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Bacteriological Profile And Antimicrobial Susceptibility Pattern Of Bacteria Isolated From Pus/Wound Swab Sample From Diabetic Patient Attending Tertiary Care Hospital, Chengalpattu District

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Abstract

Background: *Diabetic foot ulcers (DFUs) commonly result in days of significant hospital stays and are the primary cause of lower limb amputations in medicine. To choose the best antibiotic treatment, it is essential to identify the bacteria causing DFUs. Numerous studies have shown that antibiotic resistance has dramatically increased, particularly in wound infections in diabetic patients.*

Objectives: *In order to determine the prevalence of bacteria and the profiles of their antibiotic sensitivity, the current study looked at a variety of patient cultures with DFUs.*

Methods: *This is a cross-sectional study. 253 DFU patients who were attending the OPD of a tertiary care hospital at Chengalpattu district, Tamil Nadu, India was included in this study. Sterile swabs were used to collect the samples needed for testing. Routine microbiological laboratory Conventional tests were done and results were recorded.*

Results*: A total of 155 microorganisms were isolated. Klebsiella spp (36.13%), Escherichia coli (19.35%) and Proteus spp., (9.68%) were the major three organisms isolated. The most often isolated gram-positive and gram-negative bacteria were Staphylococcus species and Klebsiella pneumoniae, respectively. Vancomycin shown 100% sensitivity to all Gram-positive bacteria, although meropenem and Piperacillin/tazobactum were found to be effective against Gram negative bacteria.*

Conclusion*: In conclusion, because multiple bacteria may be implicated and multidrug-resistant strains may evolve, doctors are encouraged to take cultures into consideration before beginning empirical therapy.*

Keywords: *Diabetic foot ulcer, Bacteria, Antibiotic sensitivity, Resistance, Klebsiella spp.*

INTRODUCTION

About 415 million people are known to have diabetes since the disease's rising prevalence causes its complications to worsen. Due to the need for specialised care, diabetes and its complications are predicted to increase morbidity, death, and health spending. Diabetic neuropathy is the cause of diabetic foot ulcers (DFU), a serious and common consequence of diabetes mellitus (DM) that raises treatment costs dramatically [1-2].

Around 8.3% of people worldwide have diabetes mellitus (DM), and over 79 million people have prediabetes. In contrast, the rate of complications is quite high in developing nations like India, where 42% of people have the disease [2-3].

Nearly 40 million individuals in India have diabetes, and many of them live in poverty. Since 20% of patients have diabetic foot infections, these are the most prevalent clinical problems to address. According to a thorough analysis, people with diabetes have a high risk (12–25%) of getting foot ulcers in their lifetime. Between 40 and

80 percent of cases that are diagnosed are opportunistic infections, which are the leading cause of morbidity and mortality [4-5].

One of the most serious and debilitating complications of diabetes is diabetic foot, which is a collection of syndromes where infection, ischaemia, and neuropathy cause tissue deterioration and may even result in amputation. In their lifetime, 15% of diabetes individuals will get foot ulcers, which in 85% of cases are known to occur before amputation. Because diabetic foot ulcers do not receive enough oxygen or nutrients from the blood, they are difficult to heal and can result in lower limb amputation [6].

Patients with diabetes who are hospitalised are more likely than those who are not to have soft tissue and bone infections of the foot. Diabetic foot infections are caused by both aerobic and anaerobic microorganisms. Gram negative bacilli and a small number of anaerobic pathogens, which are primarily linked to ischaemic or necrotic wounds, are secondary to gram positive bacterial pathogens in diabetic foot ulcers. Bacterial pathogens such as *Staphylococcus aureus, Enterococcus faecalis, Pseudomonas aeruginosa, Escherichia coli, Proteus mirabilis, Acinetobacter baumanni, Klebsiella pneumoniae*, and others are frequently found in the majority of research [7-8].

Long-term infections are now the main reason for amputations, and more cases of multidrug-resistant (MDR) bacterial infections are frequently recorded. Hygiene measures and isolation procedures may promote the early healing of wounds and ulcers because cross-transmission of such infectious agents is actively presented in both community and hospital environments [9-10].

The main objective of this study includes, to isolate the aerobic pyogenic bacteria from pus samples; to determine the antibiotic susceptibility pattern of gram negative and gram-positive bacteria isolated from pus samples; to determine the socio demographic factors involved in this study; to provide relevant information to the clinicians of our hospital regarding the antibiotic to be prescribed to decrease the emergence and spread of multidrug resistant bacteria.

MATERIALS AND METHODS

Study design: This cross-sectional study was carried out in the in the department of Microbiology, Shri Sathya Sai Medical College & Research Institute, Chengalpattu district of Tamil Nadu from August 2024 to June 2025. A total of 253 specimen consisting of wound /pus swabs collected from patients of OPD of general surgery department. Clinically diagnosed diabetic patients of age 20 & above of both sexes having wound infection with pus or discharge draining from infection site or wounds were included in the study.

Sample collection and processing: Following a comprehensive clinical examination, the surgeons obtained a detailed history from the patients and evaluated the ulcers using the Wagner categorisation of ulcers. Swabs were taken from the ulcer by spinning the swab over a 1 cm portion of the lesion for 5 seconds while applying enough pressure to draw fluid from the inner section of the wound after the area had been cleansed with 0.9% sterile saline and debrided. Before the specimen was collected, care was taken to ensure that no antiseptic or antimicrobial substance was applied to the site. Two swabs were taken from the ulcers' depth. One swab was utilised for culture and the other for microscopic analysis using Gram stain.

Within 30 minutes after collection, the specimens were sent to the microbiology lab for aerobic culture, Gram staining and sensitivity testing in sterile transport containers. This study did not use anaerobic culturing. The sample was subjected to Gram-staining, and the smear was analysed. Plates were incubated at 37°C for 24 hours after swabs were further inoculated on Blood agar, Chocolate agar, MacConkey agar and Thioglycolate media. Using established procedures, the colonies were identified for their unique characteristics, such as colony morphology, staining reactions, and biochemical reactions. Kirby Bauer's disc diffusion technique was used to test for antibiotic sensitivity in accordance with the Clinical Laboratory Standard Institute (CLSI-2024) standards. Data were compiled and analysed with SPSS version 17 statistical software. Before starting the study, ethical clearance was obtained.

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RESULTS:

The study comprised 253 samples from patients with diabetic foot ulcers, 155 of which displayed organism development. Of the 155, 54 were female and 101 were male. They were between the ages of 29 and 90. 35 (22.58%) patients had diabetes for less than a year, while 120 (77.41%) patients had diabetes for more than a year. 66.45% of patients experienced ulcers for longer than a month, whereas 33.54% of patients experienced ulcers for less than a month. (Table 1)

A total of 155 bacteria included in this study. Out of this 20.65% were Gram Positive Cocci and 79.35% were Gram Negative Bacilli (Figure 1)

56 (36.13%) of the isolates in this study were *Klebsiella species*, it was the most commonly isolated organism. *Escherichia coli* (19.35%), *Staphylococcus aureus* 10.97%, Proteus *spp.* (9.68%) and *Staphylococcus other than Staphylococcus aureus* (9.68%) were the next most common bacteria. *Acinetobacter spp.* (6.45%), *Pseudomonas aeruginosa* (6.45%), *Citrobacter spp* (1.29%) were the other organisms that were isolated. (Table 2)

All of the gram-positive cocci in our investigation were extremely susceptible to vancomycin, which was followed by Co-trimoxazole (Table 3). Meropenem exhibited high sensitivity to all gram-negative bacteria (Table 4).

Table 1: Demographic and clinical details of diabetic foot ulcer patients

Demographic Details		Number of subjects included(n=155)
Gender	Male	101
	Female	54
Age	30 and below	1
	31-40	12
	41-50	78
	51-60	33
	61-70	21
	70 and above	10
Duration of Diabetics	< 1 year	35
	> 1 year	120
Duration of Ulcer (Days)	< 30 days	52
	> 30 days	103

Figure 1: Total number of organisms isolated from samples

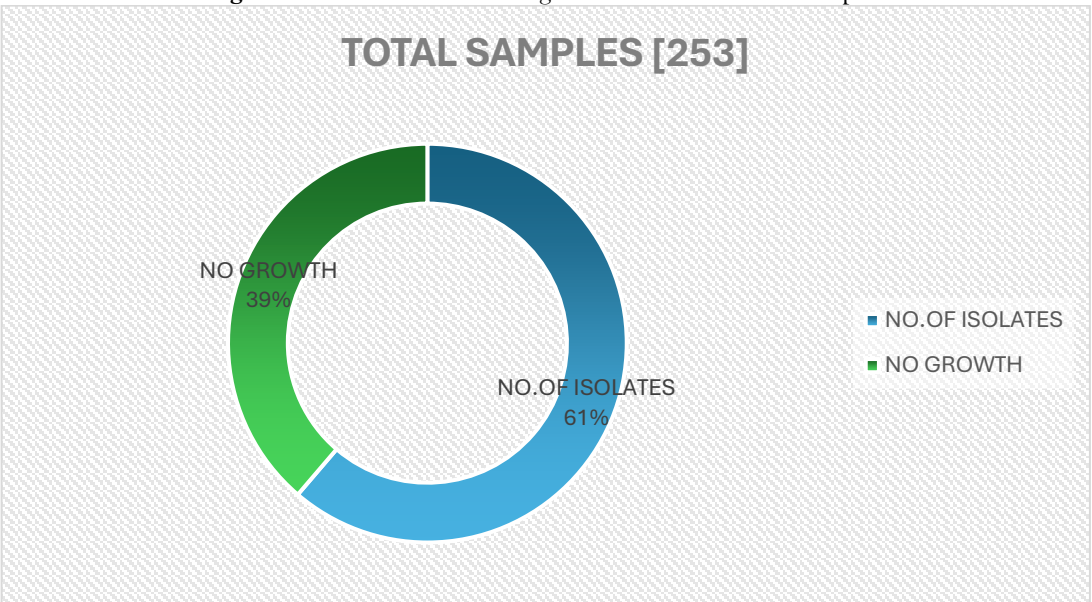

Table 2: Frequency of Bacterial isolate

Bacterial isolates	**No. of isolates [n=155]**	**Percentage**
Gram positive bacteria		
Staphylococcus aureus	17	10.97%
Staphylococcus other than staphylococci	15	9.68%
Gram negative bacteria		
Klebsiella spp.	56	36.13%
Escherichia coli	30	19.35%
Proteus spp.	15	9.68%
Pseudomonas aeruginosa	10	6.45%
Acinetobacter spp.	10	6.45%
Citrobacter spp	2	1.29%

Table 3: Antibiotic Sensitivity Pattern of Gram-Positive Cocci

Organism	**Ciprofloxacin**		**Clindamycin**		**Co-trimoxazole**		**Erythromycin**		**Tetracycline**		**Vancomycin**	
	S	%	**S**	%	**S**	%	**S**	%	**S**	%	**S**	%
Staphylococcus other than staphylococcus aureus [N=15]	4	26.7	1	6.7	10	66.7	8	53.3	10	66.7	15	100
MRSA [N=10]	5	50	5	50	7	70	5	50	4	40	10	100
Staphylococcus aureus [N=7]	3	42.8	7	100	7	100	6	85.7	5	71.4	6	85.7

Table 4: Antibiotic Sensitivity Pattern of Gram-Negative Bacilli

Organism	**Klebsiella spp. [N=56]**		**Escherichia coli [N=30]**		**Proteus spp. [N=15]**		**Pseudomonas aeruginosa [N=10]**		**Acinetobacter spp. [N=10]**		**Citrobacter spp [N=2]**	
	S	%	**S**	%	**S.**	%	**S.**	%	**S.**	%	**S**	%
Amikacin	46	82.1	20	66.7	10	66.7	7	70	10	100	2	100
Amoxicillin-Clavulanic acid	19	33.9	10	33.3	8	53.3	5	50	8	80	1	50
Ciprofloxacin	36	64.2	10	33.3	8	53.3	6	60	8	80	2	100
Ceftriaxone	29	51.7	10	50	10	66.7	7	70	10	100	2	100
Piperacillin-tazobactam	47	83.9	20	33.3	10	66.7	8	80	10	100	2	100
Meropenem	49	87.5	20	66.7	12	80	6	60	10	100	2	100
Cefoperazone-sulbactam	35	62.5	10	33.3	11	53.3	7	70	9	90	2	100
Co-trimoxazole	24	42.8	10	66.7	8	66.6	8	80	8	80	2	100
Gentamicin	24	42.8	15	53.3	10	53.3	6	60	10	100	2	100

DISCUSSION

Diabetic foot ulcers (DFUs) are one of the most common clinical outcomes that need treatment in people with diabetes. Diabetic foot infection is the term used to describe any inframalleolar infection in a diabetic. These include abscesses, cellulitis, paronychia, septic arthritis, necrotising fasciitis, osteomyelitis, and tendinitis. [11]
Gram-positive cocci showed 20.65% and Gram-negative bacilli showed 79.35% of the 155 isolates in this study. This was comparable to other studies and also contrasted in a few of them.
Similar to a small number of other research, the majority of the infections in our investigation were monomicrobial. According to several research, Klebsiella spp. is the most frequently isolated of them. [11]

Antibiotic Sensitivity Pattern of Gram-Positive Cocci:

Similar to other studies, all of the Gram-positive cocci isolated from diabetic foot ulcers in our study demonstrated high sensitivity to vancomycin and linezolid. [11-12]

Staphylococcus aureus

It is discovered that *S. aureus* had a sensitivity pattern to ciprofloxacin, clindamycin, co-trimoxazole, erythromycin and tetracycline of 26.7%, 6.7%, 66.7%, 53.3% and 66.7% respectively. Reduced sensitivity to clindamycin (6.7%) and ciprofloxacin (26.7%) was seen in this investigation. In another study, the sensitivity patterns of clindamycin, ciprofloxacin, erythromycin were determined to be 86%, 71.4% and 38.1% in contradiction with our study. [13]

Using cefoxitin discs, MRSA detection was found to be 58.8%. MRSA isolates shown 100% susceptibility to vancomycin in our study and this is same as in other studies. [12]

Staphylococcus other than staphylococcus aureus (SOSA)

SOSA had a sensitivity pattern to ciprofloxacin, clindamycin, co-trimoxazole, erythromycin and tetracyline of 26.7%, 6.7%, 66.7%, 53.3% and 66.7% respectively. A study is found to be in contrast to our study, with SOSA demonstrating 100% sensitivity to 75% sensitivity to erythromycin, and 50% sensitivity to ciprofloxacin. [13]

Antibiotic Sensitivity of Gram-Negative Bacilli

Klebsiella species

The results for *Klebsiella species* (*Klebsiella pneumoniae* and *Klebsiella oxytoca*) showed that the sensitivity of Amikacin is 82%, Amoxicillin-Clavulanic acid is 34%, Ciprofloxacin is 64%, Ceftriaxone is 52 %, Piperacillin-tazobactam is 84%, Meropenem is 88%, Cefoperazone- sulbactam 62%, Co-trimoxazole 43% and to Gentamicin the sensitivity is 42.8% This was same as in few other studies. [12-13]

Escherichia coli

E. coli in this study was 67% sensitive to amikacin, 33% sensitive to ciprofloxacin, and the same 33% sensitive to Amoxicillin-clavulanic acid, to ceftriaxone and to Cefoperazone- sulbactam. For Gentamicin the sensitivity rate of 50% was seen in this investigation while comparing with other studies, the sensitivity pattern of E. coli in our study seems to be contrast. [14]

Proteus species

In our study, Proteus spp. (*Proteus mirabilis* and *Proteus vulgaris*). exhibited 66.7% sensitivity to amikacin and to piperacillin-tazobactum. Also, with 53.3% sensitivity to ciprofloxacin and 66.7% to ceftriaxone and 53.3% of sensitivity to gentamicin. Proteus species also showed nearly 80% sensitivity to Meropenem and this was similar to few other studies. [12-13]

Pseudomonas aeruginosa

The sensitivity of piperacillin-Tazobactum was 88%, while that of ciprofloxacin is 60% and amikacin was 70%. Cefoperazone/Sulbactam is 70% and gentamicin is 60% in our study. The sensitivity of ceftriaxone is 70%,

Sensitivity of meropenem is 60%. Amoxicillin-clavulanic acid was sensitive to 50% of isolates. These results are contradictory to few other studies. [12-14]

Acinetobacter species

100% sensitivity to amikacin was shown by *Acinetobacter species* in our study and 80% to ciprofloxacin, 100% to gentamicin is observed. Amoxicillin-clavulanic acid was sensitive to 80% of Acinetobacter spp. The sensitivity pattern to Meropenem was 55% and Ampicillin was 100%, our results are correlating to other studies. [15]

Citrobacter spp.

Interestingly *Citrobacter spp.* showed 100% sensitivity to Amikacin, Ciprofloxacin, Ceftriaxone, Piperacillin-tazobactam, Meropenem, Cefoperazone- sulbactam, Co-trimoxazole and to Gentamicin. The sensitivity patter of amoxicillin-clavulanic acid was 50%.

CONCLUSION

Multidrug-resistant organisms are becoming more common in diabetic foot infection patients, and their microbial infection patterns are unpredictable. Continuous monitoring is therefore necessary to guide empirical treatment and reduce the possibility of negative outcomes. Unintentionally using wide spectrum antibiotics is best avoided. When selecting an antibiotic course of therapy, the most common isolated organisms and their patterns of antibiotic susceptibility should be taken into account. In this instance, the narrow spectrum antibiotic treatment must be initiated using the reports and subsequent microbiological tests. This will improve and promote the widespread use of antibiotics while lowering the development of bacteria resistant to a number of medications.

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Conflict of interest

The authors declare that there is no conflict of interest.

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