

## Study the susceptibility pattern of bacteria isolated from infected wounds and determine various risk factors associated with foot ulcer

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### ABSTRACT

Studying of bacteria prevalence and antimicrobial susceptibility in samples from foot ulcer patients with chronic wounds will provide the epidemiological information on chronic wound infections, representing support for diagnosis, treatment and management of this pathology, thus preventing further complications of foot infection. There are many risk factors associated with a foot ulcer, so identifying those risk factors and preventing them will help in reducing the incidence of the disease to a certain extent. Identifying the type of organisms causing the chronic wound infection, antibiotic sensitivity and resistance representing support for diagnosis, treatment and management thus preventing further complications of foot infection, and to understand the significant risk factors associated with the development of foot ulcers. An interventional study was conducted among the 80 patients with foot ulcers admitted in General surgery ward of a medical college teaching hospital from Dec 2018 to May 2019. Antimicrobial susceptibility results showed that gram-negative organism was more prevalent and among the species, the isolated majority was found to be *Staphylococcus aureus* 28 (0.35%) followed by *Klebsiella* 16(20%) and *E.coli* 15(18.75%). The most sensitive antibiotic found was Meropenam 70 (87.5%) followed by Imipenam 67(83.75%) and Linezolid 65(81.25%) The most resistant antibiotic was Cotrimoxazole 66(82.5%). This study concludes that high proportion of foot ulcers were found amongst diabetic patients than non-diabetic patients, and were often associated with trauma, cellulitis, gangrene. Some of the critical risk factors for foot ulcers included low educational status, previous history of foot ulcer, previous amputation was done, duration of ulcers, smoking, peripheral neuropathy, infection and HbA1c levels of patients.

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### INTRODUCTION

Foot ulcers are mainly associated with diabetes but can be seen in people without diabetes also. This can be a significant saddle to patients and also the health care system, especially those that recur or do not heal. A foot ulcer is defined as an open sore on the foot or a thin red crater that involves not only the superficial skin but also can be very deep [1-3]. Patients with diabetes, diabetic neuropathy and other circulation issues are more likely to get foot ulcers. Other factors that increase the risk of foot ulcers include Heart diseases, kidney diseases, obesity, nerve damage, wounded feet, tobacco use [4].

There are various risk factors in the development of foot ulcer in diabetic patients like

- Previous history of lower extremity amputation
- History of a foot ulcer
- Anatomic deformity on feet
- Peripheral vascular disease
- Smoking
- Poor control of hyperglycemia
- In dialysis patients, diabetic nephropathy can be a cause [5].

### Antibiotic therapy

Antibiotic therapy given should always be based on bacterial culture results and also the antibiotic's toxicity capability. Mild and moderate infections may be treated by oral antibiotics, such as cephalexin, amoxicillin-clavulanic, moxifloxacin or clindamycin. Severe infections will be due to polymicrobial infection, such as staphylococcus, streptococcus, Enterobacteriaceae, pseudomonas, enterococcus and anaerobic bacteria, e.g. Bacteroides, Pepto cocci, Pepto streptococci. Intravenous antibiotics used for severe infections include imipenem-cilastatin, b-lactam lactamase (ampicillin-sulbactam and piperacillin-tazobactam) and broad-spectrum cephalosporin [2].

### Antimicrobial susceptibility

It is estimated that around 15% of diabetic patients develop foot ulcer, which is highly vulnerable to infections. And these foot ulcer infections generally spread due to polymicrobial growth, mostly consisting of aerobic, gram-positive and gram-negative organisms. Hence there comes a need for adequate management of these infections, which requires appropriate antibiotic selection based on culture and susceptibility test reports. Knowledge regarding the microbes that cause infections helps decide appropriate antibiotic therapy and thereby reduce the cost spend on treatment and improve the treatment management [6]. Studies suggest that the most frequent organisms in diabetic foot ulcers are Staphylococcus aureus; followed by E.coli and Klebsiella. Most of the organisms were sensitive to Meropenem and most resistant to Cotrimoxazole [7]. Prevalence shows that gram-negative bacteria were slightly more than gram-positive bacteria in foot ulcers. Studies recommend doxycycline should be the empirical treatment of choice for gram-positive isolates and Amikacin, cefoperazone; Meropenem should be considered for most of the gram-negative aerobes [6].

**Methods**The study participants included inpatient foot ulcer patients attending The Oxford Medical

College Hospital and Research Center, Attibele, Bangalore. This study was carried out from December 2018 to May 2019. A total of 80 patients who provided written informed consent and had diabetic or non-diabetic foot ulcers, admitted in the department of surgery were included. Pregnant women, paediatrics, psychiatric patients and who are not willing to participate were excluded from the study.

Demographics of the patients (Name, Age, Sex, IP number, socioeconomic status etc.) and the data regarding past medical history, past medication history, diagnosis, culture and antibiotic sensitivity test reports were collected through the data entry form.

The obtained data were subjected for a suitable statistical method using like student t-test, chi-square test, Fischer exact test, mean, standard deviation and results were expressed in the form of suitable graphs.

### Research and ethical committee approval

Institutional Ethics Committee of The Oxford Medical College, Attibele, Bengaluru approved the study and issued a letter of permission to conduct the study.

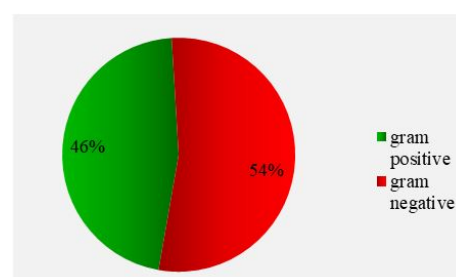
## RESULTS

### Distribution of types of organisms isolated from infected wound

This data shows the distribution of the type of organisms from the wound after the culture test [Table 1] [Figure 1].

**Table 1: Distribution of types of organisms isolated from an infected wound**

No. of patients with foot ulcer(N=80)	Gram-positive	Gram-negative
	37(46.25%)	43(53.75%)



**Figure 1: Distribution of types of organisms isolated from an infected wound**

The result showed that about 43(537.5%) of the infected wound was affected by gram-negative organisms, whereas 37(46.25%) of the infected wound was affected by the gram-negative organism.

**Distribution of types of species isolated from an infected wound** The following data shows the distribution of types of species isolated from the infected wound among the patients affected with foot infection [Table 2 ] [Figure 2 ].

**Table 2: Distribution of types of species isolated from an infected wound**

Type of organism	Name of the organism	No. of samples
Gram-negative (n=43)	E. coli	13 (16.25%)
	Pseudomonas aeruginosa	7 (8.75%)
	Klebsiella	15 (18.7%)
	Acinetobacter	2 (2.5%)
	Acetobacter	3 (3.75%)
	Klebsiella oxy-toca	1 (1.25%)
Gram positive (n=37)	Pseudomonas	2 (2.5%)
	Enterococci	10 (12.5%)
	Staphylococcus aureus	27 (33.75%)

This result shows that the most commonly isolated species from the wound was Staphylococcus aureus 27 (33.75%) among gram-positive organisms and Klebsiella species 15 (18.7%) among gram-negative organisms.

#### Distribution of antibiotics sensitivity and resistance

The following data shows the antibiotics sensitivity and resistance towards the organisms causing the foot infection [Table 3 ] [Figure 3 ].

Based on the study of antibiotics sensitivity and resistance towards the organisms causing the foot infection, the data result shows that:-

Meropenem (87.5%), Imipenem (83.75%), Linezolid (81.25%) were the most Sensitive antibiotic whereas Cotrimoxazole (82.5%), Penicillin (66.25%), Ceftriaxone (67.5%) were found to be the most Resistant antibiotic.

#### Association of various risk factors for foot ulcers

The following data shows the association between various risk factors towards the development of foot ulcers [Table 4 ].

**P-value <0.05 are significant, Chi-square test<sup>a</sup>, Fisher exact test<sup>b</sup>**

**Table 3: Distribution of Antibiotics Sensitivity and Resistance**

Drugs	Sensitive	Resistant
Amikacin	58(72.5%)	22(27.5%)
Amoxiclav	31(38.75%)	49 (61.25 %)
Cefotaxime	41(51.25%)	39 (48.75%)
Ceftriaxone	26(32.5%)	54(67.5%)
Clindamycin	42(52.5%)	38(47.5%)
Ciprofloxacin	42(52.5%)	38(47.5%)
Cotrimoxazole	14(17.5%)	66(82.5%)
Erythromycin	42(52.2%)	38(47.5%)
Gentamicin	49(61.25%)	31(38.75%)
Imipenem	67(83.75%)	13(16.25%)
Linezolid	65(81.25%)	15(18.75%)
Meropenem	70(87.5%)	10(12.5%)
Penicillin	27(33.75%)	53(66.25%)
Tetracycline	46(57.5%)	34(42.5%)
Vancomycin	39(48.75%)	41(51.25%)

Our study results showed that Educational status (p=0.001436), Previous history of foot ulcer (p=0.00052), Previous amputation done (p=0.001619), smoking (0.000001), Duration of ulcer (p=0.000025), Peripheral neuropathy (p=0.01206), Infections (p=0.008678), HbA1C level (p= <0.00001) were the risk factors for the development of foot ulcer.

#### DISCUSSION

An interventional study was carried out to study the susceptibility pattern of bacteria isolated from infected wounds and to study the risk factors among patients with diabetic and non-diabetic foot ulcer attending The Oxford Medical College, Hospital & Research Centre, Bangalore.

The study of bacteria prevalence and antimicrobial susceptibility in samples from foot ulcer patients with chronic wounds provide the epidemiological information on chronic wound infections, representing support for diagnosis, treatment and management of this pathology, thus preventing further complications of foot infection. Sekhar SM et al. [6] found gram-negative bacteria were slightly more than gram-positive bacteria in diabetic foot ulcers. The same was found in the study performed by Jain SK et al. [8]. Another study conducted by Nageen A [7]. Shows that the most effective antibiotic is Meropenam and least effective is Cotrimoxazole. A culture-based study was conducted by Noor S et al. [9] which found that the commonest isolate among gram-negative organ-

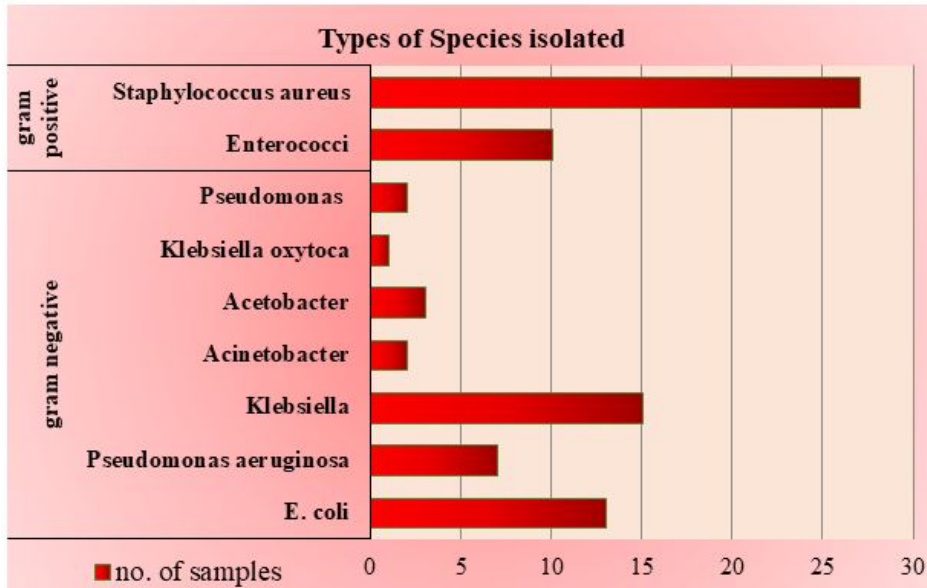


Figure 2: Distribution of types of species isolated from an infected wound

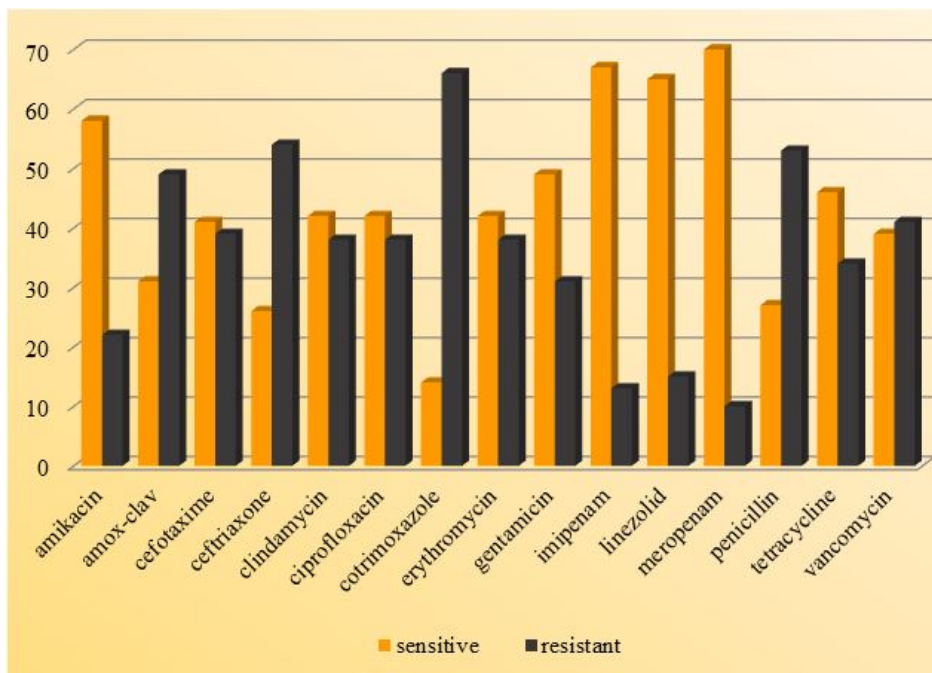


Figure 3: Distribution of Antibiotics Sensitivity and Resistance

**Table 4: Association of various Risk Factors for Foot Ulcer**

Variable	Diabeticfoot (n=48)	ulcer	Non-diabetic foot ulcer (n= 32)	$\chi^2$ value	p-value
<b>Gender</b>				2.675	0.101934 <sup>a</sup>
Male	38 (79.16%)		20 (62.5%)		
Female	10 (20.85)		12 (37.5%)		
<b>Age</b>				5.6928	0.01703 <sup>a</sup>
<50	10 (20.8%)		12 (37.5%)		
≥ 50	38 (79.1%)		20 (62.5%)		
<b>Educational status</b>				10.1587	0.001436 <sup>a*</sup>
Below PUC	40 (83.3%)		16 (50%)		
Above PUC	8 (16.6%)		16(50%)		
<b>Previous history of foot ulcer</b>				12.0409	0.00052 <sup>a*</sup>
Yes	31 (64.5%)		8 (25%)		
No	17 (35.4%)		24 (75%)		
<b>The previous amputation was done.</b>				9.9377	0.001619 <sup>a*</sup>
Yes	35(72.9%)		12 (37.5%)		
No	13(27.0%)		20 (62.5%)		
<b>Smoking</b>				25.0677	0.000001 <sup>a*</sup>
Yes	39 (81.25%)		8 (25%)		
No	9 (18.75%)		24 (75%)		
<b>Duration of ulcer</b>				17.7331	0.000025 <sup>a*</sup>
< 2 month	13 (27.0%)		24 (75%)		
> 2 month	35(72.9%)		8 (25%)		
<b>Peripheral neuropathy</b>				6.3021	0.01206 <sup>b*</sup>
Yes	14 (29.1%)		2 (6.2%)		
No	34 (70.8%)		30(93.7%)		
<b>Peripheral vascular disease</b>				2.5296	0.111725 <sup>b</sup>
Yes	9 (18.7%)		2 (6.2%)		
No	39 (81.2%)		30 (93.%)		
<b>Gangrene</b>				0.0231	0.879072 <sup>b</sup>
Yes	5 (10.4%)		3(9.3%)		
No	43 (89.5%)		29 (90.6%)		
<b>Trauma</b>				2.1818	0.139649 <sup>a</sup>
yes	12 (25%)		13 (40.6%)		
No	36 (75%)		19 (59.3%)		
<b>Infections</b>				6.888	0.008678 <sup>a*</sup>
Yes	5 (10.4%)		11 (34.3%)		
No	43 (89.5%)		21 (65.6%)		
<b>HbA1c (%)</b>				44.1944	<0.00001 <sup>b*</sup>
<6.5	6 (12.5%)		28 (87.5%)		
>6.5	42(87.5%)		4 (12.5%)		

isms was E.coli. Among gram-positive organisms, it was Staphylococcus aureus. It was also found that in non-healing ulcers, the primary causative organism was Pseudomonas aeruginosa, E.coli. Vancomycin (100%), Amikacin (90.4%) exhibited the highest sensitivity to gram-positive cocci and antibiotic Imipenam towards Paeruginosa. Likewise, our study found out that about 43(53.75%) of the infected wound was affected with gram-negative organisms, whereas 37(46.25%) of the infected wound was affected with the gram-negative organism. The most commonly isolated species from the wound was Staphylococcus aureus 27 (33.75%) among gram-positive organisms and Klebsiella species 15 (18.7%) among gram-negative organisms. The most sensitive antibiotic found in our study was Meropenam 70(87.5%) followed by Imipenam 67(83.75%) and Linezolid 65(81.25%), and the resistant antibiotic was Cotrimoxazole 66(82.5%). Preliminary evaluation of bacterial flora and their antibiotic sensitivity is crucial in the selection of potent antibiotics for the management of foot ulcer.

Identification of high-risk in foot ulcer patients is essential, so that appropriate preventative measures can be taken to reduce the incidence of ulceration. The increase in foot ulcer is becoming a burden and worrying for individual families, especially when males are the backbone and the sole earning members of the family, particularly in India. Several researchers have reported the occurrence of foot ulcers mostly in males and middle-aged subjects. Bharati Amar Taksande et al. [10] found that Less education, poor socioeconomic status, and unawareness of foot care were contributing factors of improper footwear practices, which increases the risk for the diabetic foot. Bhaktavatsalam M [11] found that males were more affected. The majority were illiterate and were found to be smokers. Shahi SK et al. [12] found that previous ulceration and amputation have been recognized as risk factors for subsequent diabetic limb ulceration. The other risk factors were neuropathy and ischemia, foot deformities, nephropathy, previous amputation of one limb and HbA1c are also regarded as contributory factors for a diabetic foot ulcer. In a study done by Khalid Al-Rubeaan et al. [3] found that PVD, neuropathy, duration of ulcer, poor glycemic control, male gender, smoking to be significant risk factors of foot ulcer. Hokkam EN [13] found risk factors for foot ulcers were previous foot ulcer, PVD, peripheral neuropathy, longer duration, poor glycaemic control and presence of infection. Our study results showed that Educational status ( $p=0.001436$ ), Previous history of foot ulcer ( $p=0.00052$ ), Previous amputa-

tion done ( $p=0.001619$ ), smoking (0.030971), Duration of ulcer ( $p=0.000025$ ), Peripheral neuropathy ( $p=0.01206$ ), Infections ( $p=0.008678$ ), HbA1C level ( $p= <0.00001$ ) were the risk factors for the development of foot ulcer.

## CONCLUSION

An interventional study was carried out to study the susceptibility pattern of bacteria isolated from infected wounds and risk factors among patients with diabetic and non-diabetic Foot Ulcer. This study emphasizes on the type of organism isolated and its antibiotic sensitivity which is found to be an essential factor for the better management of the disease. Gram-negative bacteria were slightly more than gram-positive bacteria in diabetic foot ulcers. The most commonly isolated species from the wound was Staphylococcus aureus. A most effective antibiotic is Meropenam and least effective is Cotrimoxazole.

Identification of high-risk in foot ulcer patients is essential, so that appropriate preventative measures can be taken to reduce the incidence of ulceration. Significant risk factors for foot ulcers include low educational status, previous history of foot ulcer, previous amputation was done, duration of ulcers, smoking, peripheral neuropathy, infection and HbA1c levels of patients. Less education, low socioeconomic status, and unawareness of foot care were contributing factors of improper footwear practices, which increases the risk for the diabetic foot.

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## Conflict of Interest

The authors declare that they have no conflict of interest for this study.

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