

Original Research Article

Slit skin smear or Fite-Faraco staining of tissue sample, which is a better indicator of bacillary load?

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ABSTRACT

Background: Leprosy is clinically diagnosed on the basis of presence of following cardinal signs: 1. Hypopigmented or erythematous anesthetic patch on skin, 2. Thickened and/or tender peripheral/cutaneous nerve, 3. Acid fast bacilli in slit skin smear. This study aims at comparing bacillary indices in slit skin smears with that from biopsy samples.

Methods: After obtaining informed written consent, slit skin smears were performed and observed for acid fast bacilli. Punch biopsies of lesions were taken and processed and stained with Fite-Faraco technique. The presence or absence of acid-fast bacilli in both modalities was noted.

Results: Slit skin smears were positive in 24 patients and negative in the rest. And 26 patient's biopsy reports had Fite-Faraco positivity, out of the total 46 patients. The $p=0.67$ which was not significant at significance value 0.05.

Conclusions: Skin smear is an equally reliable indicator of bacillary load as Fite-Faraco bacillary index in tissue.

Keywords: Slit skin smear, Fite-Faraco, Bacillary load

INTRODUCTION

Leprosy presents with a wide array of signs and symptoms and sometimes even remains silent without manifestations in the initial stages. It becomes important to identify, classify and provide treatment accordingly to prevent recurrence, deformities and reactions. There were 202 256 new leprosy cases registered globally in 2019.¹ This data highlights the need for the present study.

Leprosy is clinically diagnosed on the basis of presence of following cardinal signs: (i) Hypopigmented or erythematous anesthetic patch on skin, (ii) thickened /and tender peripheral nerve and (iii) acid fast bacilli in slit skin smear. The disease manifests as spectrum of different clinical forms depending upon the immune status of the host ranging from tuberculoid (TT) and borderline tuberculoid (BT) in patients having a strong cell-mediated immunity and to borderline lepromatous

(BL) and lepromatous (LL) forms in those with a robust humoral immunity with mid-borderline (BB) form in between. The spectrum of clinical manifestation has also been classified on an immune-histological and bacteriological scale by Ridley and Jopling.²

Need for study

Confirmation of leprosy can be done by laboratory investigations. Investigations like histopathological examination, PCR, serological tests, etc cannot be performed easily at all health care centres. Hence it is important to analyses the efficacy of slit skin smear (SSS) which is most routinely performed bed-side investigation.

Aim of the study

The aim of the study was to compare the bacillary indices in slit skin smears with that from biopsy samples.

METHODS

All clinically diagnosed cases of leprosy who attended dermatology OPD from January 2020 to June 2020 were included in the study after taking a written informed consent. Treated cases, relapses and patients not willing for participation in study were excluded.

Slit skin smears were taken from 2 sites (an ear-lobule and active lesion) and in the case of single lesion, two smears were taken from diametrically opposite edges of the lesion as per WHO guidelines 1988 and 1992 respectively, using universal precautionary measures.³

Ziehl-Neelsen staining was done on the air-dried smears. The stained smears were examined under an oil immersion objective (100x). In Ziehl-Neelsen (ZN) stained smears viable bacilli were seen as uniform and red stained bacilli with length 4 times more than breadth (Figure 1 and 2). The report was considered “positive” even if any of the smears showed viable bacilli and regarded “negative” if all the smears were negative. At least a hundred fields across the smear were examined before declaring a smear as negative.⁴

The lesions with active inflammatory changes were chosen. The selected site of biopsy was anesthetised with 20% lignocaine after test dose. Punch biopsies were taken from the skin lesions using 3.5 mm punch and despatched to pathology laboratory in formalin to be processed for Fite-Faraco staining and examination (Figure 2).⁴

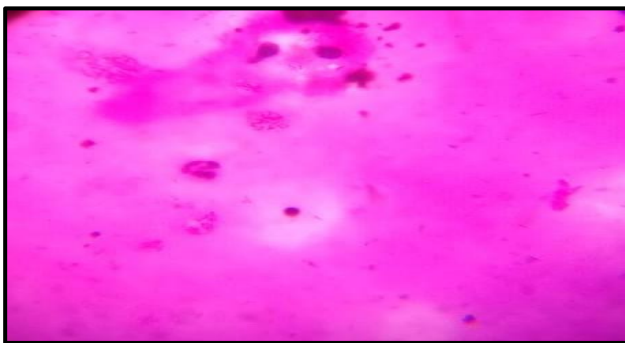


Figure 1: Slit skin smear with acid fast bacilli viewed under oil immersion field in a patient with BL leprosy.

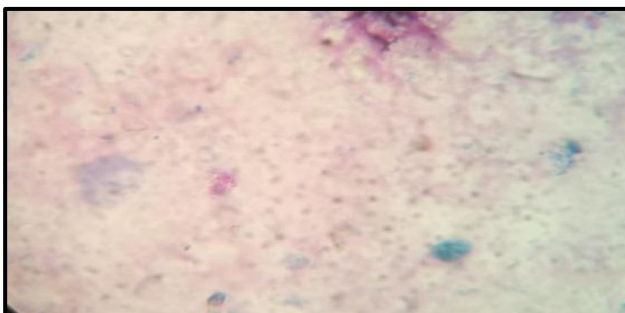


Figure 2: Slit skin smear with acid fast bacilli viewed under oil immersion field in LL leprosy.

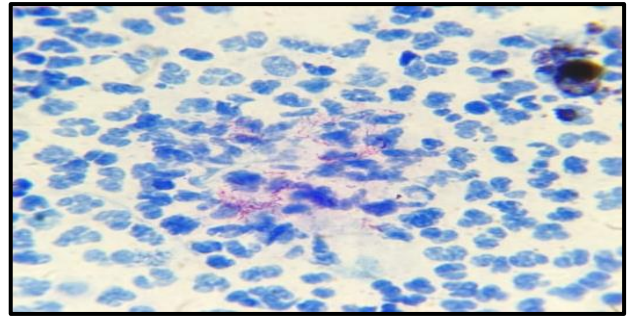


Figure 3: Fite Faraco stained tissue section with acid fast bacilli under oil immersion field in patient with BL leprosy.

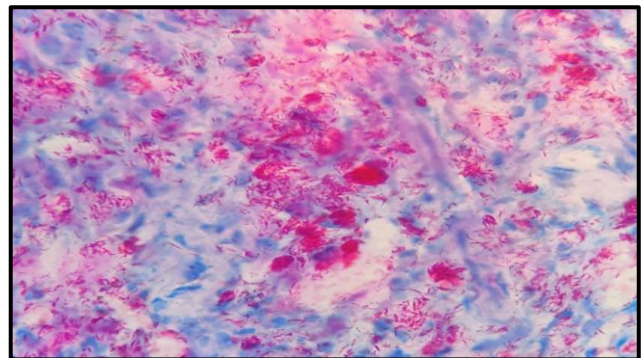


Figure 4: Fite-Faraco stained tissue section with acid fast bacilli under oil immersion field in patient with LL leprosy.

Statistical analysis

Statistical package for social sciences (SPSS) was used for statistical analysis.

RESULTS

Out of 46 clinically diagnosed cases of leprosy, 11 were tuberculoid (TT), 5 cases were borderline tuberculoid (BT), 2 were mid-borderline (BB) and there were 15 and 12 cases of borderline lepromatous (BL) and lepromatous leprosy (LL) respectively. One case was diagnosed as indeterminate leprosy (I) (Table 1). Slit skin smears were positive in 24 patients and negative in the rest. And 26 patient's biopsy reports had Fite-Faraco positivity (Table 2 and 3). The $p=0.67$ which was not significant at significance value 0.05 (Table 3).

Table 1: Number of cases in each type of leprosy.

Spectrum	Number of cases
TT	11
BT	5
BB	2
BL	15
LL	12
I	1
Total	46

Table 2: Cases of SSS and Fite-Faraco positivity.

Spectrum	SSS positive	FF positive
TT	0	0
BT	0	0
BB	00	01
BL	12	13
LL	12	12
I	0	0
Total	24	26

SSS-Slit skin smear, FF-Fite-Faraco

Table 3: Positive and negative result of each modality.

Modality	Positive	Negative	Total
Slit skin smear	24	22	46
Fite-Faraco	26	20	46

Chi-square is 0.1752 and $p=0.6754$. Not significant at $p<0.05$

DISCUSSION

A positive slit-skin smear confirms diagnosis of leprosy. Diagnosis, classification and monitoring response to treatment is aided by slit skin smear examination.⁴

In study by Sendhil et al which was done to compare bacillary index on SSS with that of granuloma, it was found Fite-Faraco is better indicator of bacillary load.⁵

Whereas in study done by Deepa et al auramine rhodamine staining technique was compared with Ziehl-Neelsen (ZN) and modified Fite-Faraco staining method, there was no statistically significant difference between 2 groups, which was similar to findings in present study.⁶

In the study by Reja et al the ability of multiplex polymerase chain reaction and modified Fite-Faraco technique over Ziehl-Neelsen staining, the detection rate was higher in Fite-Faraco compared to SSS which is not comparable to the present study.⁸

Similarly in the study by Patil et al, SSS had the highest detection rate and sensitivity in diagnosing leprosy whereas Fite-Faraco was found to be more specific.⁹

There are other modalities of diagnosis like ML flow test (serological test) which has shown correlation with slit skin smear.¹⁰ Methods like fluorescent staining and polymerase chain reaction (PCR) are superior to SSS but difficult to implement where there are financial and infrastructural constraints.⁷

There is possibility of under-diagnosis and inadequate treatment if diagnosis is not supported by demonstration of bacilli.

CONCLUSION

From this study we conclude that skin smear is an equally reliable indicator of bacillary load as Fite-Faraco bacillary index in tissue. SSS examination is simple and requires minimal expertise compared to histopathological processing. In the absence of histological examination facilities, slit skin smear can be performed. SSS is still one of the most useful diagnostic modalities that can be easily performed till newer feasible methods are unraveled.

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Conflict of interest: None declared

Ethical approval: The study was approved by the institutional ethics committee

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