



## COMPARISON OF MICROBIOLOGICAL RESULT OF DEEP BIOPSY CULTURE AND SUPERFICIAL SWAB IN DIABETIC FOOT INFECTION

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### ABSTRACT

**Introduction-** Diabetic foot a common complication of diabetes patients. This study was to compare the efficacy in identifying the organisms and the best method of specimen collection for culture study by comparing the culture of swab vs deep tissue. **Methods-** Hospital based cross-sectional study was conducted on 50 patients of diabetic foot with more than 18 yrs age. **Results-** Maximum 52% patients belonged to age group was 51-70 years. Male patients (78%) contributed to larger proportion as compared to females (22%). Most common lesion was right dorsum of foot 11 (22%) followed by left toe 7(14%), left whole foot( 10%). 60% patients were suffering from diabetes for  $\geq 10$  years. 54% patients had interphalangeal joint involvement. 54% patients were suffering from infection whereas 30% were having ulceration on diabetic foot & 16% were suffering from bone destruction. 58.0% patients were having peripheral neuropathy, 64% were having  $> 10$  years of diabetes, 46% were having peripheral arterial disease, 40% were having history of prior ulceration, 36% patients were having diabetic Charcot joint & 28% were having uncontrolled hyperglycemia 12% of patients were having ophthalmic problems whereas 4% & 2% were having cardiovascular and renal problems Staphylococcus aureus (28) was found to be the frequently isolated pathogen. Only 21 strains of S.aureus were isolated from swab culture. But tissue samples yielded the growth of 28 strains of S.aureus. Second common pathogen isolated was Coagulase negative Staphylococci (18). All 14 strains of CONS were grown in swab culture but tissue culture yielded 11 strains of CONS. **Conclusion-** In our study, tissue culture was found to be the most acceptable technique in identifying the microbial etiology of limb threatening diabetic foot infections. Appropriate specimen collection helps to overcome the unnecessary usage of antibiotic therapy.

### KEYWORDS : Diabetic, Foot, CONS

#### INTRODUCTION

Diabetes mellitus is commonest endocrine disorder and in last decade rapid increase in cases has been recorded more so in urban population. The prevalence is 2 – 4% in rural population and 5 – 8% in urban population. In western countries females are affected more than males whereas in india prevalence of diabetes is more in males.<sup>1</sup>

Diabetic foot is condition which generally arises due to long standing diabetes and negligence at the part of patient and perhaps the physician too which brings lot of suffering to the patient, bodily and mentally and worry to the surgeon because in a country like India most of patients present a late stage having tried various treatment options ranging from Ayurveda, Homeopathy, Unani, Siddha etc. and have already reached a stage in which gangrene sets up and there is heavy bacterial colonization warranting amputation otherwise patient may die of septicemia.<sup>2</sup>

Vascular assessment is an important tool which helps to determine the need for hospitalization, surgical interventions. DFIs are polymicrobial, with staphylococci (aerobic gram-positive cocci), the MC causative organism. Aerobic gram-negative bacilli are definite copathogens in chronic wounds & followed after antibiotic treatment. Obligate anaerobes may be copathogens in ischemic or necrotic wounds. Wounds without evidence any infection do not require antibiotic therapy. For infected ulcer, a post-debridement specimen is an important tool for aerobic and anaerobic culture. Broader spectrum regimens for chronic and severe infections. Osteomyelitis can be difficult to diagnose and treat. Many DFIs require surgical intervention, ranging from minor (debridement) to major (resection, amputation). Proper dressing with of loading the pressure & adequate follow up is necessary. An ischemic foot may require revascularization,

few non responding patients may benefit from selected adjunctive measures.<sup>3,4</sup>

Non healing chronic ulcers, inspite of daily dressing with local applications, it does not heal. This problem is especially seen in diabetic ulcers, venous ulcers and pressure ulcers. Treating these wound is a constant challenge for the surgeon.

The peculiarity of a chronic wound is that, they refuse to heal. Wound debridement and dressings, improving the nutritional status are all important factor in wound healing. Various studies were done on dressings in the management of DFIs. In spite of all these, treating the microbes of native wound is far most important and nidus in the management. Various debates going on for the best method of specimen collection whether it is swabbing or deep tissue culturing.<sup>5</sup>

There were studies which proved deep tissue culture, the best method of identifying the microbes. But still in many peripheral even in tertiary centres many clinicians following the SWAB TECHNIQUE for microbes culturing. This study was to compare the efficacy in identifying the organisms and the best method of specimen collection for culture study by comparing the culture of SWAB VS DEEP TISSUE.

#### MATERIAL & METHOD:

**1.STUDY DESIGN:** Cross sectional Hospital based study

**2.STUDY PLACE:** Department of Surgery, PBM Hospital, Bikaner

**3.STUDY DURATION:** one year

**4.STUDY POPULATION:** All Diabetic foot patient admitted in ward of department of surgery

**5.SAMPLING TECHNIQUE:** Consecutive sampling

**6.SAMPLE SIZE:** all eligible patients admitted in surgery ward were included in study.

**7.INCLUSION CRITERIA:**

- i. patients willing to participate.

**8.EXCLUSION CRITERIA:**

- i. Not Willing to participate.
- ii. Severly ill patients.
- iii. Using antibiotics before hospitalisation

**9.STUDY TOOL:**

1. A pre tested pre structured questionnaire with both open and close ended questions will be used.
2. Wagner's Classification for Diabetic Foot Ulcer
  - Grade 0: no ulcer in a high-risk foot.
  - Grade 1: superficial ulcer involving the full skin thickness but not underlying tissues.
  - Grade 2: deep ulcer, penetrating down to ligaments and muscles, but no bone involvement or abscess formation.
  - Grade 3: deep ulcer with cellulitis or abscess formation, often with osteomyelitis.
  - Grade 4: localized gangrene.
  - Grade 5: extensive gangrene involving the whole foot.

**10.DATA COLLECTION & ANALYSIS:**

After obtaining permission from Ethical Committee and informed verbal consent of study population selected through analyzing inclusion and exclusion criteria and with help of simple random sampling, the questionnaire will be administered to study subjects by the researcher. All relevant information related to study subjects' socio demographic details, anthropometry, clinical profile, biochemical parameters were taken and Two specimens were collected from each wound after the wound had been cleansed (using sterile saline and gauze) and debrided (removal of necrotic tissue, foreign material, calluses, and undermined wound edges). No antimicrobial agent (e.g., alcohol or iodine) or antiseptic was introduced into the wound before specimen collection. Each wound was swabbed using the Levine technique, involving rotation of a wound swab over a 1cm area of the wound for 5 seconds, using sufficient pressure to extract fluid from the inner part of the wound. A specimen of 4mm in diameter was obtained from the furthest extension of abscess is performed in Deep Tissue debridement . The specimens were placed into sterile transport containers and sent to the microbiology laboratory for culturing within 15 minutes. Only one site is sampled for each patient. Cultures were processed following the same standard procedures for the swab and tissue samples. All data collected were entered into Microsoft Excel and was analysed with help of appropriate software and tests of significance considering level of significance as  $p < 0.05$ .

**OBSERVATIONS:**

In present study, maximum 52% patients belonged to age group was 51-70 years followed by 13(26%) in 31-50 age group , 3 (6%)cases in 0-30 age group and 8(16%) cases in more than 70 year age group. Male patients (78%) contributed to larger proportion of our study population as compared to females (22%)

**Table-1: Distribution of cases according to Occupation (N=50 cases)**

Occupation	No.	Percentage
Businessman	8	16%
Driver	2	4%
Farmer	17	34%
House wife	6	12%

Labour	5	10%
Student	2	4%
Retired govt. employee	4	8%
Govt. employee	6	12%
Total	50	100%

Major occupation in present study was farming (34%) followed by Businessman 16% & equal proportion of government employees and housewives (12%).

**Table- 2: Distribution of cases according to site of lesion involved (N=50 cases)**

Lesion	No.	Percentage
Left Dorsum of foot	3	6%
Left Toe	7	14%
Left fore foot	4	8%
Left plantar foot	2	4%
Left whole foot	5	10%
Right Dorsum foot	11	22%
Right Toe	4	8%
Right Fore foot	4	8%
Right plantar foot	4	8%
Right whole foot	3	6%
Right heel	3	6%
Total	50	100%

In this study, most common lesion was right dorsum of foot 11 (22%) followed by left toe 7(14%), left whole foot( 10%).

**Table-3: Distribution of cases according to risk factors responsible for ulceration:**

RISK FACTOR	No.	(%)
Peripheral neuropathy	29	58.0
Foot Deformity	13	26.0
Diabetic Charcot Joint	18	36.0
Trauma	4	8.0
Improperly fitted shoes	5	10.0
Peripheral Arterial Disease	23	46.0
Callus	8	16.0
H/O Prior Ulceration	20	40.0
Higher Plantar Foot Pressure	9	18.0
Limited joint mobility	16	32.0
Uncontrolled hyperglycemia	14	28.0
Chronic Renal Insufficiency	2	4.0
Diabetes duration (> 10 yrs)	32	64.0
Older Age	4	8.0
Poor Foot Hygiene	11	22.0
Poor Vision	6	12.0

Above table shows that 58.0% patients were having peripheral neuropathy, 64% were having >10 years of diabetes, 46% were having peripheral arterial disease, 40% were having history of prior ulceration, 36% patients were having diabetic Charcot joint & 28% were having uncontrolled hyperglycemia.

**Table 4: Comparison of swab and tissue culture techniques.**

Pathogen	Swab	Tissue
Staphylococcus aureus	21	28
Coagulase negative staphylococci	14	11
Enterococci	1	2
Streptococci	2	2
Klebsiella species	7	9
Pseudomonas aeruginosa	6	6
Non fermenting Gram negative bacilli	2	2
Proteus	2	2
E.coli	2	2

Samples were collected by two different techniques from the diabetic foot ulcer and subjected for microbiological methods to isolate the etiology of diabetic foot infection. Staphylococcus aureus (28) was found to be the frequently isolated pathogen. Only 21 strains of S.aureus were isolated from swab culture.

But tissue samples yielded the growth of 28 strains of *S.aureus*. Second common pathogen isolated was Coagulase negative Staphylococci (18). All 14 strains of CONS were grown in swab culture but tissue culture yielded 11 strains of CONS.

## DISCUSSION

This hospital based cross-sectional study was conducted in surgery dept. PBM hospital Bikaner attached with S.P.M. college Bikaner. 50 patients reporting to the General Surgery dept. and eligible as per inclusion criteria were included in the study.

The present study was carried out to analyze two various methods of sample collection in identifying the etiology of diabetic foot infections. In our study it was observed that tissue samples showed higher yield than wound swab specimens, hence providing more information on wound flora. These findings are in agreement with the study conducted by Nelson et al.<sup>6</sup> He observed that tissue sampling had a better yield than wound swab tests, offering more knowledge about wound flora as a result. Although total tissue sampling identified more species than wound swabs, certain species were left out on both techniques. They thus provide similar knowledge to an extent and both methods can be useful. However, yield was differed from bacteria to bacteria.

In our study, total number of *S.aureus* isolated were 28. Swab samples yielded only 21 strains of *S.aureus* but tissue sample yielded 28 strains of *S.aureus*. In case of CONS, total number was 14. Swab samples yielded 14 strains of CONS and tissue samples yielded only 11 strains. Higher number of CONS isolation in superficial swab samples could be due to contamination of the skin flora. It was noticed that Enterococcal growth was predominally seen in tissue samples than swab specimen. Surprisingly, no disparities were noticed in isolation of Gram negative bacilli which is not in agreement with the study conducted by Ying Huang et al.<sup>7</sup>

As per their study, swab culturing is associated with a high risk of missing pathogens, especially Gram-negative bacteria. According to the study conducted by Mutluoglu et al. the findings of swab culture-taken specimens did not compare well with those collected for tissue culture. Which indicates that swab samples may be less accurate than tissue samples to direct antimicrobial therapy.<sup>8</sup>

Another study conducted by Demetriou et al. assessed the diagnostic efficiency of swabs versus tissue cultures, and the discrepancies in bacterial isolates between neuropathic and neuroischemic patients. Swab cultures are highly sensitive but less specific, and have an outstanding negative predictive value in both neuropathic and neuroischemic foot ulcer patients with diabetes. There are no variations in microbial load between the types.<sup>9</sup>

According to Macías et al. report, swab culture is a sensible option for assessing the microbiology of diabetic foot which is not in line with our research findings.<sup>10</sup>

All Gram positive bacterial isolates from Grade II ulcer yielded similar results in both sampling techniques compared to grade 3 and 4. Therefore, for grade 2 wounds, ulcer swabbing, which is easier to conduct and comparatively non-invasive, could be a suitable sampling procedure compared to deep tissue biopsy, which could cause skin, blood vessels and nerves to sustain damage.<sup>11</sup>

Our findings show improved tissue yield relative to wound swab specimens; the full details will be possible as data are collected by using both sampling techniques. This, in conjunction with the literature currently available, supports the suggestions that tissue samples should be chosen over

swab specimens when choosing one form.

Our study has few limitations. Sample size was limited to analyse the data. Obtained results were analysed with the aid of simple percentage method. In addition, fungal microbiota and anaerobic bacteria culture was not performed.

## CONCLUSION

In our study, tissue culture was found to be the most acceptable technique in identifying the microbial etiology of limb threatening diabetic foot infections. Appropriate specimen collection helps to overcome the unnecessary usage of antibiotic therapy.

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