

Original Research Article

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A Study of Bacterial Culture Isolates and Antibiogram in Diabetic Foot Ulcer in GGG Hospital, Jamnagar, Gujarat, India

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ABSTRACT

A diabetic foot is common complications of diabetes and cause of the hospitalization. Major increase in mortality among diabetic patients, observed over the past 20 years is considered to be due to the development of macro and micro vascular complications, including failure of the wound healing process. The individuals with diabetes have at least a 10-fold greater risk of being hospitalized for soft tissue and bone infections of the foot than individuals without diabetes. This study was carried out to determine the bacteriological profile of infected diabetic foot ulcers and their antibiotic sensitivity pattern. Total 280 pus samples were collected from diabetic foot by using sterile swabs and they were processed. Study was carried out over the period of 5 months from January 2014 to may 2014 in microbiology department, Shri M.P. Shah Government medical College, GGGH, Jamnagar. A total of 77(27.5%) bacterial culture isolates were obtained from 280 samples with diabetic foot. Gram negative bacilli were more prevalent (87.02%) than gram positive cocci (12.98%). The antibiogram (antibiotic sensitivity pattern) of the bacteria were also studied. Gram negative bacilli were sensitive to Piperacillin+Tazobactam (79.10%), Amikacin (40.29%), Imipenem (38.80%), Netilmicin (28.35%) Gentamicin (17.91%), Cefoperazone (14.92%), Ceftazidim (4.47%). Gram positive cocci were sensitive to Linezolid(100%), Vancomycin (70%), Clindamycin and Tetracycline (50%), Azithromycin and Gentamicin (30%), Cotrimoxazole (20%), Cephalaxin(10%). Knowledge on the antibiogram of the isolates will be helpful in determining the antibiotics for the treatment of diabetic foot ulcers rather than prescribing the empirical antibiotics.

Keywords

Bacterial culture isolates, antibiogram, diabetic foot ulcer.

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Introduction

Diabetes mellitus is a metabolic disorder present with chronic hyperglycaemia and disturbances in carbohydrate, fat and protein metabolism. It is due to defects in insulin secretion, action or both. It manifested as damage or dysfunction in various organs like retina, kidney, Cardio vascular system,

Cerebro vascular system, bone, joints & soft tissue like foot ulcer. It also includes physiological disturbances like sexual dysfunction. The major contributory factors for the high prevalence of the complications are; delayed diagnosis of diabetes, inadequate control of glycaemia,

hypertension, and lack of awareness about the disease among majority of the public (Yashpal *et al.*, 2012).

Diabetes mellitus is more prevalent within increasing age specially middle age and elderly people (Park *et al.*, 2009).

Diabetic foot is infection, ulceration and/or gangrene of foot due diabetes mellitus. It is actually a triad of neuropathy, ischaemia and infection which required hospitalisation and prolonged stay. The presence of infection rapidly worsens the clinical picture often requiring limb amputation. Diabetic foot ulcers are common and estimated to affect 15% of all diabetics. In India, it is estimated that approximately 40,000 legs are being amputated every year, of which 75% are neuropathic feet which are potentially preventable (Yashpal *et al.*, 2012, Bhavin Dalal *et al.*, 2003). This study was carried out to determine the bacteriological profile in diabetic foot ulcers and their antibiotic sensitivity pattern.

Materials and Methods

Study duration & sample size

Total 280 pus samples were studied over the period of 5 months from January 2014 to May 2014 in Microbiology department, M.P. Shah government medical College, GGGH, Jamnagar.

Pus samples were collected by using sterile swabs and they were processed. After obtaining proper patient informed consent, samples were collected from the deeper portion of the ulcers (ulcer base) by using 2 sterile swabs. The samples were collected by making a firm, rotatory movement with the swabs. One swab was used for Gram staining and the other was used for culture. A direct Gram stained smear of the

specimen was examined. The specimens were inoculated onto blood agar, chocolate agar, Mac Conkey's agar. The inoculated plates were incubated at 37°C overnight and the plates were examined for growth on the next day. The further processing was done according to the nature of the isolate, as was determined by Gram staining, culture characteristic and their biochemical reactions.

The antibiogram (antibiotic sensitivity pattern) of the bacteria were also studied by the Modified Kirby Bauer disc diffusion method by using Mueller Hinton Agar according to CLSI (Clinical and Laboratory Standard Institute) guideline.

The antimicrobial discs which were used were those of Piperacillin/Tazobactam (100+10mcg), Amikacin (30mcg), Netilmicin (30mcg), Imipenam (10mcg), Ciprofloxacin (5 mcg), Cefoperazone (30mcg), Gentamicin (10 mcg) for the Gram negative bacilli. Vancomycin (30mcg), Linezoli (30mcg), Azithromycin (30mcg), Clindemycin (2mcg), Tetracycline (30mcg), Cotrimoxazole (25mcg), Gentamicin (10mcg) were used to study the susceptibility patterns of the Gram positive cocci .

Inclusion and exclusion criteria

Total two swabs were collected in each & every patients. One was tested by direct gram staining and results of direct gram staining were excluded from study while second was tested by culture method and results were included in study.

Ethical clearance

It was a retrospective analysis of samples collected for routine diagnosis, so ethical consideration was not necessary.

Results and Discussion

A total of 77 (27.5%) bacterial isolates were obtained from 280 pus samples with diabetic foot as shown in table 1.

The study included 57 (74.01%) males and 20 (25.99%) females .The ages ranged from 20-80 years. Most numbers of foot infections were seen between the age group of 51-60 years followed by 41 to 50 years as shown in table 2.

Gram negative bacilli 67 (87.02%) were more prevalent than gram positive cocci 10 (12.98%) as shown in table 3.

A total of 67(87.02%) Gram negative bacilli were isolates among which *Pseudomonas* spp. 43 (55.86%) was the commonest organism followed by *Proteus* spp. 8 (10.38%), *Klebsiella* spp. 6 (7.79%), *Acinetobacter* spp. 6(7.79%) and *E-coli* 4(5.19%). A total of 10(12.98%) Gram positive cocci were isolates among which *Staphylococcus aureus* 5 (6.5%) was the commonest organism followed by *Enterococcus* spp 4 (5.19%) and Coagulase Negative *Staphylococcus aureus* 01 (1.29%).

In present study gram negative isolates were more sensitive to Piperacillin + Tazobactam 79.10% followed by Amikacin 40.29%, Imipenem 38.80%, Netilmicin 28.35% and Gentamicin 17.91% as shown in figure 1.

In present study gram positive isolates were more sensitive to Linezolid 100% followed by Vancomycin 70%, Clindamycin and Tetracyclin 50%, Azithromycin and Gentamicin 30 %, Cotrimoxazole 20% as shown in figure 2.

In present study total of 77 bacterial isolates were obtained from 280 pus samples with diabetic foot as shown in table-1. The bacterial culture positivity rate is 27.5% in our study which is lower than the study of Jayshree *et al* 38% (Jayshree Konar *et al.*, 2013).

Rate of diabetic foot infections in male was 74.01% in current study as shown in table-2. Almost same result has been identified in study of Pathare *et al.*, with 78.5% positivity rate in male (Mendes *et al.*, 2012). This could because diabetes mellitus is more common in male and male are prone to trauma because of their outdoor occupation.

Table.1 Total positive cases.

| +Ve (%) | -Ve (%) | Total (%) |
|------------|------------|------------|
| 77 (27.5%) | 203 (72.5) | 280 (100%) |

Table.2 Age and sex distribution

| Age range | Male (%) | Female (%) | % (n=77) |
|--------------|-------------------|-------------------|-----------------|
| 20-30 | 03 (3.89) | 01 (1.3) | 04 (5.19) |
| 31-40 | 03 (3.89) | 02 (2.60) | 05 (6.49) |
| 41-50 | 15 (19.49) | 06 (7.80) | 21 (27.29) |
| 51-60 | 29 (37.66) | 08 (10.39) | 37 (48.05) |
| 61-70 | 04 (5.19) | 02 (2.60) | 06 (7.79) |
| 71-80 | 03 (3.89) | 01 (1.3) | 04 (5.19) |
| Total | 57 (74.01) | 20 (25.99) | 77 (100) |

Table.3 Isolated Gram negative and Gram positive organisms

| Organisms no (%) (n=77) | Total (%) |
|--|------------------|
| Gram Negative Bacilli 67 (87.02%) | 77 (100%) |
| <i>Pesudomonas</i> spp 43 (55.86%) | |
| <i>Proteus</i> spp 08 (10.38%) | |
| <i>Klebsiella</i> spp 06 (7.79%) | |
| <i>Acinetobacter</i> spp 06 (7.79%) | |
| <i>Escherichia coli</i> 04 (5.2%) | |
| Gram Positive cocci 10 (12.98%) | |
| <i>Staphylococcus aureus</i> 05 (6.5%) | |
| <i>Enterococcus</i> 04 (5.19%) | |
| Coagulase Negative <i>Staphylococcus aureus</i> 01 (1.29%) | |

Fig.1 Antibiotic sensitivity pattern of gram negative isolates

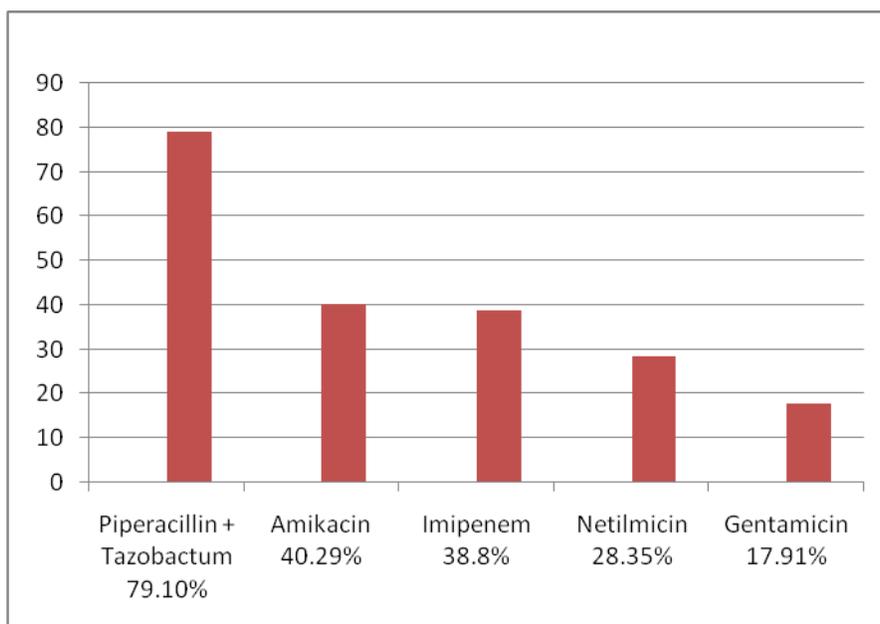
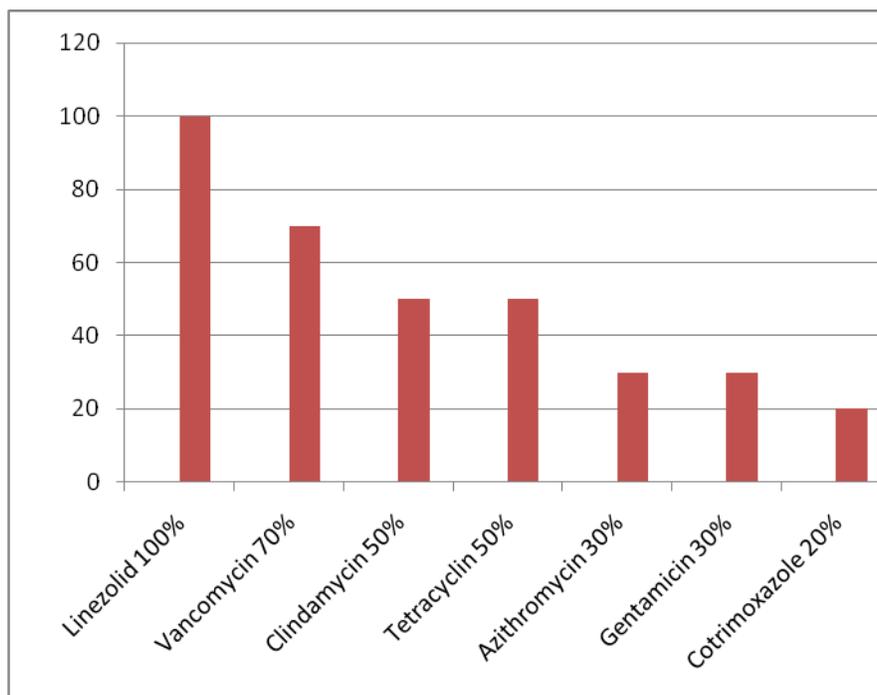


Fig.2 Antibiotic sensitivity pattern of gram positive isolates



In present study diabetic foot infections were more common in 51 to 60 years (37.66%) followed by 41 to 50 years (19.49%) as shown in table-2. The most common age group in study of Leela *et al.*, was 51 to 60 years (45.3%) followed by 41 to 50 years (31.33%) (Leela Rani *et al.*, 2013) which is little higher than current study. Both the study shows that diabetic foot infection cases are increasing with advancing age, exact reasons for this are unknown. but may be due to increasing diabetes with advancing age which favour the bacterial infection.

In present study GNB (87.02%) were more common than GPC (12.98%) as shown in table-3. High rate by GNB than GPC was also noticed by Gadepalli R *et al* with 51.4% participation by GNB & 33.3% participation by GPC (Gadepalli *et al.*, 2006). Most common bacteria among the GNB category was *Pseudomonas spp* (55.86%) while from GPC category it was *Staphylococcus aureus* (6.5%) in our study

as shown in table-3. Some research also suggest highest infection rate by *Pseudomonas spp* (30.67%) from GNB category and *Staphylococcus aureus* (17%) from GPC category in diabetic foot ulcer (Jain Manisha *et al.*, 2012; Vaidehi Mehta *et al.*, 2014).

Sensitivity pattern of present study suggest that all GNB isolates from diabetic foot ulcer were most sensitive to Piperacillin + Tazobactam (79.10%) and less sensitive to other tested drugs as shown in figure-1, while GPC from diabetic foot ulcer were most sensitive to Linezolid (100%) and less sensitive to other tested drugs as shown in figure-2. Research by Vaidehi *et al* indicates that GNB isolates from diabetic foot ulcer were also most sensitive to Piperacillin + Tazobactam but with sensitivity rate is almost more than half about 38% while GPC isolates shows same result with 100% sensitivity rate to Linezolid.

In conclusion, in present study, Gram negative bacilli were more predominant than

Gram positive cocci. Pseudomonas species and staphylococcus aureus were responsible for most of the Diabetic foot infections .Gram negative bacilli were most sensitive to Piperacillin+Tazobactam, and Gram positive cocci were most sensitive to Linezolid. Knowledge on the antibiogram of the isolates will be helpful in determining the antibiotics for the treatment of diabetic foot ulcers rather than prescribing the empirical antibiotics.

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