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BACTERIOLOGICAL PROFILE OF DIABETIC FOOT ULCER

Dr. K. LEELA RANI[1] , Dr. MADHUMATHI[1], Dr. SANDHYA BELWADI[2], Dr. B. V. RAMANA[3]

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1. Assistant Professor, Department of Microbiology, Vydehi institute of medical sciences and research centre Whitefield, Bangalore 66.
2. Professor, Department of Microbiology, Vydehi institute of medical sciences and research centre Whitefield, Bangalore 66.
3. Assistant Professor, Department of Microbiology, Sri Venkateswara Institute of Medical Sciences, Tirupati- 517 507, Andhra Pradesh, India.

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Corresponding Author

Dr. K. LEELA RANI

Abstract

Diabetic foot infection is an important complication in Diabetes mellitus. Despite recent medical advances in the treatment foot infection remains a major cause for morbidity and mortality in patients with this disorder. Proper management of these infections requires microbial isolation, appropriate or accurate selection of antibiotics. Understanding the bacteriology and accuracy of culture methods is important in selecting antibiotics in these patients **AIM**: To study the bacteriological profile of diabetic foot ulcers and to find out the antimicrobial susceptibility of those organisms **METHODS:** A total of 150 patients attending the medical college hospital at Tirupati , Andhra Pradesh over a period of 6 months. **RESULTS:** *Escherichia. coli* was the commonest organism followed by *Proteus. mirabilis, Pseudomonas aeruginosa , Klebsiella pneumoniae, Staphylococcus aureus*. Gram negative organisms were sensitive to Imipenem, Piperacillin/Tazobactum, Amikacin and third generation Cephalosporins. Gram positive organisms were sensitive to Vancomycin. **CONCLUSION:** Gram negative organisms were more common in diabetic foot ulcers and are sensitive to number of widely used antimicrobial agents.

INTRODUCTION:

Diabetes mellitus is a disease as old as mankind itself and is a major health care challenge. Diabetes mellitus confers a special vulnerability to infection due to defects in both cell mediated and humoral immunity, probably due to hyperglycemia. Once these infections occur, they are more difficult to treat and pose a great threat to the diabetic than to a healthy person. Foot infections are common and represent a serious problem in diabetic patients.[1]

Despite recent medical advances in the treatment of diabetes mellitus, foot infection remains a major cause for morbidity and mortality in patients with this disorder. Three main factors are responsible for this: neuropathy, angiopathy and immunopathy. Neuropathy is probably the most important factor: minor irritations and trauma can lead to limb threatening infections without the patient feeling the changes. Angiopahty plays only a minor role, while immunopathy has implications for antibiotic treatment, in that bactericidal agents are needed.[2]

Proper management of these infections requires microbial isolation, appropriate or accurate selection and identifications of complications that require surgical intervention as over half of these patients need amputations3. Understanding the bacteriology and the accuracy of the culture methods is important in selecting antibiotics in these patients. In India, studies on the microbiology of diabetic foot infections are scanty and the magnitude of this problem hence goes unnoticed. Further studies are necessary to assess the role of bacteria in this etiology and to develop a more appropriate treatment regimen. The present study was, therefore, undertaken to assess the role of the aerobic bacteria as pathogens and the antimicrobial susceptibility of the aerobes isolated.

MATERIALS AND METHODS: The present study was carried out in the Department of Microbiology, S. V. Medical College, Tirupati. A total 150 diabetic patient with foot ulcerations were studied. Criteria for inclusion in the study were: presence of foot infection in the diabetics of grade I and above and hospitalized patients.

Samples were collected in the surgical wards where the dressing was being done. The ulcer was cleaned with sterile normal

saline and the surrounding area was cleaned with 70% alcohol. Debris, dead and devitalized tissue overlying the ulcer was removed using a sterile forceps and scissors. Swabs were collected from the depth of the ulcers on the feet of the diabetic patients. From each patient, two swabs were collected. One swab was used for the isolation of aerobic bacteria and the other for preparation of smear for Gram stain.

All the samples collected were immediately brought to laboratory and then processed. They were processed as follows: a). One of the two swabs was used for direct smear examination by Gram stain to look for the presence of pus cells and bacteria.b). Aerobic culture – For the isolation of aerobic bacteria, the sample was inoculated onto 5% sheep blood agar and MacConkey agar and incubated at 37C for 24 hours under aerobic conditions.

The morphology of the colonies on Blood agar, and MacConkey agar were studied and smears from the colonies were prepared and stained with Gram's stain. Organisms were further processed with relevant biochemical tests as per standard methods. The antibiotic susceptibility testing of the isolates was done on Mueller Hinton agar using Kirby-Bauer disc diffusion method. The antibiotic discs Ampicillin (10mcg), Gentamycin (10mcg), cotrimaxazole(25mcg) , Netilmycin(30mcg), Ofloxacin(5mcg), Amikacin(30mcg), Erythromycin(15mcg), Cefatoxime(30mcg), Ceftazidime(30mcg) , Ceftriaxone(30mcg) Imepenem(10mcg) piperacillin(100mcg) piperacillin\tazobactum100/10mcg) and Clindamycin(2mcg) (obtained from Hi-Media Laboratory) were used .

RESULTS: A total of 150 cases of foot infected among diabetics were included in the present study .The study includes 107 males and 43 females .The ages ranged from 30-85 years. Most number of foot infections were seen between the age group of 51-60 years . Of the 216 isolates, 28 isolates are Gram positive cocci, accounting for 12.96% of the isolates and 188 isolates (87.03%) were Gram negative bacilli.

Out of 150 cases samples from 13 patients (8.66%) cultures were negative, while from 84 patients (56%) cultures revealed polymicrobial aetiology. The most frequent

organism isolated was *Escherichia. coli*(25.46%), followed by *Proteus. mirabilis*(23.14%), *Pseudomonas aeruginosa*(13.42%), *Klebsiella pneumoniae*(12.03%), *Staphylococcus. aureus*(10.64%).The other species were *Proteus. vulgaris*(6%), Enterobacter cloacae(3.7%), *Enterococcus faecalis*(1.85%), *Citrobacter freundii* (1.85%), *Morganella morganii*(1.38%), and *Streptococcus pyogenes*(0.46%).

DISCUSSION: The feet of diabetic patients are prone to soft tissue lesions. It is important for the treating physician to recognize that the appearance of a diabetic foot ulcer on the plantar surface does not mean that an infection is present. The aetiology of diabetic foot ulcers is multifactorial, with mechanical factors playing a large role.[3] In the absence of ischaemia, the vast majority of diabetic foot ulcers are the result of increased pressure on the soft tissues. Diabetic foot infections are limb threatening and life threatening illness in the diabetic patients. Diabetic foot infections are also the major cause for non traumatic limb amputations. Diabetic foot infections not only lead to pain, suffering and disability but also to prolonged hospitalization hampering both mental and economic status of the patients, leading to great expenses to the patient as well as the community.

In the present study, diabetic foot infections were common in men (71.33%) compared to study of Pathare *et al* (78.5%), Sapico *et al* (61.5%) and Anandi *et al* (65.4%). [4 5 6]This could because diabetes mellitus is more common in men and are prone for trauma because of their outdoor occupation. In our study diabetic foot infections were more common in 40-60 year age group accounting for 70% of the total cases. Which was similar to the study of Pathare *et al* (40-60 years) and Anandi *et al* (43 years). [4 5 6] Out of 150 cases studied, 98.66% were of non-insulin dependent diabetes mellitus (NIDDM) type as compared to Pathare *et al* (96%) and Ravishekar *et al* (88.8%) . In the present study 53.6% of the cases were suffering from diabetes mellitus for more than 6 years sapico et al reported that 61.5% of the patients had diabetes mellitus for more than 5 years.

Enterococus species were isolated from 4 of our cases (1.85%). Most of the

other studies recovered Enterococus species in a range of 4 - 30%. Gram negative bacilli were the most common organisms isolated in the present study. Among the 188 (87.03%) of the aerobic Gram negative pathogens. Escherichia coli was the commonest isolate accounting for 25.46% of the total isolates which is similar to the study of Anandi 5 etal (27.7%).[5] The next common isolates in the present study were *Proteus mirabilis* (23.14%),similar to the study of the Anandi *et al* (16.9). *Pseudomonas aeruginosa* (13.42%), similar to the study of Anandi etal (11.3%) and Ravishekar etal (9.8%). *Klebsiella pneumoniae* (12.03%) similar to the study of Pathare *et al* (14.1%) and Anandi *et al* (16.9%).

The other Gram negative bacilli isolated in the present study were *Proteus vulgaris (6%). Citrobacter freundii (1.85%). Enterobacter species (3.7%) Morganella morganii (1.38%)*. All these isolates have been reported by most of the workers. An empiric /specific therapy is an important factor in the management of diabetic foot infections. Extended spectrum cephalosporins have been used with good results for treatment of foot infections in diabetic patients. In patients who appear severely ill or having necrotising infections broader antibiotic coverage is indicated.

In the present study, Staphylococcus aureus was most sensitive to Vancomycin (100%) 8 Clindamycin cefotaxime (65.2%) Ceftazidime (56%) C (52.21%) Clindamycin (34.78%) Gentamicin (34%) and Ampicillin (34%) Ofloxacin (34%) and Amikacin (30.4%).The present study showed higher rate of resistance among most of the isolates. This could be because most of the patients had received treatment primarily which could have eliminated the sensitive organisms and left out only the resistant ones.

Enterococcus species isolated in the present study showed higher resistance. They showed sensitivity to Vancomycin(100%) Amikacin (50%) Gentamycin (25%) Netilmycin (25%) Ofloxacin (25%) Cefotoxime (25%) Ceftazidime (25%) Ceftrioxone (25%). The Gram negative bacilli isolated in the present study were most sensitive to imepenem,Piperacillin /Tazobactum,Piperacillin,Amikacin, Cefotaxime, Ceftazidime, Ceftriaxone, and Gentamycin, Ofloxacin, *Escherichia coli*,

which was most commonest isolate (25.46%) of all the isolates and was most susceptible to Imepenem(96%) Piperacillin/Tazobactum(90%) ,Cefatoxime (76.6%),Piperacillin(72%) Amikacin (65.45%) Ceftriaxone (54.5%) Ceftazidime (49%) Gentamycin (30.9%) and Ofloxacin (30.9%) Proteus mirabilis isolates demonstrated Imepenem 98% sensitivity,Piperacillin/Tazobactum90%, 60% sensitivity to Cefotaxime, 58% sensitivity to Ceftriaxone and 90% to Amikacin. Sensitivity of *Proterus vulgaris* is 79% to Amikacin and 38.46% to Cefotaxime and showing maximum resistance to other routinely used antibiotics.[7] Enterobacter species sensitivity to Ofloxacin (87.5%) Cefatoxime (75%) Amikacin (75%) Ceftazidime (62.5%) Gentamycin (50%) Ceftriaxone (50%). Of the 4 isolates of *Citrobacter freundii* sensitivity to Cefotaxime is (50%) and Ceftriaxone is (50%). Of the 3 isolates of Morganella morganii were sensitivite to Cefatoxime (66%) and to all other antibiotics was (33%). Pseudomonas sensitivity to Imepenem93% Piperacillin/Tazobactum 83%, Piperacillin 83%, Amikacin is (55%) and less sensitive to other routinely used antibiotics.[8]

CONCLUSIONS: From these findings the necessity of in patient management of the diabetic foot infections that could be treated effectively with antibiotics will come into vouge. Just prior to onset of antibiotics therapy sample should be collected. The Gram stain will guide us to the possible pathogens causing the infection and aid in the commencement of therapy until the antibiotics susceptibility report is available. Therefore culture results are of value in directing antimicrobial selection to better match the antibacterial susceptibility of the organisms. The collection of a sample prior to onset of antibiotic therapy is a must with the primary gram staining of the direct smear guiding both the microbiologist and clinician on the predominant microbial flora and enabling in the commencement of empirical antibiotic therapy until the culture and sensitivity report is made available to further enable in the specific treatment and management. Last but not the least patient education is the most important aspect. Once a patient is diagnosed as diabetic, it is the doctor's responsibility to enlighten the patient regarding the foot care in diabetic

and making him/her to understand the consequences of foot infections.

TABLE 1

AGE DISTRIBUTION OF THE STUDY GROUP

Age range	No. of patients	% of patients
20-30	1	0.66
31-40	7	4.66
41-50	47	31.33
51-60	68	45.3
61-70	21	14
71-80	5	3.3
81-90	1	0.66

TABLE 2

TYPE OF DIABETES

Type of Diabetes	N = 150	%
I	2	1.33
II	148	98.66

TABLE 3

SEX DISTRIBUTION OF PATIENTS

Males	Females
107	43
71..33%	28.66%

TABLE 4

Antibiotic susceptibility pattern of Gram negative isolates:

Antibiotic	P.aeuriginosa(29)	P.mirabilis(50)	P.vulgaris(13)	K.pneumoniae(26)	E.coli(55)	Enterobacter cloacae(8)	M.morganii(3)	C.freundii(4)
Ampicillin	NT	6(12)	3(23%)	5(19%)	4(7.2)	1(12.5)	1(33)	1(25)
cotrimoxozole	NT	0	2(15%)	2(7.6)	13(23.6)	0	1(33)	0
Amoxy clav	NT	34(68)	7(54)	14(53)	32(58.1)	3(38)	2(66)	3(75)
Gentamicin	10(34.4)	15(30)	3(23)	4(15)	17(30.9)	4(50)	1(33)	1(25)
Netilmicin	6(20.68)	10(20)	1(7.6)	7(26.9)	15(27.2)	3(37.5)	1(33)	1(25)
Amikacin	16(55.1)	45(90)	10(79)	17(65.3)	36(64.45)	6(75)	2(67)	3(75)
Cefperazone	15(51.7)	30(60)	5(38.5)	15(57.6)	39(73.5)	4(50)	1(33)	2(50)
Ceftazidime	5(17.24)	29(58)	2(15)	15(57.6)	27(49)	6(75)	2(66.6)	2(50)
Cefatoxime	6(20.68)	30(60)	5(38.5)	18(69.2)	42(76.6)	5(62.5)	1(33)	2(50)
Ceftriaxone	6(20.68)	29(58)	4(30.8)	42(76.6)	30(54.5)	6(75)	2(66.6)	2(50)
Ciprofloxacin	3(10.31)	23(46)	1(7.6)	11(42.3)	17(30.9)	7(87.5)	1(33)	1(25)
Imepenem	27(93.1)	49(98)	11(84.6)	25(96)	53(96.36)	8(100)	3(100)	4(100)
Piperacillin	24(82.75)	40(80)	9(69.2)		40(72)	6(75)	2(66.6)	3(75)
Piperacillin/Tazobactum	24(82.75)	45(90)	10(76.9)	23(88.4)	50(90)	7(87.5)	2(66.6)	3(75)

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