Clinical Presentation and Bacteriology of Diabetic Foot Ulcers at Mulago Hospital: A Prospective Case Series Study.

Justin Pario Aldo[a], Agatha Nambuya P.[a], Frederick Nelson Nakwagala[a], Moses Kamya[a], David Livingstone Ejalu[b,*]

[a] *School of Medicine, College of Health of Sciences, Makerere University, Kampala Uganda.*
[b] *Faculty of Health Sciences, Martyrs University Kampala Uganda*

Abstract

Background:

Diabetic foot ulcer is a common cause of mortality among patients admitted to Mulago hospital. Peripheral neuropathy and peripheral arterial disease are the major risk factors for diabetic foot ulceration and the severity of these two conditions may determine treatment options and ulcer outcomes.

General objective:

The study assessed the clinical presentation of foot ulcers in diabetic patients and identified the common bacterial isolates from diabetic foot ulcers and their susceptibility patterns.

Methods:

This was a prospective study involving 60 consecutive patients with diabetic foot ulcers admitted to the endocrinology unit at Mulago hospital between July and December 2010. Patients were recruited after the provision of written informed consent. Deep tissue swabs from the ulcers were then taken at debridement for both aerobic and anaerobic cultures and susceptibility tests using standard microbiological methods.

Results:

Of the 60 patients, 57% had neuroischemic foot ulcers, 18% had neuropathic ulcers, 18% had ischemic ulcers and 7% had unclassified ulcers. 98.3% of the samples grew 93 aerobic bacteria and no anaerobe was isolated. Polymicrobial infection occurred in 41% of samples of which 10% had mixed growth. 80.6% of the isolates were Gram-negative aerobes, mainly E.coli Klebsiella, Pseudomonas, and Proteus while eighteen (19.4%) were Gram-positive, mainly Staphylococcus aureus and Enterococcus faecalis. The prevalence of MuMultidrug-resistant organisms (MDRO) was 84% and Methicillin-resistant staphylococcus aureus (MRSA) was 60% while Extended-spectrum beta-lactamases (ESBL) was 43.5%.

Conclusion:

Diabetic foot ulcers were mostly neuroischemic with moderate neuropathy severity. Infection was mostly due to aerobic Gram-negative organisms with a high prevalence of MDRO, ESBL, and MRSA. Most isolates were susceptible to vancomycin, imipenem, and ciprofloxacin.

Recommendation:

Deep tissue culture and sensitivity tests should be done to make appropriate antibiotic choices for diabetic patients with foot ulcers.

Keywords: Bacteriology, diabetic foot ulcer, Extended-spectrum beta-lactamases, Multidrug-resistance, Submitted: 15th/10/2022 Accepted: 07th/12/2022

1. Background

Globally, Diabetes has reached epidemic proportions and the World Health Organization (WHO) projections indicate that there will be over 366 million worldwide and about 328,000 people in Uganda living with diabetes in 2030 (Wild et al., 2004). A diabetic foot ulcer is a common cause of morbidity, prolonged hospital admissions, lower extremity amputations, and death (Vamos et al., 2010) (Ploeg et al., 2005). Patients often present late with chronic, extensive, and infected ulcers that take a long to heal. The chronicity and poor healing have been attributed to peripheral arterial disease, severe peripheral neuropathy, and multidrug-resistant organisms (MDRO) including Methicillin-resistant staphylococcus aureus (MRSA) and extended-spectrum beta-lactamases (ESBL) (Gadepalli et al., 2006), (Nyamu et al., 2004), (Abdulrazak et al., 2005).

Studies done in Uganda among diabetic patients in general diabetic clinics showed that the prevalence of foot ulcers, a precursor for infection ranges from 4-6.3 % and 60% of the patients had high-risk foot for ulceration (Nambuya et al., 1996) (Bateganya et al., 2003). In Mulago Hospital, the total outpatient attendances due to diabetes increased from 5,179 to 6,600 (27%) during the period July 2007 to June 2008 and July 2008 to June 2009. The corresponding inpatient increase was 34% (572 to 770) in the same period (Hospital records 2009). In the endocrinology unit and surgical wards at Mulago hospital, a total of 160 cases were admitted with diabetic foot ulcers from July 2008 to June 2009 and foot ulcers accounted for 90 out of 755 (12%) of all diabetes-related admissions in the endocrinology unit (Hospital records). As the prevalence of diabetes increases, the long-term complications such as peripheral neuropathy and peripheral arterial disease will also rise in parallel (Norgren et al., 2007).

The clinical presentation of peripheral neuropathy and peripheral arterial disease in diabetic patients with foot ulcers and the bacteriological profile and their susceptibility pattern to common antibiotics (Bansal et al., 2008) in Mulago hospital are not known. Clinicians do not routinely screen and treat peripheral neuropathy and peripheral arterial disease in diabetic patients with foot ulcers. This study, therefore, set out to describe the clinical presentation of foot ulcers in diabetic patients and identify the common bacterial isolates from diabetic foot ulcers and their susceptibility patterns. We intended to describe the clinical presentation of peripheral neuropathy, peripheral arterial disease and to identify the common bacterial isolates and their antibiotic susceptibility pattern in diabetic patients with foot ulcers admitted to Mulago hospital.

2. METHODOLOGY

2.1.

2.1.1. Study design and setting

This was a prospective case series to describe the clinical presentation and bacteriological profile of diabetic foot ulcers from July to December 2010. The study was carried out in Mulago hospital which is Uganda's National referral and teaching Hospital located in the capital city Kampala. The hospital admits about 120 patients with diabetic foot ulcers in one year with an average of 10 patients per month. Most patients with diabetic foot ulcers are admitted to the endocrinology unit of ward 4B.

2.1.2. Study participants

The study targeted males and females above 18 years old with diabetic foot ulcers in Uganda. 60 participants with diabetic foot ulcers admitted to Mulago Hospital were sampled using convenience sampling method from July to December 2010 and included in the study. Patients admitted to the endocrinology unit were recruited to the study. Only those who consented were enrolled consecutively for six months to achieve the sample size of 60 participants. The sample size was

*Corresponding author.
Email address: davidejalu@gmail.com (David Livingstone Ejalu)

purposively sampled because of the limited number of patients admitted at the unit. All patients admitted during the study period were prospectively recruited in to the study based on the set criteria.

2.1.3. Data collection procedure

Face-to-face interviews were held with participants using a semi-structured data collection tool that was checked at the end of the interview for completeness. For each of the study subjects, a history was obtained detailing the patient's sociodemographic characteristics such as age, gender, marital status, usual residence, and level of formal education. History regarding the diabetes including duration of disease (estimated from the year of diagnosis), the mode of the treatment from either the patient or the available hospital records, reported compliance to treatment was documented. The cause of the ulcer, duration of ulcer, previous use of antibiotics before admission, and current antibiotics administered on the ward were noted. Symptoms of peripheral neuropathy and peripheral arterial disease such as numbness, pins, and needles, and intermittent claudication or rest pain respectively were recorded. The ulcer was then examined for its location and signs of infection such as purulence, induration, redness, tenderness, and warmth. A physical examination of both feet was performed for peripheral arterial disease and peripheral neuropathy using standard clinical methods as described below.

2.2. ***Clinical assessment for peripheral neuropathy***

Pressure sensation: 10g (5.07) monofilament at 4 of the 10 standard sites of the sole of the feet (planter base of the big, 2^{nd}, 5th toes and heal), avoiding callosity areas. Vibration sense was elucidated using the 128 Hz turning fork at the hallux of the big. Achilles deep tendon reflex was tested using standard patellar hammer and technique and graded as either present (normal), with reinforcement, or absent. Pain perception was assessed by application of a pin prick on the proximal part of the great toe to barely depress skin and results were classified as present when the patient can distinguish sharpness or absent when the patient cannot distinguish it. Thereafter the Neurological Disability Scoring (NDS) system according to Flynn et al 1992 was used to award scores to each foot and the sum score obtained. In a situation of previous foot amputation, the score awarded to the examined foot was doubled. The NDS system is as outlined below.

2.3. Clinical assessment of peripheral arterial disease:

Both feet were inspected for features of ischemia such as skin darkening (hyperpigmentation), shining (sausage toes), nail thickening, and loss of hair. The dorsalis pedis and posterior tibial arterial pulses were palpated with the patient in the supine position by the investigator in the standard way and these were graded as normal pulse, impaired or absent pulse.

2.4. ***Clinical classification of foot ulcers***

Based on the neuropathy severity score and impairment or absence of foot pulses, the feet were classified as neuropathic, ischemic, or neuroischemic. Foot ulcers were classified as ischemic when the peripheral arterial disease was present but the neurological disability score was ≤ 2; neuropathic when the NDS was ≥ 3 but no obvious peripheral arterial disease and neuroischemic when the NDS ≥ 3 and evidence of peripheral arterial disease was present.

2.5. ***Obtaining deep tissue samples for bacteriological culture***

Any overlying necrotic debris was removed by scrubbing the wound with saline-moistened sterile gauze to eliminate surface contamination. Deep tissue pus swabs were obtained at the base of the ulcer or the deep edge. Sinus tracts were opened and deep pus swabs were taken at the base. The pus swabs were put in BD Port-A-Cult tubes (sterile tubes for both aerobic and anaerobic organisms) and promptly sent to the microbiology laboratory within 30 minutes for processing. The standard operating procedure (SOP) in appendix 5 was used for Gram stain, culture, and sensitivity testing.

Table 1: The NDS system is as outlined below.

Parameter	**Right foot**		**Left foot**	
Sensation	Normal	Abnormal	Normal	Abnormal
Pin (pin prick)	0	1	0	1
Vibration (turning fork)	0	1	0	1
Pressure (monofilament)	0	1	0	1
Achilles tendon reflex				
Present	0			0
Reinforced	1			1
Absent	2			2

The range of neuropathy score is 0-10.
Neuropathy severity classification: 0-2 Noneuropathy; 3-5 Mild neuropathy; 6-8 Moderate neuropathy; >9 severeneuropathy

3. Data analysis

Data were cleaned and coded using EPI-INFO version 3.3.2., descriptive statistics (means, frequencies, and proportions) were generated in SPSS to summarize variables and presented in form of tables, bar graphs, and pie charts.

4. RESULTS

4.1.

4.1.1. Patient demographic data

Describes the social demographics of the study patients. Of the 60 patients, 42 (70%) were females. The mean age was 54.7 years with 40 (66.7%) of the patients above 40 years. Thirty-four (56.6%) of the patients attained primary education, 9 (15%) did not go to school and only 4 (6.6%) had tertiary education. Thirty-four (56.6%) came from rural areas and 39 (65%) patients were married.

Table 2: describes the clinical profile of the study patients. Fifty (83.3%) of the patients were known diabetic patients with a duration of more than one year. Thirty three (55%) patients were taking oral hypoglycemic agents only, twenty (33.3%) were on insulin only, patients were only on dietary control and none was on herbs for diabetes control. Less than half of the patients, 29(48.4%), reported taking their medicines all the time as prescribed by health workers.

Table 3 Describes the clinical history and treatment of the ulcers in the study patients. Thirty-nine (65%) of the patients had ulcers that had occurred for the first time. Forty-four (73.3%) ulcers occurred spontaneously, and less than 2% were caused by the burn. Forty-seven (78.3%) of the ulcers were chronic (more than two weeks). Forty-two (70%) patients used unknown antibiotics at home before admission and of these thirty six (60%) were for less than one week. While on the ward, 52 (86.7%) patients were initiated on a cocktail of intravenous antibiotics mainly Ampicillin, Cloxacillin and Metronidazole (73.3%), ceftriaxone, and metronidazole (10%), and Ampicillin/Cloxacillin/Gentamicin (3.3%). However, eight patients (13.3%) were not given antibiotics as decided by the attending clinicians.

Table 4: describes the clinical features of peripheral neuropathy and severity of neuropathy using the NDS. Symptoms of peripheral neuropathy (numbness and pricking pains) were present in 55 (91.7%) of the patients. The sensory modalities tested were pin prick, monofilament, and vibration sense. Pin prick was abnormal in 28 (46.7%), monofilament was abnormal in 47(78.3%) and vibration was abnormal in forty (66.7%). Deep tendon reflex were absent in thirteen (21.7%) patients, reinforced in twenty four (40%), and normal in twenty (38.3%). Classification on NDS showed that 25 (42%) of the patients had moderate neuropathy, 15(25%) had mild neuropathy, and only 5(10%) patients had severe neu-

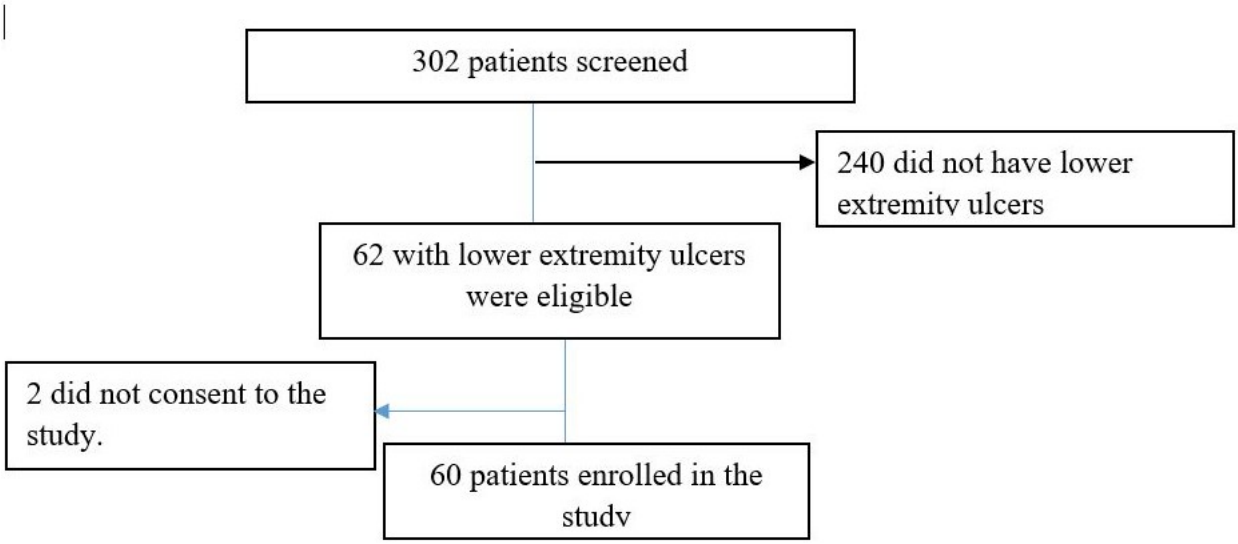

Figure 1: **Flow chart of participant recruitment**

Table 2: **Social demographic characteristics of the study patients**

Social Demographics	**Number**	**%**
Age group(years)		
>40	40	66.7%
<40	20	33.3%
Sex		
Female	42	70%
Male	18	30%
Marital Status		
Married	39	65%
Widowed	11	18.3%
Divorced	9	15%
Single	1	1.7%
Residence		
Urban	26	43.3%
Rural	34	56.7%
Level of education		
Primary	34	56.6%
Secondary	13	21.6%
None	9	15%
Tertiary	4	6.6%

Table 3: **Diabetic history of the study patients**

Clinical profile	**Number**	**Percentage**
Duration of diabetes		
Known Diabetic more than 1 year (Duration in Years)		
1-5	21	35%
6-10	12	20%
>11	17	28.3%
Diagnosed diabetic less than 1 year		
Duration in months		
<1	6	10%
1-11	4	6.7%
Treatment for Diabetes		
Oral hypoglycaemics only	33	55%
Insulin only	20	33.3%
Insulin+ oral hypoglycaemics	5	8.3%
Herbs plus western medicine	2	3.3%
Interruption of treatment		
Yes	31	51.6%
No	29	48.4%

ropathy. Fifteen patients (25%) were classified as having no neuropathy.

Table 5 shows clinical features of peripheral arterial disease in the study patients.Thirty-nine (65%) patients reported symptoms of peripheral arterial disease (intermittent claudication and or rest pain). Fifty-three patients (83.3%) had dystrophic nails, Loss of hair on the foot was present in 39 (65%) patients. Foot skin colour was abnormal (shiny/hyperpigmented) in 35 (58.3%) patients. The posterior tibial artery pulse was absent in 33 (55%) patients and impaired in ten (16.7 %.). Most ulcers occurred in the setting of palpable dorsalis pedis artery, being normal in 32 (53.3%) patients, impaired in 10 (16.7%), and absent in 18 (30%) patients.

4.2. Prevalence of the various types of diabetic foot ulcers in the study patients.

Based on the International Working group of Diabetic Foot classification, Neuroischemic ulcers were the commonest observed in 34(56.7%) patients followed by ischemic and neuropathic ulcers both of which occurred with a similar frequency 11 (18.3%). Four (6.7%) patients presented with unclassified foot ulcers.

Table 7 shows the profile of bacteria isolated from infected foot ulcers of the study patients. Out of the sixty patients studied, 59 samples (98.3%) were culture positive. There were 93 isolates in all samples with some samples having up to 4 organisms. Twenty-five (41.7%) samples had a polymicrobial infection and 10(16.7%) of these were mixed with Gram negative and Gram-positive bacteria. All isolates were facultative aerobes (100%) and no anaerobe was isolated from all samples. Gram negative bacteria were the most common isolates 75(80.6%) with E. coli isolated in 23 (24.7%) patients, Klebsiella in 23 (24.7%), Pseudomonas aureginosa in 11 (12.9%), and Proteus species in 9 (7.7%) patients. Gram positive aerobes accounted for 18(19.4%) of all isolates with Staphylococcus aureus as the predominant bacteria 10 (10.6%) followed by Enterococcus faecalis 6(6.5%).

Table 8 shows the antimicrobial susceptibility of Gram positive aerobes in the study patients. Staphylococcus aureus was the main Gram

Table 4: **Foot ulcer history and antibiotic treatment of the study patients**

Ulcer history and treatment	**Number**	**%**
Previous foot ulcer		
No	39	65%
Yes	21	35%
Cause of current ulcer		
Spontaneous	44	73.3%
Trauma	15	25%
Burns	1	1.7%
Duration of ulcer		
<2 weeks	13	21.7%
2-4weeks	33	55%
>4weeks	14	23.3%
Use of antibiotics before admission		
Yes	42	70%
No	18	30%
Duration of antibiotic use		
<1 week	36	60%
1-2 weeks	9	15%
>2 weeks	15	25%
Antibiotics prescribed on the ward		
Intravenous Ampiclox and metronidazole	44	73.3%
Intravenous Ceftriaxone, metronidazole	6	10%
Intravenous Ampiclox, Gentamicin	2	3.3%
None	8	13.3%

positive aerobe isolated. It was most sensitive to vancomycin 6(100%) followed by ciprofloxacin 9(90%) and was 100% resistant to co-trimoxazole. Methicillin resistance using cefoxtin discs was 60%. Streptococcus pyogens was sensitive to all the tested antibiotics (100%) and Enterococcus faecalis showed 100% resistance to most drugs except Vancomycin where it was 100% sensitive. six (60%) of the staphylococcus aureus isolates, were MRSA while four (40%) were non-MRSA.

Table 9 shows the antimicrobial susceptibility pattern of the main Gram negative isolates from the study patients. All the isolates tested showed 100% resistance to ampicillin and varying high resistance to augumentin as follows; E.coli (82.6%), Klebsiella(85%), proteus(77.8%), and Morganelle morganii(100%). The resistance to the third generation cephalosporin ceftriaxone was 26.3% for E.coli, 64.7% for Klebsiella, 12.5% for Proteus species, and 66.7% for Morganelle morganii. Pseudomonas aureginosa showed relatively better sensitivities to antipseudomonal drugs as follows, gentamicin 72.7 %, ceftazidime 81.8%, cefepime 75%, piperacillin/tazobactum 50%, piperacillin 54.5%, and ciprofloxacin 72.7%. All isolates were 100% sensitive to imepenem. Resistance to chloramphenicol and ciprofloxacin respectively were as follows; E.coli (29.4% and 43.5%), Klebsiella (75% and 60%), Proteus (71.4%and 55.6%) and Morganelle morganii (0% and 75%).

4.3. ***Prevalence of ESBL producing isolates in the study.***

We isolated 46 ESBL producing isolates (23 E.coli and 23 Klebsiella) from the 60 patients. Out Of the 23 E.coli, 7(30.4%) were ESBL. Out of the 23 Klebsiella isolates, 13 (56.5%) were ESBL. The total prevalence of ESBL producing organisms in our study was 43.5%.

Table 5: **Clinical findings of peripheral neuropathy and neuropathy severity scores**

Neuropathy features	**Number**	**%**
History of pricking pains, numbness of feet		
Yes	55	91.7%
No	5	8.3%
Pin prick test		
Abnormal	28	46.7%
Normal	32	53.3%
Monofilament test		
Abnormal	47	78.3%
Normal	13	21.7%
Vibration test		
Abnormal	40	66.7%
Normal	20	33.3%
Deep tendon reflex		
Reinforced	24	40%
Absent	13	21.7%
Normal	23	38.3%
Neurological Disability Score (NDS)		
No neuropathy 0-2	15	25%
Mild 3-5	15	25%
Moderate 6-8	25	42%
Severe >9	5	8%

5. DISCUSSION:

5.1. Clinical presentation of peripheral neuropathy in the study patients.

In this study, 78.3% of the patients had peripheral neuropathy by the standard monofilament testing method. The prevalence of peripheral neuropathy by the composite score of monofilament, pin prick, vibration, and deep tendon reflexes (Neurological Disability Score-NDS) was 75%. Previous studies reported the prevalence of peripheral neuropathy in Mulago hospital to be 46.4% and 35.3% and these are lower compared to our findings. However, the current study reported similar symptoms that occurred more frequently in patients above 30 years(Nambuya et al., 1996) (Obbo, 2002). This difference may be explained by the fact that the two studies were done among all diabetic outpatients while our study was only in diabetic patients with foot ulcers admitted to the ward. However, studies done in Kenya and Tanzania among diabetic patients with foot ulcers showed similar symptoms but with higher peripheral neuropathy prevalence of 78% and 100% respectively compared to our study (Gulam-Abbas et al., 2002), (Nyamu et al., 2004). The slight variation in the prevalence may be due to the subjective nature of the sensory examination. The fact that the majority of the patients had peripheral neuropathy coupled with ulcers that mostly occurred spontaneously, means that our patients probably had an unrecognized injury to the feet due to loss of protective sensation.

We found out that both peripheral neuropathy and peripheral arterial disease coexisted in 56.7% of our study patients while 18.3% of patients had ischemic ulcers, 18.3% had neuropathic ulcers and 6.7% were unclassified ulcers. This prevalence of neuroischemic ulcers is higher compared to other studies elsewhere. In Tanzania, a related study showed that the prevalence of neuroischemic ulcer was 17.5%, 21% for ischemic, and 61.5% for neuropathic ulcers (Gulam-Abbas et al., 2002).

Table 6: **Clinical features of peripheral arterial diseaseamong study patients**

Clinical features	**Number**	**(%)**
History of intermittent claudication or rest pain		
Yes	39	65%
No	21	35%
Loss of hair		
Yes	39	65%
No	21	35%
Nail changes		
Yes	53	83.3%
No	7	16.7%
Foot skin colour		
Shiny/hyperpigmented	35	58.3%
Normal	25	41.7%
Dorsalis pedis artery pulse		
Absent	18	30%
Impaired	10	16.7%
Normal	32	53.3%
Posterior tibial artery pulse		
Absent	33	55.0%
Impaired	10	16.7%
Normal	17	28.3%

Table 7: **Profile of bacterial isolates from infected diabetic foot ulcers in the study**

Profile of bacteria isolated from infected foot ulcers in diabetic patients (n=60)		
Bacteria category	**Number**	**Frequency(%)**
Aerobic and facultative isolates	93	100%
Gram-negative	**75**	**80.6%**
E.coli	23	24.7%
Klebsiella	23	24.7%
Pseudomonas	11	12.9%
Proteus mirabilis	9	7.7%
Morganelle morganii	4	4.3%
Citrobacter freundii	2	2.2%
Acinetobacter species	2	1.1%
Enterobacter species	1	1.1%
Gram-positive	**18**	**19.4%**
S.aureus	10	10.6%
Enterococcus faecalis	6	6.5%
S. pyogenes	2	2.2%

Table 8: Antimicrobial susceptibility pattern of the main aerobic gram positive organisms isolated from infected foot ulcers of the study patients.

Antimicrobial agent(μg)	**Proportion susceptible (%)**		**Enterococcus feacalis (n=6)**
	S.aureus (n=10)	**S.pyogens (n=2)**	
Methicillin sensitive	4(40)	N/A	N/A
Methicillin resistant (MRSA)	6(60)	N/A	N/A
Cefoxtin	4(40)	N/A	N/A
Erythromycin	3(30)	2(100)	0(0)
Co-trimoxazole	0(0)	N/A	N/A
Gentamycin	7(70)	N/A	0(0)
Chloramphenicol	8(80)	2(100)	0(0)
Ciprofloxacin	9(90)	N/A	0(0)
Clindamicin	6(67)	2(100)	N/A
Vancomycin	6(100)	N/A	6(100)
Ceftriaxone	N/A	2(100)	N/A
Penicillin G	N/A	2(100)	N/A
Ampicillin	N/A	N/A	0(0)

MRSA: MethicillinResistant Staph. Aureus

Table 9: **Antimicrobial susceptibility pattern of the main aerobic gram negative bacteria isolated from infected foot ulcers of the study patients.**

Anti-microbial agent(μg)	**Proportion susceptible (%)**				
	E.coli(n=23)	Klebsiella species(n=23)	Pseudomonas(n=12)	Proteus species(n=9)	Morganelle morganii(n=4
Gentamicin	10(43.5)	8(40)	8(66.7)	4(44.4)	0(0)
Ampicillin	0(0)	0(0)	N/A	0(0)	0(0)
Ceftriaxone	14(73.7)	6(35.3)	N/A	8(87.5)	1(25)
Augmentin	4(17.4)	3(15)	N/A	2(22.2)	0(0)
Cefuroxime	2(12.5)	0(0)	N/A	1(11.1)	0(0)
Co-trimoxazole	3(14.3)	3(15.8)	N/A	0(0)	0(0)
Ceftazidime	12(57.1)	4(21.1)	9(75)	6(66.7)	3(75)
Chloramphenicol	12(70.6)	4(25)	N/A	2(22.2)	0(0)
Ciprofloxacin	13(56.5)	8(40)	8(72.7)	4(44.4)	1(25)
Imepenem	17(100)	17(100)	3(100)	7(100)	4(100)
Piperacillin/Tazobactum	N/A	N/A	5(50)	N/A	N/A
Cefepim	N/A	N/A	6(75)	N/A	N/A
Piperacillin	N/A	N/A	6(50)	N/A	N/A
Amikacin	N/A	N/A	2(100)	N/A	N/A

In Kenya, the prevalences were 30.5% for neuroischemic ulcers, 47.5% for neuropathic ulcers, 18% for ischemic ulcers, and only 4% for unclassified ulcers (Nyamu et al., 2004). These differences may be explained by the subjectivity of measurement by different authors and also variations in the study populations. The presence of these comorbidities is responsible for delayed wound healing and calls for multiple interventions to target both conditions.

5.2. ***Bacteriological profile of diabetic foot infection in the study patients.***

Of the 60 cultures performed, fifty nine (98.3%) were culture positive and we isolated ninety three facultative aerobes with an average yield of 1.6 organisms per sample. No anaerobes were isolated. The only organism grew in 58.3% of the samples while 41.7% were polymicrobial. Gram negative bacteria constituted 80.6% while Gram positive bacteria were 19.4%. The fact that we did not isolate any anaerobe is consistent with similar studies done in Tanzania (Gulam-Abbas et al., 2002), Kenya (Nyamu et al., 2004), and Pakistan (Alavi et al., 2007) where only aerobic bacteria were isolated. However, a similar study was done in Nigerian isolated anaerobes in 4.6% and aerobes in 95.4% of samples (Unachukwu et al., 2005). In India, anaerobes were present in 15.3% of samples (Gadepalli et al., 2006) while a multicenter study done in the United States of America isolated purely anaerobic cultures in 1.3% and mixed anaerobic and aerobic in 43.7% of cultures (Citron et al., 2007). In Kuwait, the prevalence of anaerobic organisms was 10.5% in one study (Abdulrazak et al., 2005). However, these studies were done in patients who were antibiotic naive and where samples were obtained by deep tissue curettage compared to our study which was done in antibiotic experienced patients and where deep tissue swabs were taken. These differences may probably be because anaerobes are deep-seated in the wounds or because of previous use of antibiotics before presenting to the hospital. In our study, 70% of the patients mentioned the use of unknown antibiotics before presentation to the hospital. The findings in these two studies may probably mean that anaerobes are difficult to isolate in antibiotic experienced patients.

Polymicrobial infection in our study is consistent with most studies done in Africa, India, America, and Kuwait (Gadepalli et al., 2006), (Gulam-Abbas et al., 2002), (Citron et al., 2007), (Abdulrazak et al., 2005). However, the mean isolate was lower in our study compared to other studies. We found that 58.3% of infections were mono microbial. This means that routine use of combination antibiotics for diabetic foot infections may not be justified in our setting. The Gram negative predominance in our study is consistent with results of studies done in India, Pakistan, and Tanzania that reported prevalences of 51.5%, 54.8%, and 65% respectively in mostly chronic ulcers but this was lower compared to 80.6% in our study (Alavi et al., 2007), (Gadepalli et al., 2006), (Gulam-Abbas et al., 2002). However, in Nigeria mainly aerobic Gram positive organisms of 56.1% with *staphylococcus aureus* accounting for 24.4% of total isolates (Akanji et al., 1989). Our study similarly revealed that *Staphylococcus aureus* was the main Gram positive aerobe but not the most frequently isolated organism. The Gram negative predominance may be due to the longer duration of ulcers before presentation to the hospital.

5.3. ***Antibiotic susceptibility patterns of the bacterial isolates in the study.***

Our study showed the prevalence of 84% for multidrug resistant organisms (MDRO), 60% for methicillin resistant staphylococcous aureus (MRSA), and 43.5% for extended spectrum beta lactamases (ESBL). All Gram negative isolates were 100% resistant to ampicillin but 100% sensitive to imepenem while Gram positive isolates were 100% sensitive to vancomycin. Both Gram positive and Gram negative isolates were more susceptible to ciprofloxacin compared to other common antibiotics that were tested. Similarly, high prevalences of MDRO of 85% and 83.8% have been reported in India and the United States respectively (Gadepalli et al., 2006), (Citron et al., 2007). A lower prevalence of 65% has been reported in Pakistan (Alavi et al., 2007).

For MRSA, our prevalence was similar to the 56% found in India (Gadepalli et al., 2006) and higher compared to 22.2% and 4.4% in Pakistan and the United States respectively (Alavi et al., 2007), (Citron et al., 2007). These findings suggest that Vancomycin may only be the best option for treating MRSA. This poses a great threat in the treatment of diabetic foot infection in our setting because most patients present late and may not be able to afford efficacious therapy.

Gram negative isolates also showed high resistance to common antibiotics. Only *Pseudomonas* had the least multidrug resistance of 25%. The study showed a considerable prevalence of extended spectrum beta lactamase (ESBL) of 43.5%. No such studies have been published in Africa to compare our findings but a similar prevalence of 44.7% has been reported in India (Gadepalli et al., 2006). This finding once again threatens the use of 3^{rd} generation caphalosporins in the management of diabetic foot infection. This means that the most effective drug that targets Gram negative bacteria can only be 4^{th} generation cephalosporins that are very expensive. However, 3^{rd} generation cephalosporins may be used based on the sensitivity pattern of the isolated microbe. For *pseudomonas*, ceftazidime, ciprofloxacin, and gentamicin may be used since there was relatively low resistance to these drugs.

6. Limitations of the study:

Measurements for peripheral neuropathy and peripheral vascular disease were clinical and therefore subjective. Alternative methods such as doppler ultrasound peripheral arterial disease could be used. However, monofilament testing is currently the recommended screening tool for peripheral neuropathy and exclusive measurement by the principal investigator was done to minimize interobserver bias in measurements.

7. Acknowledgments:

Management and Administration of Mulago National Referral hospital provided all the support for the research.

8. Author Contribution:

PAJ collected the data, performed the analysis, and drafted the manuscript. EDL extensively reviewed the manuscript, MK, NFN, and MPA supervised the entire research and gave approval to the final version.

9. Funding:

There was no funding for this study.

10. Availability of data and materials:

Supplementary material has not been provided for the data sources.

11. Consent for publication

Not applicable.

12. Conflict of interests:

The authors declare that they have is no conflict of interests.

13. Ethics statement :

This study was approved by the Research Ethics committee of Makerere University College of Health Sciences, School of Medicine. Participants who participated signed a written informed consent to participate in the study.

14. Publisher details:

Publisher: Student's Journal of Health Research (SJHR)
(ISSN 2709-9997) Online
Category: Non-Governmental & Non-profit Organization
Email: studentsjournal2020@gmail.com
WhatsApp: +256775434261
Location: Wisdom Centre, P.O.BOX. 148, Uganda, East Africa.

References

[1] C. N. Unachukwu, O. K. Obunge, O. J. Odia, The bacteriology of diabetic foot ulcers in Port Harcourt, Nigerian. Nigerian Journal of Medicine 14 (2) (2005) 173–176.

[2] M. H. Bateganya, J. R. Luie, A. P. Nambuya, M. A. Otim, Morbidity and mortality among diabetic patients admitted to Mulago Hospital, Uganda. Malawi Medical Journal: The Journal of Medical Association of Malawi 15 (3) (2003) 91–94.

[3] L. Norgren, W. R. Hiatt, J. A. Dormandy, M. R. Nehler, K. A. Harris, F. G. R. Fowkes, Inter-Society Consensus for the Management of Peripheral Arterial Disease (TASC II), Journal of Vascular Surgery 45 (1) (2007) 5–67.

[4] A. Abdulrazak, Z. I. Bitar, A. Shamali, L. A. Mobasher, Bacteriological study of diabetic foot infections, Journal of Diabetes and Its Complications 19 (3) (2005) 138–141.

[5] P. N. Nyamu, C. F. Otieno, E. O. Amayo, S. O. Mcligeyo, Risk factors and prevalence of diabetic foot ulcers at Kenyatta National Hospital, Nairobi. East African Medical Journal 80 (1) (2004) 36–43.

[6] E. P. Vamos, A. Bottle, A. Majeed, C. Millett, Trends in lower extremity amputations in people with and without diabetes in England, Diabetes Research and Clinical Practice 87 (2) (1996) 275–282.

[7] R. Gadepalli, B. Dhawan, V. Sreenivas, A. Kapil, A. C. Ammini, R. Chaudhry, A Clinico-microbiological Study of Diabetic Foot Ulcers in an Indian Tertiary Care Hospital, Diabetes Care 29 (8) (2006) 1727–1732.

[8] A. O. Akanji, O. O. Famuyiwa, A. Adetuyibi, Factors influencing the outcome of treatment of foot lesions in Nigerian patients with diabetes mellitus, The Quarterly Journal of Medicine 73 (271) (1989) 1005–1014.

[9] E. Bansal, A. Garg, S. Bhatia, A. K. Attri, J. Chander, Spectrum of microbial flora in diabetic foot ulcers, Indian Journal of Pathology and Microbiology 51 (2) (2008).

[10] A. J. Ploeg, J. W. Lardenoye, M.-P. F. M. V. Peeters, P. J. Breslau, Contemporary Series of Morbidity and Mortality after Lower Limb Amputation, European Journal of Vascular and Endovascular Surgery 29 (6) (2005) 633–637.

[11] A. P. Nambuya, M. A. Otim, H. Whitehead, D. Mulvany, R. Kennedy, D. R. Hadden, The presentation of newly-diagnosed diabetic patients in Uganda, QJM 89 (9) (1996) 705–712.

[12] O. Obbo (2002). [link].
URL http://makir.mak.ac.ug/handle/10570/1335

[13] Z. Gulam-Abbas, J. K. Lutale, S. Morbach, L. K. Archibald, Clinical outcome of diabetes patients hospitalized with foot ulcers, Tanzania. Diabetic Medicine 19 (7) (2002) 575–579.

[14] D. M. Citron, E. J. C. Goldstein, C. V. Merriam, B. A. Lipsky, M. A. Abramson, Bacteriology of Moderate-to-Severe Diabetic Foot Infections and In Vitro Activity of Antimicrobial Agents, Journal of Clinical Microbiology 45 (9) (2007) 2819–2828.

[15] S. Alavi, A. D. Khosravi, A. Sarami, A. Dashtebozorg, E. Montazeri, Bacteriologic study of diabetic foot ulcer, Pakistan Journal of Medical Sciences 23 (2007) 681–684.

[16] S. Wild, G. Roglic, A. Green, R. Sicree, H. King, Global Prevalence of Diabetes, Diabetes Care 27 (5) (2004) 1047–1053.

Author biography

Justin Pario Aldo School of Medicine, College of Health of Sciences, Makerere University, Kampala Uganda

Agatha Nambuya P. Lecturer School of Medicine, College of Health of Sciences, Makerere University, Kampala Uganda.

Frederick Nelson Nakwagala Lecturer School of Medicine, College of Health of Sciences, Makerere University, Kampala Uganda. nakwagala@yahoo.com

December 11, 2022

Moses Kamya Professor School of Medicine, College of Health of Sciences, Makerere University, Kampala Uganda.

David Livingstone Ejalu Lecturere Uganda Martyrs University, Faculty of Health Sciences, Kampala Uganda.

December 11, 2022