to be more than gram-positive organisms. *Pseudomonas aeruginosa* was the most frequent followed by *Klebsiella pneumonia* and *E.coli*. These findings correlated well with those of studies carried out in India which showed that gram-negative bacilli as the most common organism and *pseudomonas* being the predominant pathogen.<sup>[19-21]</sup> However, few studies reported gram positive as the most common organism and *Staphylococcus aureus* as the most common isolate.<sup>[22-24]</sup> Therefore, there seems to be a changing trend in the organisms causing diabetic foot infections with gram-negative bacteria replacing gram-positive bacteria as commonest agents. It also confirms the fact that the diabetic foot infections do not have a clear etiology.

Knowledge about the antibiotic susceptibility pattern of the isolates is also essential for proper management of diabetic foot infections. Antibiotics such as Vancomycin, Tigecycline, and Linezolid showed >85% susceptibility towards gram- positive isolates. In the present study, *Staphylococcus* species isolated were susceptible to Gentamicin, Tetracycline, Vancomycin, Teicoplanin, and Linezolid. So, these antibiotics seem to be appropriate for empirical treatment of diabetic foot infections. Coagulase negative staphylococcus showed only 73% susceptibility towards Teicoplanin, Linezolid, and Vancomycin. Most of the gram-positive organism showed low susceptibility to Penicillin, Cephalosporins, and Fluoroquinolones.

In the present study, Imipenem and Meropenem showed very good susceptibility against *Pseudomonas* and *Klebsiella* species. Amikacin showed 100% susceptibility against *E.coli* whereas Piperacillin with Tazobactam combination, Gentamicin, Meropenem, Imipenem, and Cefoperaxone with Sulbactam combination showed more than 80% susceptibility. *Proteus* species showed 100% susceptibility to Piperacillin with Tazobactam combination and more than 80% susceptibility to Cefepime, Amikacin, Meropenem, and Cefoperazone with Sulbactam combination. *Acenobacter* species showed low susceptibility towards most of the fluoroquinolones, aminoglycosides, cephalosporins, and carbapenams.

Considering the resistance pattern of antibiotics towards the bacterial isolates, in our study, many organisms showed multi-drug resistance towards gram negative and gram-positive organisms. This increased incidence of multi-drug resistant organisms is a potential risk factor in the management of diabetic foot infections which may lead to the occurrence of complications like systemic toxicity, gangrene formation, and amputation of lower extremity. Initiating a combination therapy has been considered as the better option for the successful treatment of diabetic foot infections.

Management of gram-negative infections is extremely challenging. Future studies should also focus on identifying the risk factors for the development of these infections so that appropriate treatment can be initiated early and can prevent or minimize drug resistance and fatal outcomes.

The present study demonstrates that a variety of organisms can be isolated from diabetic foot ulcers. Knowledge about the microbes that cause infection and their susceptibility towards the antibiotics will allow physicians to make best out their choice. Considering the nature of the organism and the type of isolate appropriate empirical antibiotic therapy should be initiated especially for the patients who are at risk categories. Once the nature of the organism and the probable pathogens are isolated, de-escalation of empiric therapy with a single drug or combination therapy can be guided by relevant culture results.

## CONCLUSION

This study showed the predominance of gram-negative organisms over gram-positive organisms with the majority of the infections to be monomicrobial in nature. It is necessary to evaluate the culture sensitivity test from the infected wound and the knowledge on the antibiotic

sensitivity pattern of the isolates helps in planning treatment with the appropriate antibiotic regimen. This, in turn, helps to prevent the emergence of drug-resistant organisms and minimizing healthcare costs. Clinical pharmacists can play a vital role in suggesting suitable antibiotic treatment regimen for the proper management of diabetic foot ulcers. They can also be involved in educating the patients about the importance of maintaining optimal glycemic control and avoiding the risk factors for developing ulcers which will help in improving the quality of life.

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## Conflicts of interest

There are no conflicts of interest.

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