DIABETIC FOOT ULCERS MICROBIOLOGICAL STUDY

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ABSTRACT

BACKGROUND AND OBJECTIVES

Infections of all types are more common in patients with diabetes, on the basis of outcome of retrospective study in Canada. Many types of infections are very common in diabetic than non-diabetic patients. Foot is the most common site. Diabetic foot infections range from mild infections to limb threatening conditions. Most require emergency medical attention. Diabetic foot infection is a global burden and projected to increase from 246 million people to over 380 million people by the year 2025. Many people with diabetes develop complications that seriously affect their quality and length of life. Lower limb complications are common, particularly foot ulcers and gangrene. Development of these complications is attributed to individual risk factors, poverty, racial and ethnic differences, and quality of local and national health care systems. The wide variations noted suggest that best practices in low incidence areas could easily be adapted in high incidence areas to reduce the burden of complications. Almost every infection begins in a wound, often as neuropathic ulceration or a traumatic break in the skin. Infections that begin as a small problem may progress to involve soft tissue, bones and joints.

Because of these morbidity and occasional mortality by these foot infections several authoritative groups have recently developed guidelines for assessing and treating diabetic foot.

METHODOLOGY

100 Diabetic patients with foot ulcers were admitted and wounds were classified using wagner's classification. Pus was sent for culture and sensitivity and treated accordingly.

RESULTS

In our study the most common organism cultured from the wound with diabetes mellitus was staphylococcus. The most sensitive drug for these organisms was found to be chloramphenicol on most occasions.

CONCLUSION

The rationale of pus culture and sensitivity is not only to definitively treat the diabetic wound after the culture sensitivity report is available, but also to treat the wounds in places where the culture sensitivity facilities are not available or delayed. So as to target commoner organisms with the drugs which are sensitive on more occasions so as to avoid drug resistance and in a cost effective manner.

KEYWORDS

Diabetic foot, Wagner's classification, Osteomyelitis, Staphylococcus species & chloramphenicol.

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INTRODUCTION: Infections of all types are more common in patients with diabetes on the basis of outcome of retrospective study in Canada. Many types of infections are very common in diabetic than non-diabetic patients. Foot is the most common site. Diabetic foot infections range from mild infections to limb threatening conditions. Most require emergency medical attention. Diabetic foot infection is a global burden and projected to increase from 246 million people to over 380 million people by the year 2025. Many people with diabetes develop complications that seriously affect their quality and length of life.

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Lower limb complications are common, particularly foot ulcers and gangrene. Development of these complications is attributed to individual risk factors, poverty, racial and ethnic differences, and quality of local and national healthcare systems. The wide variations noted suggest that best practices in low incidence areas could easily be adapted in high incidence areas to reduce the burden of complications. Almost, every infection begins in a wound, often as neuropathic ulceration or a traumatic break in the skin. Infections that begin as a small problem may progress to involve soft tissue, bones, and joints. Because of these morbidity and occasional mortality by these foot infections, several authoritative groups have recently developed guidelines for assessing and treating diabetic foot.

Ulcer Severity Classification:
Wagner's System.
Grade 0: Pre-ulcerative lesion.
Grade 1: Partial thickness wound up to, but not through dermis.
Grade 2: Full thickness wound extending to tendons or deeper subcutaneous tissues, but without bony involvement or osteomyelitis.
Grade 3: Full thickness wound involving bone.
Grade 4: Localised gangrene.
Grade 5: Gangrene of entire foot.

University of Texas System Classification: Addition to Wagner system:
Stage A: Clean wounds.
Stage B: Non-ischaemic, infected wounds.
Stage C: Ischaemic, non-infected wound.
Stage D: Ischaemic, infected wounds.

Diabetic foot ulcers represent a major clinical problem. Successful treatment requires a thorough understanding of the pathophysiology, the surgical debridement, and updating various treatment modalities. Failure to recognise the cause, pathology, and associated infectious process may lead to amputation, septicaemia, and death.

Study Design:
- Descriptive Longitudinal Study.
- Analysis Plan: SPSS software.

Study Centre: M.G.M.G.H. Trichy.
Duration of Study: 1 Year (DEC 2014 to DEC 2015).
Sample Size: 100 Cases.

Inclusion Criteria:
- Patients with foot ulcers and diabetes mellitus.
- Age Group: Patients age more than 13 yrs.
- Both Sexes.
- Patients Willing to Participate in the Study.

Exclusion Criteria:
- Wagner's grade 4 and 5.
- Non-diabetic foot ulcers.
- Paediatric age group.
- Death or absconded from ward.
- Patients not willing to participate in the study.

METHODOLOGY:
- Diabetic foot patients were admitted in wards.
- Consent regarding the study was obtained.
- Detailed history recorded.
- Routine blood and urine investigations were done.
- X-ray foot taken.
- Foot ulcers were classified according to Wagner's classification.

OBSERVATION AND RESULTS:
- Total number of patients-100.
- Males-75.
- Females-25.

Diabetes Type:
- Type 2 diabetes mellitus-100.
- Right foot-52.
- Left foot-48.
- Wagner's grade 1-16.
- Wagner's grade 2-62.
- Wagner's grade 3-22.
- Most common organisms- Staphylococcus species (50 cases).
- Most sensitive drug- Chloramphenicol (38 cases).

Mode of Treatment:
- Debridement alone-66.
- SSG-29.
- Amputations-5.

Outcome:
- Good outcome-85.
- Poor outcome-15.

Epidemiology: Clinical studies and some retrospective data have identified several types of infections to occur commonly in diabetics. The frequency of foot cellulitis is 9 times more frequent in diabetics compared to non-diabetics.[1] Osteomyelitis of foot and joints appear to be more common in foot than in any other location. Significant independent risk factors include wounds that penetrated to bone, wounds with more than 30 days duration, recurrent wounds, traumatic aetiology, presence of peripheral vascular disease.[2] The risk factors for severe foot infections are history of previous foot infections, previous vascular disease, neuropathy, but not social and economic factors.[3] Fortunately, most foot infections are superficial, but about a quarter spread to involve deeply involving even the bone.

Around half of diabetic patients who have one foot infections will go on to suffer from another infection within few years. Foot infections and ulcerations are now the leading cause of hospital admissions in our Country. Clinical studies have reported that 25% to 50% of diabetic foot infections lead on to minor amputation and around 10% to 40% of patients go on for major amputations.[4] Of importance here is around 10% to 30% of individuals with diabetic foot ulcer will eventually progress to amputation. About 60% of amputations are preceded by infected foot ulcer. Thus, infection is often a proximate cause leading to tragic outcome.[5]

Pathophysiology: Many numbers of physiologic and metabolic disturbances place the diabetic patients at high risk of foot wounds. Various factors include metabolic derangements, faulty wound healing, vasculopathy, and neuropathy. Microbiological colonisation is unavoidable usually with endogenous organisms, but these become potentially pathogenic in wound environment.

The risk of wound infection rises when local conditions favour bacterial growth than host defences. Avoiding infection in a wound is most effectively achieved by removing the dead tissue and foreign bodies and ensuring good tissue perfusion.[6]

Patient's immune status is also very important predisposing factor for infections. Defects in host immune system in these diabetic foots include mainly impairment in polymorphonuclear leukocyte functions. Many of these defects are the result of metabolic disturbances due to poorly-controlled diabetes. Ketosis in particular impairs leukocyte functions.[7]

Hyperglycaemia also appears to impair the complement functions at least in experimental situations. Impaired wound healing, prolonged persistence of abscess, and poor granulation formation are further accompaniments of diabetes that may predispose to infectious complications. These patients also have staphylococcus as commensals in body elsewhere. This colonisation may predispose to skin infections with this virulent pathogen when there is a break in protective dermal surface.

In addition, different types of skin infections as well as skin and fungal nail infections disproportionately plague diabetic patients. In one study, evidence of pedal fungal infections was found in over 80% of people with long-term type 1 diabetes. Fungal infections provide breaks in the skin, which makes bacterial colonisation easier. The typical anatomy of foot makes foot infections potentially serious. The structure of various compartments, tendon sheaths, neurovascular bundles tend to favour proximal spread of infections. The deep space of foot is divided into medial, lateral, and central compartments. Because of the rigidity of these spaces due to tendons and bones, oedema associated with acute infection may rapidly elevate the compartmental pressure causing ischaemic necrosis of the compartmental tissues. Infections spread from one compartment to another at the proximal calcaneal convergence or by direct septal perforation. But, dorsal or lateral spread is a late sign of infection.

Microbiological Considerations:

Definitions: Skin is coated with bacteria present in a harmless association known as colonisation. Many of these organisms are present permanently, but some are transient. Bacteria are found commonly in disrupted epithelium. When microbial multiplication ensues, wound gets infected. Infection may either follow colonisation or may occur as a primary event. For example in the setting of acute trauma, infection involves invasion of host tissue by microorganisms (Pathogens) with subsequent host inflammatory response.

Some believe that foul odour, tissue friability, and lack of granulation tissue also suggest infection. In a wound, factors such as number and types of organisms, their interaction with each other and with the wound environment, vascular status, and host resistance collectively influence whether or not a wound heals or gets infected. Superficial infection is confined to skin and subcutaneous tissue while deep infection involves involvement of fascia, tendons, joints, and bones. Infection can be due to single organism or more than one (polymicrobial). Bacteria are broadly divided into groups depending on their cell wall reaction to Gram stain (Positive or Negative), requirement for oxygen (Aerobes or Obligate Anaerobes), and their morphology (Cocci or Bacilli).

The predominant organism of normal skin is gram-positive aerobes, particularly low virulent coagulase-negative staphylococci, alpha haemolytic streptococci, and corynebacterium (short rods). When skin is unhealthy, the flora becomes polymicrobial. Virulent gram-positive aerobic notably Staph. aureus and beta haemolytic streptococci may flourish. Antibiotic therapy can also alter the normal flora of skin or wounds favouring organisms that are resistant to agents administered. Lesions infected for short duration appear to be monomicrobial and commonly due to gram-positive pathogens. Chronic wounds develop complex flora with aerobic gram-negative rods, anaerobes, and enterococci in addition to gram-positive aerobes. Fungi also appear to disproportionately colonise the skin of diabetic patients.[8]

Wound Cultures: Culturing, a clinically uninfected wound is unnecessary unless the purpose is to seek presence of epidemiologically significant pathogen (MRSA). When a wound is infected, identifying the microbiological cause will assist in management. The culture will identify the aetiologic agents only if the specimens are cultured in a proper way. Since, it is needed to traverse the skin and subcutaneous tissue of the wound to collect the sample, they may become contaminated with colonisers. As mixed flora usually grow, such reports denote it's a poorly-obtained specimen. Cleaning and debriding the wound before obtaining the tissue specimen will lessen the likelihood of unhelpful microbiology report. Patients with severe, longstanding, or complicated infection or who have already received antibiotics may have polymicrobial infections. Even in this situations, culture and sensitivity results generally help to tailor the antibiotic regimen. In antibiotic naïve, patients with simple infections, normal commensals are usually seen.

Materials obtained from depth usually contain actual pathological organisms.[9] Clinicians frequently culture superficial wounds by rolling a cotton swab across the surface often without prior cleaning or debriding. This sample will contain total colonising flora from which the infecting organism originated lowering the culture specificity. The hostile environment of air-filled swab inhibits growth of anaerobes and fastidious organisms lowering the sensitivity. The scrapings from depth of wound provide good reliable results.[10]

In the beginning of an infection, usually a single organism is identified and when the wound becomes more complex, the flora becomes polymicrobial. Initially, it would have only aerobic organisms; as the wound worsens, it becomes both aerobic as well as anaerobic with mixed flora.[11] Pseudomonas infection occurs in patients who keep their foot for long duration in water. E. coli is common in patients who have already received antibiotics. Some suggest that directing therapy to these organisms in diabetic foot infections might be unnecessary. Obligate anaerobes are frequent in ischaemic wounds with necrosis or that involve deep tissue. Anaerobes are rarely the sole pathogen, but mostly they are mixed with aerobes. Antibiotic resistant organisms especially MRSA is common in patients who had already been treated with other empirical antibiotics.

Diagnosing Infection: Because all skin wounds will be colonised by microorganisms, infection is diagnosed clinically not microbiologically. Infection should be suspected if a local foot problem has pain, swelling, ulcer, sinus, or crepitation. A systemic infection is suspected if fever, vomiting, rigors, tachycardia, confusion, and malaise exist or metabolic disorders like hyperglycaemia, azotaemia, or ketosis exists. It should be considered even if the local signs are less severe. In some cases, these changes can be seen even in non-infectious disorders such as gout, acute Charcot disease.[12] Uninflamed ulcers can have bony involvement, which is evident on x-ray.

Signs of multiorgan involvement are usually not seen in diabetic foot infections even in patients with limb-threatening infections.[13] When infection is suspected, diagnosis should be pursued aggressively, as these infections can worsen in few hours. The most important laboratory tests include visualising and culturing microorganisms from samples of tissues, body fluids, or pus.

Clinical Presentation: The clinical characteristics of patients with diabetic foot infections are similar in most of the reported series. The average age is 60 years; most had diabetes for 15 to 20 years. Two third of patients had peripheral vascular disease (absence of pedal pulse) and about 80% of patients had lost peripheral sensations in foot more commonly in the fore foot region. Most of these patients had some form of treatment before they came to hospital for admission. Most patients have loss of sensation because of sensory neuropathy. But, new onset of pain in previously neuropathic foot is ominous. More number of these patients in this group showed no systemic manifestations.[14]

Judging the Disease: Many grading systems are available for diabetic foot ulcers. Wagner's system is being widely used, it is imprecise. Only Wagner's grade three addresses infection, the key factor in classifying foot infection are assessing depth of wound, presence of ischaemia, and presence of infection.[15] The University of Texas incorporates these features and has been validated in prospective trials.[16]

Bone Infection: Diabetic patients may have coexistent neuropathy that is called neuroarthropathy or Charcot disease. These diseases must be distinguished from bone infections. Bone disease usually moves from the surface of the bone to the deeper surface to involve the marrow. While many patients with foot osteomyelitis have peripheral vascular disease, this is not the major pathophysiologic problem in this disease. About 40-50% of patients have bone marrow involvement.[17] Every patient with chronic ulcer must be suspected of bone involvement.

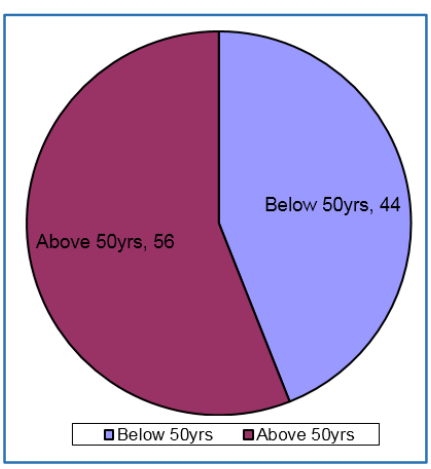
Bigger (more than 2 cm) and deeper (more than 3 mm) ulcers are associated with osteomyelitis. A swollen erythematous digit suggests osteomyelitis. A markedly elevated ESR (more than 70 mm) increases the likelihood of bone infection.[18] Clinical evaluation includes probing the wound with a metal probe.

Various imaging modalities can be used to assess osteomyelitis. Plain x-rays should be ordered for diabetic foot patients except for only cellulites and superficial ulcers. X-rays take up to two weeks for the bony involvement to become evident. Sensitivity of which is only 55%, specificity is 75%. The characteristic changes include bone density loss, cortex involvement, and sequestration of sclerotic bone. When in doubt about bone infection, plain x-ray can be repeated in few weeks interval. If clinical and radiological findings are inconclusive, other imaging modalities can be used. Scanning bones with technetium are 85% sensitive, but because they show high uptake with other bone disorders, they are nonspecific (45%). Leukocyte scans (Indium-111) are highly specific as well as sensitive. Unlike bone scans, leukocyte scans may be useful to define when the disease is halted. But, these are time consuming.

Combining bone scan and leukocyte scan increases, the accuracy and localisation of infection, but also the cost. Radiolabelled anti-granulocyte fragments (ex. sulesomab) and Tc-99 dextran scintigraphies are newer techniques that show promise. Other newer diagnostic techniques are high resolution ultrasound and PET scans. But, the procedure of choice now is MRI with sensitivity of more than 90% and specificity of more than 80%.[19] MRI has the advantage of showing high resolution of not only bone, but also the soft tissue. Its limitations are that early cortical involvement can be missed and marrow oedema can show false positive results.

Several recent studies have shown that soft tissue specimens do not accurately reflect pathogens in bone.[20] Bone specimens can be obtained by open method and percutaneous image-guided method. These are easy to perform. Patients who are on treatment may show normal flora on culture. Bone biopsy is often needed when other investigations are not confirmatory regarding the microbiological results. Microbiological studies of diabetic foot bone disease have uniformly found that staphylococcus species is the most common aetiologic agent in around 40% of infections. S. epidermidis in around 25%, Streptococcus in around 30%, Enterobacteriaceae in around 40%. Some studies have shown that majority of cultures are polymicrobial.[21]

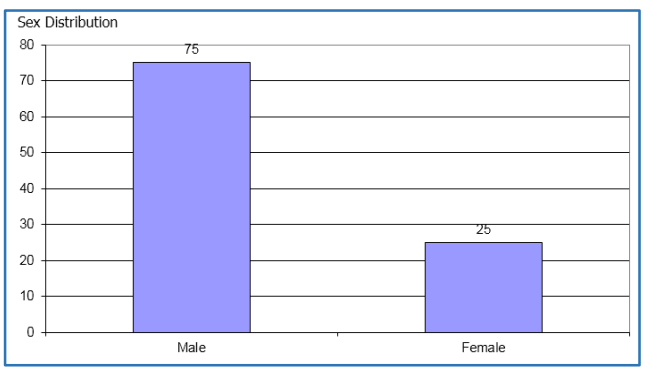
OBSERVATION AND RESULTS:



Age:

Particulars	Frequency (n=100)	Percentage (100%)
Below 50 yrs.	44	44.0
Above 50 yrs.	56	56.0

In my study, out of 100 patients, 44 patients were aged below 50 years and 56 patients were aged above 50 years.



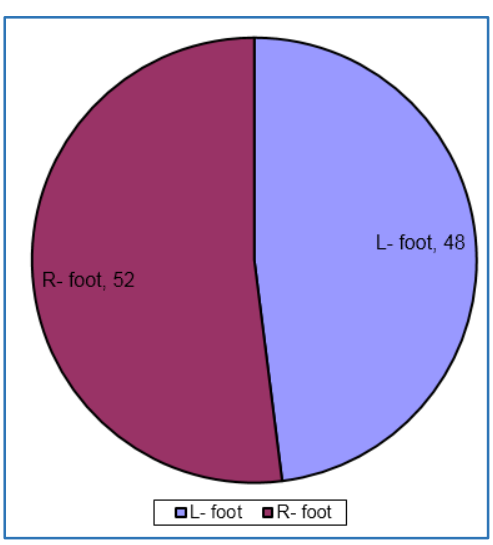
Sex:

Particulars	Frequency (n=100)	Percentage (100%)
Male	75	75.0
Female	25	25.0

In my study, out of 100 patients, 75 were male and 25 were female.

Anatomical Site:
Site:

Particulars	Frequency (n=100)	Percentage (100%)
L-foot	48	48.0
R-foot	52	52.0



In my study, out of 100 patients, 52 patients had lesion in the right leg and 48 patients had lesion in left foot.

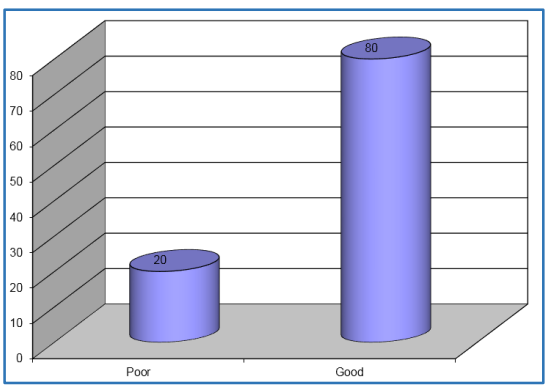
Diabetes type:
DM Type:

Particulars	Frequency (n=100)	Percentage (100%)
Type 2	100	100.0

In my study, all the patients were type 2 diabetics.

Diabetes Control:
Control:

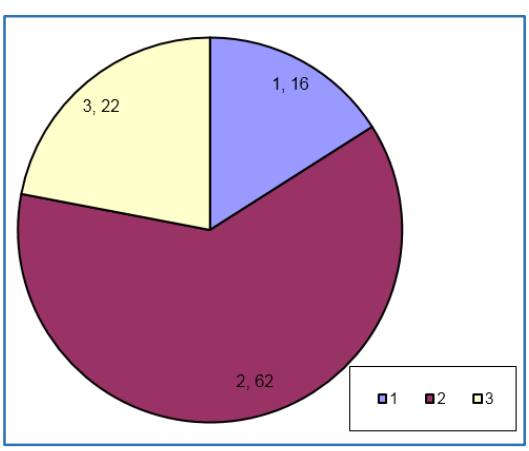
Particulars	Frequency (n=100)	Percentage (100%)
Poor	20	20.0
Good	80	80.0



In my study, out of 100 patients, 80 had good control of diabetes and 20 had poor control of diabetes.

Wagner's Grading:
Wagner Grade:

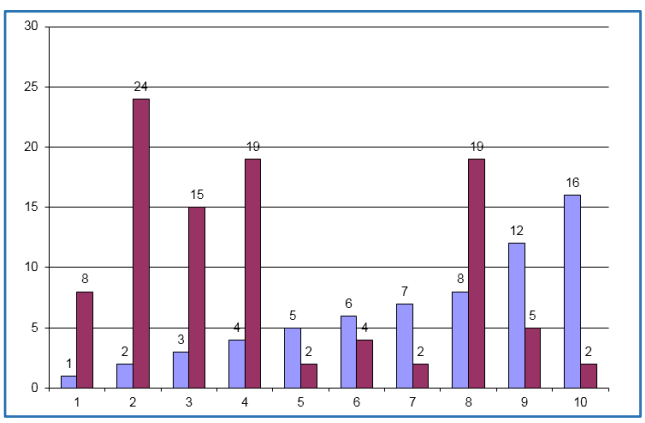
Particulars	Frequency (n=100)	Percentage (100%)
1	16	16.0
2	62	62.0
3	22	22.0



In my study, diabetic foot patients with grade 1-3 were included and patients with grade 1 were 16 in number, with grade 2 were 62 in number, and with grade 3 were 22 in number.

Duration in Weeks:

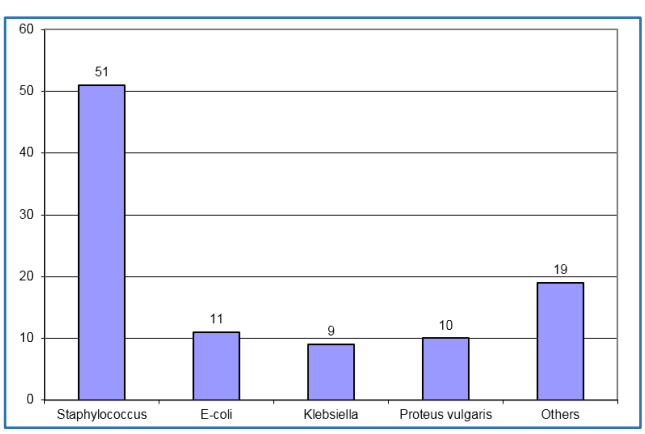
Weeks	Frequency (n=100)	Percentage (100%)
1	8	8.0
2	24	24.0
3	15	15.0
4	19	19.0
5	2	2.0
6	4	4.0
7	2	2.0
8	19	19.0
12	5	5.0
16	2	2.0



In my study, patients with diabetic foot presented with one week to 16 weeks' duration.

Pus Culture Reports:
Pus Culture:

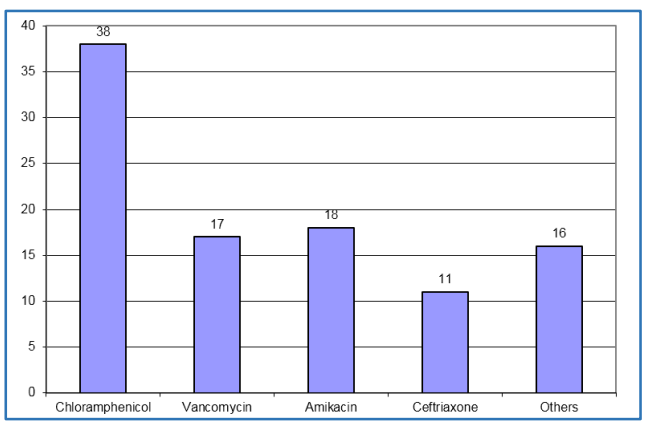
Particulars	Frequency (n=100)	Percentage (100%)
Staphylococcus's	51	51.0
E. coli	11	11.0
Klebsiella Sp.	9	9.0
Proteus Sp.	10	10.0
Others	19	19.0



Out of 100 patients, culture staphylococcus species was found in 51 patients with MRSA in 8 patients, E. coli in 11 patients, Klebsiella species in 9 patients, Proteus species in 10 patients, and rest organisms in minor occasions.

Sensitivity Reporting:
Sensitivity:

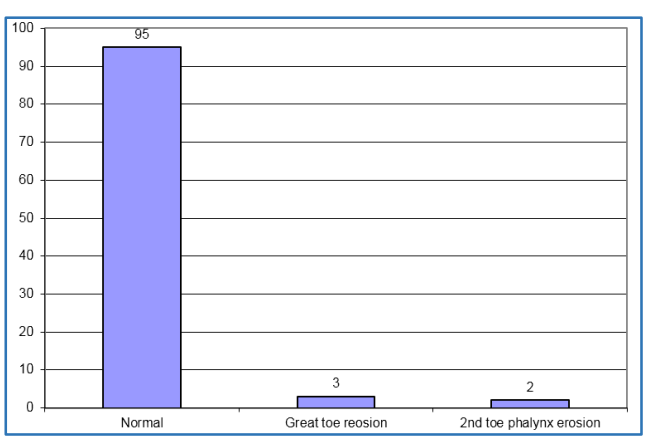
Particulars	Frequency (n=100)	Percentage (100%)
Chloramphenicol	38	38.0
Vancomycin	17	17.0
Amikacin	18	18.0
Ceftriaxone	11	11.0
Others	16	16.0



In my study, in most of the occasions, organisms were sensitive to Chloramphenicol in 38 cases, Vancomycin in 17 cases, Amikacin in 18 cases, Ceftriaxone in 11 cases, and other drugs 16 cases.

X-Ray Foot:

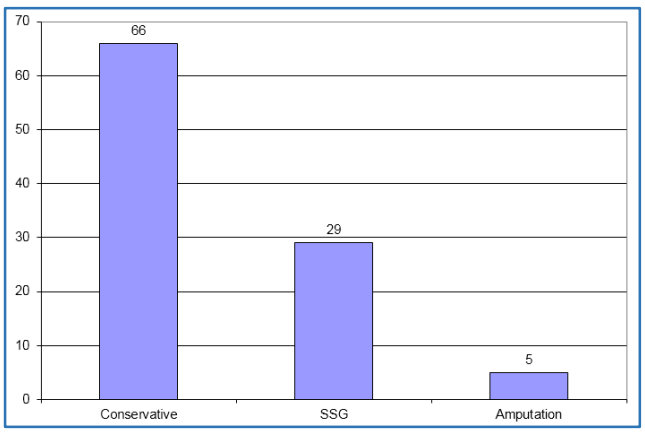
Particulars	Frequency (n=100)	Percentage (100%)
Normal	95	95.0
Great toe erosion	3	3.0
2nd toe phalanx erosion	2	2.0



In my study, x-ray foot was taken for all 100 cases, bone was not involved in 95 cases, 3 cases had great toe erosion, 2 cases had 2nd toe phalanx erosion.

Mode of Treatment:

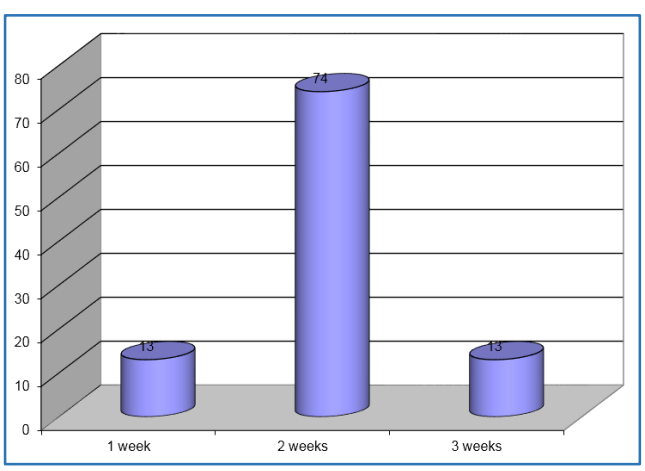
Particulars	Frequency (n=100)	Percentage (100%)
Conservative	66	66.0
SSG	29	29.0
Amputation	5	5.0



In my study, out of 100 patients, 66 patients were treated conservatively with antibiotics and mechanical debridement alone. 29 patients were treated with split skin graft, 5 patients had bone erosion who did not respond to two to three weeks of antibiotics, and they were treated with toe amputations.

Duration of Antibiotics:

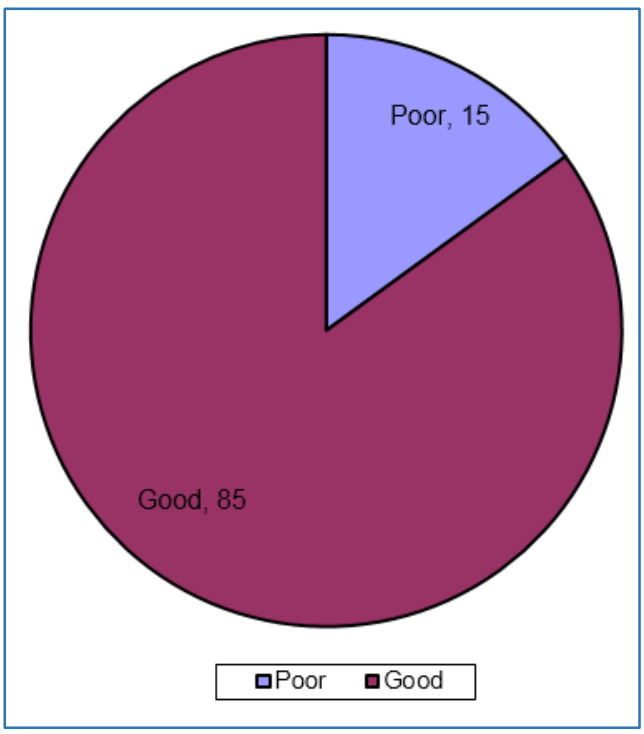
Particulars	Frequency (n=100)	Percentage (100%)
1 week	13	13.0
2 weeks	74	74.0
3 weeks	13	13.0



Out of 100 patients, antibiotic treatment was given for 1 week in 13 cases, 2 weeks in 74 cases, and 3 weeks in 13 cases.

Wound Healing:

Particulars	Frequency (n=100)	Percentage (100%)
Poor	15	15.0
Good	85	85.0



Out of 100 patients treated, 85 patients had good wound healing, 15 patients had poor wound healing.

DISCUSSION: In this study, staphylococcus was the most predominant pathogen either as a sole pathogen or as a combined pathogen in the polymicrobial flora. Staphylococcus was identified in 51% of cases, E. coli was identified in 11% of cases, Klebsiella species in 9% of cases, Proteus species in 10% of cases, rest others in 19% of cases. Regarding sensitivity of drugs, Chloramphenicol was sensitive in 38% of cases, Amikacin in 18% of cases, Ceftriaxone in 11% of cases, Gentamicin in 14% of cases, Meropenem in 6% of cases, Vancomycin in 17% of cases, Piperacillin/tazobactam in 1% of case. Total of 100 patients with Wagner's grade 1-3, 95% of patients had normal x-ray foot. 5 patients progressed to involve the bone for which toe amputation had to be done.

Out of 100 patients, 66% of patients were treated conservatively, 29% of patients were treated with SSG, rest 5% of patients were treated with toe amputation. Antibiotics were given for 1 week in 13% of patients, 74% of patients received it for 2 weeks, and rest 13% of cases received it for 3 weeks. In this study, wound recovered well in 85% of patients and in 15% of patients, the wound healing was not satisfactory.

CONCLUSION: The results from my study shows that diabetic foot ulcers are more common in middle-aged male patients with type 2 diabetes mellitus. The predominant pathogen being staphylococcus species like elsewhere in most parts of India and world as cited in the references in my study. The most sensitive drug being chloramphenicol on most occasions. The rationale of pus culture and sensitivity is not only to definitively treat the diabetic wound after the culture sensitivity report is available, but also to treat the wounds in places where the culture sensitivity facilities are not available or delayed, so as to target commoner organisms with the drugs, which are sensitive on more occasions and to avoid drug resistance in a cost-effective manner.

REFERENCES

1. Boyko EJ, Lipsky BA. Infection and diabetes mellitus. In: Harris MI, ed. Diabetes in America. 2nd edn. NIH publication no 95-1468. Bethesda, MD: National Institute of Health 1995:485-499.
2. Lavery LA, Armstrong DG, Wunderlich RP, et al. Risk factors for foot infections in persons with diabetes. Diabetes Care 2006;29(6)1288-1293.
3. Peters EJ, Lavery LA, Armstrong DG. Diabetic lower extremity infection: influence of physical, social, and psychological factors. J Diabetes Complications 2005;19(2):107-112.
4. Apelqvist J, Bakker K, Van-Houtum WH, et al. International working group on diabetes foot: international consensus on the diabetic foot. Amsterdam 1999:1-96.
5. Pecoraro RE, Ahroni JH, Boyko EJ, et al. Chronology and determinants of tissue repair in diabetic lower extremity ulcers. Diabetes 1991;40(10):1305-1313.
6. Bowler PG. Wound pathophysiology, infection, and therapeutic options. Ann Med 2002;34(6):419-427.
7. Sentochnik DE, Eliopoulos GM. Infection and diabetes. In: Khan CR, Weir GC, eds. Joslin's diabetes mellitus. 13th edn. Philadelphia: Lea and Febiger 1994:867-888.
8. Tan JS, Joseph WS. Common fungal infections of the feet in patients with diabetes mellitus. Drugs Ageing 2004;21(2):101-112.
9. Pellizzer G, Strazzabosco M, Presi S, et al. Deep tissue biopsy vs. superficial swab culture monitoring in the microbiological assessment of limb-threatening diabetic foot infection. Diabet Med 2001;18(10):822-827.
10. Sapico FL, Witte JL, Canawati HN, et al. The infected foot of diabetic foot patient: quantitative microbiology and analysis of clinical features. Rev infect Dis 1984;6(Suppl 1):171-176.
11. Lipsky BA, Pecoraro RE, Wheat JL. The diabetic foot, soft tissue, and bone infection. Infect Dis Clin North Am 1990;4(3):409-432.
12. Pakarinen TK, Laine HJ, Honkonen SE, et al. Charcot arthropathy of the diabetic foot. Current concepts and review of 36 cases. Scand J Surg 2002;91(2):195-201.
13. Armstrong DG, Lavery LA, Sariaya M, et al. Leucocytosis is a poor indicator of acute osteomyelitis of the foot in diabetes mellitus. J Foot Ankle Surg 1996;35(4):280-283.
14. Eneroth M, Apelqvist J, Stenstrom A. Clinical characteristics and outcome in 223 diabetic patients with deep foot infections. Foot Ankle Int 1997;18(11):716-722.
15. Armstrong DG, Lavery LA, Harkless LB. Validation of a diabetic wound classification system. The contribution of depth, infection, and ischaemia to risk of amputation. Diabet Care 1998;21(5):855-859.
16. Oyibo SO, Jude EB, Tarawneh I, et al. A comparison of two diabetic foot ulcer classification systems: the Wagner and the University of Texas Wound Classification Systems. Diabetes care 2001;24(1):84-88.
17. Snyder RJ, Cohen MM, Sun C, et al. Osteomyelitis in diabetic patients: diagnosis and treatment. Part 1: overview, diagnosis, and microbiology. Ostomy Wound Manage 2001;47(1):18-22.
18. Kaleta JL, Fleischli JW, Reilly CH, et al. The diagnosis of osteomyelitis in diabetes using erythrocyte sedimentation rate: a pilot study. J Am Podiatry Med Assoc 2001;91(9):445-450.
19. Craig JG, Amin MB, Wu K, et al. Osteomyelitis of diabetic foot: MR imaging pathologic correlation. Radiology 1997;203(3):849-855.
20. Zuluaga AF, Galvis W, Saldarriaga JG, et al. Aetiologic diagnosis of chronic osteomyelitis: a prospective study. Arch Internal Med 2006;166(1):95-100.
21. Lipsky BA. Osteomyelitis of the foot in diabetic patients. Clin infect Dis 1997;25(6):1318-1326.