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Establishing the microbiological and clinical profile in subjects having a diabetic foot in Indian healthcare centre

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***Abstract*---**Introduction: Proper management of diabetic foot infections needs identification and isolation of associated pathogens along with a selection of appropriate antibiotic therapy based on culture and sensitivity. Aims and Objectives: The present study was conducted to assess the prevalence, microbiologic, and clinical profile of subjects having diabetic feet. Materials and Methods: The present retrospective clinical study assessed bacterial culture on tissue samples, pus swabs, and bacterial pus isolates from subjects with diabetic foot admitted to the institute. Results: Enterococcus spp was seen in one subject with Grade 3 disease, streptococcus spp was seen in 1 subject with Grade 3 disease and 2 subjects with Grade 4 disease, and in a total of 3.40% (n=3) subjects, MRSA was seen in 1 subject of Grade 2 disease, 1 in grade 3, and 3 subjects with grade 4 disease, and total of 5.68% (n=5) study subjects and staphylococcus aureus was seen in 2 subjects with grade 1 disease, 2 subjects with grade 3 disease, 4 subjects with grade 4 disease, and 1 subject with grade 5 disease, and in a total of 10.22% (n=9) study subjects. Acinetobacter spp has been seen in 3 (3.40%) subjects with grade 3 disease and NFGNB in 1, 1, 2, and 2 subjects with grade 1, 2, 3, and

5 study subjects making a total of 6.81% (n=6) subjects. Pseudomonas spp had the highest prevalence with 5, 3, 4, 2, and 8 study subjects with grade 1, 2, 3, 4, and 5 study subjects making a total of 25% (n=22) study subjects. Proteus spp were seen in 14,77% (n=13) of study subjects with 4, 2, 3, and 4 subjects with grade 2, 3, 4, and 5 diseases respectively. Enterobacter spp has seen in 4.54% (n=4) study subjects with 2 each in grade 4 and 5 diseases. Klebsiella spp was seen in 11.36% (n=10) of study subjects with 1, 2, 2, 3, and 2 study subjects respectively. E. coli was seen in 13.63% (n=12) study subjects comprised of 2, 3, 3, and 4 subjects with grade 2, 3, 4, and 5-grade disease. Conclusion: The present study concludes that more predominance of gram-negative bacteria is seen with an increase in Wagner's grade. For deciding the empirical antibiotic therapy, regular antibiotic-resistance pattern surveillance should be conducted.

***Keywords*---**Diabetic foot infections (DFI), Clinical and microbiological profile, Polymicrobial, Sensitivity pattern, Wagner's Grading.

Introduction

Chronic foot infections in subjects with diabetic Mellitus are usually seen and are usually difficult to manage with conservative treatment. The literature data reports that diabetic subjects are at 10 times higher risk of hospitalization secondary to soft tissue infection in the foot region compared to non-diabetic subjects. It is estimated that the diabetic population in India will increase to nearly 57 million by the end of 2025.[1] The infection in subcutaneous tissues can easily spread to deeper tissues resulting in the amputation of the limbs secondary to gangrenous changes. In diabetic subjects, uninfected ulcerations are complicated after minor trauma in diabetic subjects with neuropathy leading to tissue necrosis om neuropathic ulcers/ osteomyelitis and sinus which usually draining.[2]

Moist gangrene development is largely governed by the persistence of the infection. The organisms that persist in the gangrenous infection are Proteus spp, Enterococcus spp, and pseudomonas spp which cause extensive destruction in tissues secondary to poor circulation of blood in the affected foot.[3]

Appropriate and accurate management of infections in diabetic feet needs identification and isolation of pathogens associated along with a selection of appropriate antibiotic therapy based on culture and sensitivity.[4] This retrospective study was to assess associated bacterial pathogen with diabetic foot and their effect on outcomes following treatment of the diabetic foot.

The present study was conducted to assess the prevalence, microbiologic, and clinical profile of subjects having a diabetic foot with susceptibility to antibiotics and to assess these microbiologic and clinical profiles based on various grades of Wagner classification and outcomes in diabetic foot infection subjects.

Materials and Methods

The present retrospective clinical study was conducted to assess the prevalence, microbiologic, and clinical profile of subjects having a diabetic foot with susceptibility to antibiotics and to assess these microbiologic and clinical profiles based on various grades of Wagner classification and outcomes in diabetic foot infection subjects. The study was conducted after obtaining clearance from the concerned Ethical committee. The study population was comprised of the subjects visiting the institute with gangrenous infection of foot in diabetic subjects.

The inclusion criteria were subjects with a confirmed diagnosis of diabetes, having foot infections that worsen with time, not responding to conventional antibiotic therapy, subjects who were not terminally ill, subjects in sound mental state, and subjects who were willing to participate in the study. Exclusion criteria were subjects with associated diseases contributing to infections and subjects who were not willing to participate in the study.

After the final inclusion of the subjects tissue samples, pus swabs, and bacterial pus isolates were collected from all the subjects admitted to the institute with diabetic feet. Following collection, based on standard procedures, the samples were processed for antimicrobial susceptibility pattern, bacterial identification, and culture.

The records were maintained concerning first and second-line antimicrobial therapy, sensitivity patterns, and isolated microorganisms where all the assessments were done based on outcomes in the subjects, risk factors, and ulcer gradings. The collected data were subjected to the statistical evaluation using SPSS software version 21 (Chicago, IL, USA) and one-way ANOVA and t-test for results formulation. The data were expressed in percentage and number, and mean and standard deviation. The level of significance was kept at $p<0.05$.

Results

The present retrospective clinical study was conducted to assess the prevalence, microbiologic, and clinical profile of subjects having a diabetic foot with susceptibility to antibiotics and to assess these microbiologic and clinical profiles based on various grades of Wagner classification and outcomes in diabetic foot infection subjects. The study included a total of 132 subjects from both genders within the age range of 58-72 years. Demographic and disease-related characteristics of the study subjects are described in Table 1. It was seen that the mean age of the study subjects was 63.4±3.67 years. There were 79.54% (n=105) males and 20.45% (n=27) females in the present study. On assessing the microbial growth, monomicrobial growth was seen in 78.78% (n=104) subjects, whereas, polymicrobial growth was seen in 21.21% (n=28) study subjects. There were 10.60% (n=14) bacterial isolates that had Wagner's grade I, 12.87% (n=17) had Wagner's grade II, 25.75% (n=34) subjects had Wagner's grade III, and 22.72% (n=30) subjects had Wagner’s grade IV (Table 1).

On assessing the distribution of gram-positive cocci in the various W grades in the study subjects with diabetic foot, it was seen that Enterococcus spp was seen

in one subject with Grade 3 disease, streptococcus spp was seen in 1 subject with Grade 3 disease, and 2 subjects with Grade 4 disease, and in a total of 3.40% (n=3) subjects, MRSA was seen in 1 subject of Grade 2 disease, 1 in grade 3, and 3 subjects with grade 4 disease, and total of 5.68% (n=5) study subjects and staphylococcus aureus was seen in 2 subjects with grade 1 disease, 2 subjects with grade 3 disease, 4 subjects with grade 4 disease, and 1 subject with grade 5 disease, and in a total of 10.22% (n=9) study subjects (Table 2).

For gram-negative bacilli, it was seen that Acinetobacter spp was seen in 3 (3.40%) subjects with grade 3 disease and NFGNB in 1, 1, 2, and 2 subjects with grade 1, 2, 3, and 5 study subjects making a total of 6.81% (n=6) subjects. Pseudomonas spp had the highest prevalence with 5, 3, 4, 2, and 8 study subjects with grade 1, 2, 3, 4, and 5 study subjects making a total of 25% (n=22) study subjects. Proteus spp were seen in 14,77% (n=13) of study subjects with 4, 2, 3, and 4 subjects with grade 2, 3, 4, and 5 diseases respectively. Enterobacter spp has seen in 4.54% (n=4) study subjects with 2 each in grade 4 and 5 diseases. Klebsiella spp was seen in 11.36% (n=10) of study subjects with 1, 2, 2, 3, and 2 study subjects respectively. E. coli was seen in 13.63% (n=12) study subjects comprised of 2, 3, 3, and 4 subjects with grade 2, 3, 4, and 5-grade disease (Table 3).

Discussion

The present retrospective clinical study was conducted to assess the prevalence, microbiologic, and clinical profile of subjects having a diabetic foot with susceptibility to antibiotics and to assess these microbiologic and clinical profiles based on various grades of Wagner classification and outcomes in diabetic foot infection subjects. The study included a total of 132 subjects from both genders within the age range of 58-72 years. It was seen that the mean age of the study subjects was 63.4±3.67 years. There were 79.54% (n=105) males and 20.45% (n=27) females in the present study. On assessing the microbial growth, monomicrobial growth was seen in 78.78% (n=104) subjects, whereas, polymicrobial growth was seen in 21.21% (n=28) study subjects. There were 10.60% (n=14) bacterial isolates that had Wagner's grade I, 12.87% (n=17) had Wagner's grade II, 25.75% (n=34) subjects had Wagner's grade III, and 22.72% (n=30) subjects had Wagner's grade IV. These findings were consistent with the results of Bajuri MY et al[5] in 2017 and Alva KA et al[6] in 2013 where authors assessed subjects with comparable demographics as in the present study.

The present study also assessed the distribution of gram-positive cocci in the various W grades in the study subjects with diabetic foot, it was seen that Enterococcus spp was seen in one subject with Grade 3 disease, streptococcus spp was seen in 1 subject with Grade 3 disease and 2 subjects with Grade 4 disease, and in a total of 3.40% (n=3) subjects, MRSA was seen in 1 subject of Grade 2 disease, 1 in grade 3, and 3 subjects with grade 4 disease, and total of 5.68% (n=5) study subjects and staphylococcus aureus was seen in 2 subjects with grade 1 disease, 2 subjects with grade 3 disease, 4 subjects with grade 4 disease, and 1 subject with grade 5 disease, and in a total of 10.22% (n=9) study subjects. These results were in agreement with the studies of Stacey HJ et al[7] in

2019 and Girish MB et al[8] in 2010 where authors reported the presence of similar gram-positive cocci in the culture of pus from the diabetic foot.

For gram-negative bacilli, it was seen that Acinetobacter spp was seen in 3 (3.40%) subjects with grade 3 disease and NFGNB in 1, 1, 2, and 2 subjects with grade 1, 2, 3, and 5 study subjects making a total of 6.81% (n=6) subjects. Pseudomonas spp had the highest prevalence with 5, 3, 4, 2, and 8 study subjects with grade 1, 2, 3, 4, and 5 study subjects making a total of 25% (n=22) study subjects. Proteus spp were seen in 14,77% (n=13) of study subjects with 4, 2, 3, and 4 subjects with grade 2, 3, 4, and 5 diseases respectively. Enterobacter spp has seen in 4.54% (n=4) study subjects with 2 each in grade 4 and 5 diseases. Klebsiella spp was seen in 11.36% (n=10) of study subjects with 1, 2, 2, 3, and 2 study subjects respectively. E. coli was seen in 13.63% (n=12) study subjects comprised of 2, 3, 3, and 4 subjects with grade 2, 3, 4, and 5-grade disease. These results were comparable to the results by the studies of Goh TC et al[9] in 2020 and Mahamoud BA et al[10] in 2013 where authors reported a similar pattern of distribution for gram-negative bacilli in the culture from the samples of subjects with a diabetic foot infection.

Conclusion

Within its limitations, the present study concludes that the prevalence and predominance of gram-negative microorganisms increase with the increased grade of Wagner's disease. For deciding the empirical antibiotic therapy, regular antibiotic-resistance pattern surveillance should be conducted. The antibiotics given to these subjects should be assessed based on culture and sensitivity. However, the present study had a few limitations including a small sample size, shorter monitoring period, and geographical area biases. Hence, more longitudinal studies with a larger sample size and longer monitoring period will help reach a definitive conclusion.

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TABLES

Table 1: Demographic and disease-related characteristics in the study subjects

Characteristics	**Percentage (%)**	**Number (n=132)**
Mean age (years)	63.4±3.67	
Age Range (years)	58-72	
Gender		
Males	79.54	105
Females	20.45	27
Microbial Growth		
Monomicrobial	78.78	104
Polymicrobial	21.21	28
Wagner's grades		
I	10.60	14
II	12.87	17
III	25.75	34
IV	22.72	30

Table 2: Distribution of gram-positive cocci in the study subjects

Gram-positive cocci	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**	**Total (n=18)**	**Percentage (%)**
Enterococcus spp			1			1	1.13
Streptococcus spp			1	2		3	3.40
MRSA		1	1	3		5	5.68
Staphylococcus aureus	2		2	4	1	9	10.22

Table 3: Distribution of gram-negative bacilli in the study subjects

Gram negative bacilli	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**	**Total (n=70)**	**Percentage (%)**
Acinetobacter spp			3			3	3.40
NFGNB	1	1	2		2	6	6.81
Pseudomonas spp	5	3	4	2	8	22	25
Proteus spp		4	2	3	4	13	14.77
Enterobacter				2	2	4	4.54
Klebsiella spp	1	2	2	3	2	10	11.36
E. coli		2	3	3	4	12	13.63