

A STUDY ON BACTERIOLOGICAL AND MYCOLOGICAL PROFILE OF DIABETIC FOOT ULCER PATIENTS ATTENDING IN GAUHATI MEDICAL COLLEGE & HOSPITAL



Microbiology

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ABSTRACT

Introduction: Diabetes mellitus is a chronic metabolic disease known for chronic complications such as peripheral vascular disease, cardiovascular disease, nephropathy, retinopathy, neuropathy and infections. The objectives of the study are to determine the Bacteriological and Mycological profile of infected diabetic foot ulcers and to study the Antibiotic susceptibility pattern of the bacterial isolates.

Materials and methods: A hospital based cross-sectional study was carried out in a tertiary Care Hospital of Assam between the periods of June/2018 to May/2019 in the department of Microbiology.

Results: 92.31% sample showed growth of Aerobic bacteria; however, anaerobic bacterial growth was not detected in any of the samples. Fungus was isolated in 15.38% samples. 79.17% patients had monomicrobial infections and 20.83% patients had polymicrobial infections.

Conclusions: Both Gram positive cocci and Gram negative bacilli caused diabetic foot infections and this study showed a preponderance of Gram negative bacilli.

KEYWORDS

Diabetes, Bacteriological, Mycological, Antibiotic

INTRODUCTION:

Diabetes mellitus is a chronic metabolic disease which affects a large part of the human population and also the main endocrine cause of morbidity and mortality all over the world.⁽¹⁾ Globally, an estimated 425 million adults were living with diabetes in 2017, compared to 108 million in 1980. Low and middle income countries carry almost 80% of the diabetes burden. India today leads the world with its largest number of diabetic subjects in any given country. It is said that presently 62 million Indians are diabetics and these numbers are likely to increase to 89.4 million by the year 2025.^(2,3)

Diabetes mellitus is a disease known for chronic complications such as peripheral vascular disease, cardiovascular disease, nephropathy, retinopathy, neuropathy and infections. Among them diabetic foot infection is one of the devastating complications. Uncontrolled hyperglycemia, atherosclerotic vascular disease, sensory neuropathy are the most important risk factors developing diabetic foot ulcer. In addition to causing severe morbidities, they now account for the largest number of diabetes-related hospital bed-days and are the most common proximate, non-traumatic cause of amputations. Foot infections are among the most common bacterial infections encountered in patients with diabetes mellitus in clinical practice.^(4,5)

Diabetic patients are at greater risk for bacterial and fungal infections and also have an increased susceptibility to develop skin and soft tissue infections.⁽⁶⁾ It is essential to assess the magnitude of bacterial and fungal infection of the lesions to avoid further complications and save the diabetic foot.⁽⁷⁾

Since Most of the diabetic foot infections are of mixed type, so the proper management of these infections require an appropriate antibiotic selection based on the culture and the antimicrobial susceptibility testing results.⁽⁷⁾ In view of the that, a hospital based cross-sectional study was carried out with the following.

AIMS AND OBJECTIVES

- To determine the Bacteriological and Mycological profile of infected diabetic foot ulcers.
- To study the Antibiotic susceptibility pattern of the bacterial isolates.

METHODS:

The present study was conducted in a tertiary Care Hospital of Assam between the period of June/2018 to May/2019 in the department of Microbiology. Approval from the Institutional Ethical Committee was

obtained prior to initiation of the study. Wound swab (two from each patient) and pus samples were collected from 104 diabetic foot ulcer patients attending surgery department using a sterile disposable swab following thorough cleaning to avoid contaminations and after taking written informed consent of the patients. Samples were immediately processed as per standard guidelines in Microbiology department.

One part of the sample was inoculated into 5% Sheep blood agar and MacConkey agar for isolation of aerobes and facultative anaerobes, One part was inoculated into the Sabouraud's dextrose agar (SDA) media with chloramphenicol (5%) for isolation of fungus and last part of the specimen was used to make smear for direct microscopy. Isolates were identified and confirmed based on colony morphology, Gram stain morphology and biochemical properties.

Antibacterial susceptibility testing was done according to Kirby Bauer method using Mueller Hinton Agar as per CLSI guidelines.⁽⁸⁾ Detection of ESBL was done phenotypically by screening method followed by confirmation with standard disc diffusion method. MRSA was detected with 30 microgram cefoxitin disc by Kirby Bauer disc diffusion method. Quality control was performed using *Pseudomonas aeruginosa* ATCC strain 27853 for *Pseudomonas aeruginosa* and *Staphylococcus aureus* ATCC strain 43300 for MRSA detection.⁽⁸⁾

RESULTS:

Among 104 diabetic foot ulcer patients, the age of the study population ranged from 30-79 years with a median age of 55.97 years. Of them, 84% were males and 16% were female with a male: female ratio of 5:1. 95.19% patients had type 2 diabetes mellitus whereas only 4.81% were type 1 diabetes mellitus patients. Great toe (39.42%) was the most affected site of ulcer followed by Dorsum (32.69%) of foot. In the present study 96 (92.31%) sample showed growth of Aerobic bacteria; however, anaerobic bacterial growth was not detected in any of the samples. Fungus was isolated in 16 (15.38%) samples. 76 (79.17%) patients had monomicrobial infections and 20 (20.83%) patients had polymicrobial infections.

The most common bacteria isolated was *Pseudomonas aeruginosa* 31 (29.81%) followed by *Staphylococcus aureus* 23 (22.12%), *Klebsiella pneumoniae* 18 (17.31%), *Proteus mirabilis* 12 (11.54%), *Escherichia coli* 10 (9.62%), *Enterococcus faecalis* 3 (2.88%) and *Enterobacter aerogenes* 3 (2.88%). (Figure 1)

Among the fungal isolates, 87.5% were *Candida albicans* and 12.5% were *Fusarium solani*.

Antimicrobial Susceptibility test revealed that most effective antibiotic among the Gram negative bacilli was Polymyxin B followed by Meropenem, Imipenem etc (Figure-2)) and Linezolid, Teicoplanin and Clindamycin etc were the most effective antibiotic for the Gram positive cocci (Figure-3). Among the Gram negative bacilli, 50% of the *Escherichia coli* and 55.56% of the *Klebsiella pneumoniae* were ESBL producers. MRSA was detected in 17.39% of the *Staphylococcus aureus*. 4% strains were detected as Multidrug resistant strains.

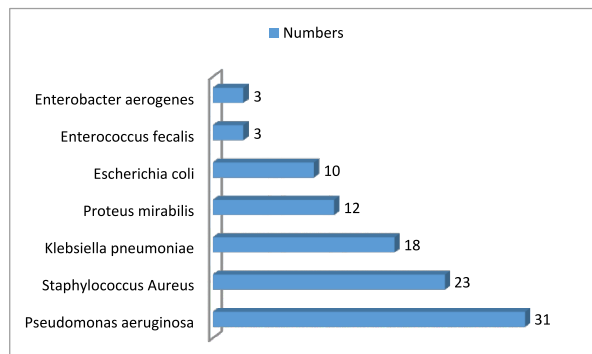


Fig1: Distribution of bacterial isolates associated with diabetic foot ulcer (N=104)

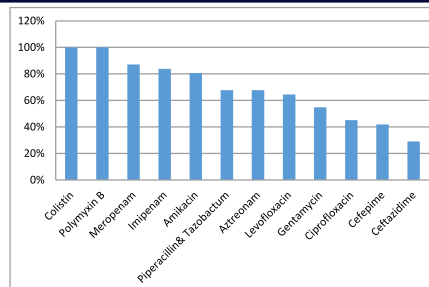


Fig 2: Antimicrobial susceptibility pattern of Gram negative bacilli

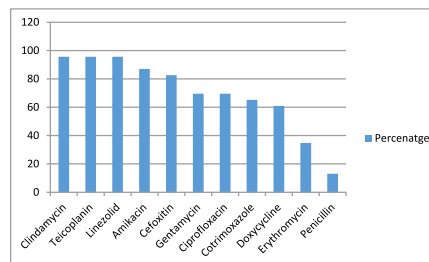


Fig3: Antimicrobial susceptibility pattern of Gram positive cocci

Table 1: Comparison of various bacterial isolates

Study	<i>P. aeruginosa</i>	<i>S. aureus</i>	<i>K. pneumoniae</i>	<i>P. mirabilis</i>	<i>E. coli</i>	<i>E. faecalis</i>	<i>E. aerogenes</i>
Present study	29.81%	22.12%	17.31%	11.54%	9.62%	2.88%	2.88%
Mehta VJ <i>et al</i> ⁽⁹⁾	34.70%	75%	27.70%	8.33%	23.60%	15%	0
Sanjith Saseedharan, <i>et al</i> ⁽¹⁰⁾	20.9%	26.9%	0	0	12%	12.7%	0
Priyadarshani S <i>et al</i> ⁽¹¹⁾	16%	13.3%	8%	6.6%	14.6%	5.3%	5.3%

DISCUSSION:

In the present study mean age of patients were 55.97 years. Raza M *et al*⁽¹²⁾, Malepati S *et al*⁽¹³⁾ also observed almost similar findings in their studies. Male preponderance was observed in the present study. Malepati S *et al*, Raza M *et al*, Anandi C *et al* also observed more males than females in their studies which is in concordance to the present study.^(7,12,13) This could be explained by the fact that men have more outside activity than have women, which may lead to more foot exposure to different risks and more plantar pressure on their feet.

In the present study, 96 (92.31%) specimens yielded bacterial growth out of which 20(20.83%) showed polymicrobial growth with predominance of Gram negative bacilli. Shashanka R *et al*, Priyadarshani S *et al* in their study also showed Gram-negative bacilli were more prevalent than Gram-positive cocci in diabetic foot.^(11,14)

Various studies have reported multiple bacterial isolates associated with diabetic foot ulcers. (Table1). In the present study, among the bacterial isolates *Pseudomonas aeruginosa* (29.81%) was predominant followed by *Staphylococcus aureus* (22.12%).

Fungus was isolated from 15.38% diabetic foot ulcer patients in the present study. Out of this 87.5% were *Candida albicans* and 12.5% were *Fusarium solani*. Raza M *et al*⁽¹²⁾ also found *Candida* (26.32%) as the most common fungal isolates whereas Wijesuriya *et al*. in Sri Lanka reported *Aspergillus niger* as the most common fungal isolates.⁽¹⁵⁾

MRSA was found to be 17.39% in the present study whereas Priyadarshani S *et al* and Sugandhi P *et al* found 8% and 50% MRSA respectively in their study^(11,16).

ESBL production was seen in 50% *Escherichia coli* isolates in the present study. Babypadmini *et al*.⁽¹⁷⁾ have shown 41% of *Escherichia coli* isolates to be ESBL producers in their study. Motta *et al*.⁽¹⁸⁾ found prevalence of ESBL was only 6% among *E. coli* isolates.

In the present study 4% isolates were multidrug resistant strains which were not tested for presence of metallo beta lactamase (MBL) enzyme but its presence can't be ruled out among these isolates.

Both Gram positive cocci and Gram negative bacilli caused diabetic foot infections and this study showed a preponderance of Gram negative bacilli. There was a variation in the bacterial aetiologies of the DFIs, based on the geographical location. Knowledge on the antibiotic susceptibility pattern of the isolates from diabetic foot infections is crucial for planning the appropriate treatment of these cases, prior to getting the susceptibility reports from the laboratory. Appropriate usage of antibiotics based on local antibiogram pattern can certainly help the clinician in reducing the burden of diabetic foot infections.

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CONCLUSIONS: