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Original Article

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A study of histomorphological spectrum of leprosy and its correlation with bacteriological index at a tertiary care center

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Abstract

***Background & Aims*:** Hansen's disease, Leprosy, is a chronic granulomatous infectious disease caused by Mycobacterium leprae, principally affecting the skin and peripheral nerves. Histopathological examination plays an important role in early diagnosis and management. The aim of this study was to study the correlation of histomorphological findings with the bacteriological index in different types of leprosy, and to inspect the histopathological spectrum of leprosy.

***Materials & Methods*:** The retrospective study was carried out on the skin punch biopsies from 121 cases of leprosy taken in the Department of Dermatology at a tertiary care center and reported in the histopathology section of the Department of Pathology between January 2022 to December 2022. Hematoxylin-Eosin and Fite-Faraco-Stained sections were evaluated for features confirming leprosy and further categorized as per Ridley-Jopling system.

***Results*:** Among total 121 skin biopsies examined histopathologically, the most common type seen was Borderline Tuberculoid (BT) Leprosy (32.23%), followed by Lepromatous Leprosy (LL) (20.66%), Borderline Lepromatous (BL) Leprosy (16.52%), Histoid Leprosy (HL) (12.39%), Tuberculoid Leprosy (TL) (9.91%), Erythema Nodosum Leprosum (ENL) (6.61%), and Neuritic (NL) and mid borderline leprosy diagnosed each 1 case (0.82%). Fite-Faraco staining to identify acid-fast bacilli (AFB) was done in all 121 cases, which was positive in 64 (52.8%) of the cases. No bacilli were noted in all cases of TT leprosy, whereas all cases of Histoid types showed the presence of acid-fast bacilli. The correlation of histopathological diagnosis and bacteriological index was seen in 52.8% of the cases. The highest correlation was seen in Histoid Hansen's disease (100%), LL (80%), ENL (75%), BL (70%) followed by BT (23%), TT (0%), Neuritic Leprosy (0%), and BB (0%).

***Conclusion*:** The correlation between clinical manifestations, histopathological, and bacteriological features were required for diagnosis and classification of leprosy. Nerve damage is irreversible, therefore, early detection and treatment is important to prevent disabilities.

***Keywords*:** Bacteriological Index, Fite-Faraco Stain, Hansen's Disease, Leprosy, Ridley Jopling Classification

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Introduction

Leprosy is a chronic granulomatous infectious disease caused by noncultivable Mycobacterium leprae (1). Leprosy has been declared eliminated (prevalence rate < 1 per 10,000 population) as an important public health problem in our country,vas still cases are being reported with varying prevalence throughout many areas in India (2).

Leprosy is one of the leading causes of physical disabilities, which contribute to intense social stigma, resulting in discrimination of patients and their families. Leprosy is known since ancient times as “Kushtaroga”. The causative agent of leprosy, Mycobacterium Leprae, was discovered in 1873 by Armauer Hansen (3).

Depending on the degree of immunity, clinical and histopathological features, various types of leprosy gradually may develop (3). Since exact typing of leprosy is sometimes clinically not possible, added to this, the poor results obtained by slit skin smear leads to false negative diagnosis. Histopathological examination of skin or nerve biopsies as well as demonstration of acid-fast bacilli in histopathological section and in slit skin smear aid in the diagnosis of leprosy (3).

In 1966, Ridley and Jopling proposed the leprosy classification as follows: tuberculoid (TT), borderline tuberculoid (BT), mid-borderline (BB), borderline lepromatous (BL), and lepromatous (LL) (4). Bacteriological Index (BI), the concentration of bacilli in smears, is known as the bacterial or bacteriological index and includes living and dead bacilli.

This study aimed to analyze the correlation of histomorphological findings with the bacteriological index in different types of leprosy as well as to inspect the histopathological spectrum of leprosy.

Materials & Methods

A retrospective hospital-based study was conducted in 121 patients of leprosy. The study was carried out on the skin punch biopsies from untreated cases of leprosy taken in the Department of Dermatology at a tertiary care center and reported in the histopathology section of the Department of Pathology between January 2022 to December 2022. After adequate fixation for about 8-12 hours in 10% formalin, the biopsies were submitted for routine processing, followed by paraffin embedded sections of 3-5µ thickness and stained with hematoxylin and eosin stain. All cases of leprosy were examined for: a) Epidermal atrophy, epithelioid granulomas, number and distribution of lymphocytes, histiocytes and foam cells, b) Infiltration of nerves, blood vessels and adnexa, and c) Grenz zone.

Sections stained with Fite Faraco stain were examined for lepra bacilli in all cases and bacteriological index was reported. Histopathological findings were graded into TT, BT, BB, BL, and LL, according to Ridley and Jopling scale.

The most common index is Ridley's logarithmic measurement, which is based on the number of bacilli for the purpose of oil immersion, and graded as:

6+ = more than 1000 bacilli in an average field,
5+ = 100 to 1000 bacilli in an average field,
4+ = 10 to 100 bacilli in an average field,
3+ = 1 to 10 bacilli in an average field,
2+ = 1 to 10 bacilli in 10 fields, and
1+ = 1 to 10 bacilli in 100 fields.

At least 100 immersion oil smears should be checked before reporting bacterial index slides.

All the clinically diagnosed cases of leprosy were included in the study. Cases were selected regardless of their age, sex, religion, occupation and socioeconomic status. Inadequate biopsies, inconclusive reports, and poorly preserved biopsy were excluded from the study.

Results

The present study included 121 skin biopsies from the patients who were clinically diagnosed as leprosy from January 2022 to December 2022. Age of patients ranged from 11 years to 70 years. Maximum number of cases was seen in the age group of 31-40 years (28.09%), followed by 21-30 years (23.14%), and then 41-50 years’ age group (19%). Less number of cases was seen in 51-60 years (12.39%) and 11-20 years’ age group (8.26%) (Table 1).

Table 1. Distribution of patients According to Age wise

Lesions	11-20 yrs	21-30 yrs	31-40 yrs	41-50 yrs	51-60yrs	61-70 yrs	>70 yrs	Total
Lepromatus leprosy	2	7	7	4	4	1	-	25
Tuberculoid leprosy	1	3	4	-	1	2	1	12
Borderline Tuberculoid leprosy	4	7	13	9	3	3	-	39
Borderline lepromatous leprosy	3	4	4	4	2	3	-	20
Histoid leprosy	-	5	4	3	3	-	-	15
ENL	-	1	1	3	2	-	1	8
Neuritic leprosy	-	-	1	-	-	-	-	1
Mid borderline leprosy	-	1	-	-	-	-	-	1
Total	10(8.26%)	28(23.14%)	34(28.09%)	23(19%)	15(12.39%)	9(7.43%)	2(1.65%)	121(100%)

The study showed marked male predominance; Seventy-one cases (58.67%) males and 50 cases (41.32%) were females. The male-to-female ratio was 1.42 (Table 2).

Table 2. Distribution of Patients According to Sex wise

Lesion	Male	Female	Total cases
Lepromatous leprosy	14(11.57%)	11(9.09%)	25(20.66%)
Tuberculoid leprosy	6(4.95%)	6(4.95%)	12(9.91%)
Borderline tuberculoid leprosy	24(19.83%)	15(12.39%)	39(32.23%)
Borderline lepromatous leprosy	12(9.91%)	8(6.61%)	20(16.52%)
Indeterminate leprosy	-	-	-
Histoid leprosy	8(6.61%)	7(5.78%)	15(12.39%)
ENL	6(4.95%)	2(1.65%)	8(6.61%)
Neuritic leprosy	-	1(0.82%)	1(0.82%)
Mid borderline leprosy	1(0.82%)	-	1(0.82%)
Total	71(58.67%)	50(41.32%)	121

On histopathological examination, among total 121 skin biopsies, the most common type seen was borderline tuberculoid leprosy comprised of 39 cases (32.23%), followed by lepromatous leprosy with 25 cases (20.66%), borderline lepromatous leprosy with 20 cases (16.52%), Histoid leprosy with 15 cases (12.39%), tuberculoid leprosy with 12 cases (9.91%), erythema nodosum leprosum (ENL) with 8 cases

(6.61%), and Neuritic and mid borderline leprosies diagnosed each with 1 case (0.82%) (Table 3).

Table 3. Distribution of Patients According to Histopathological Diagnosis

Histopathological Diagnosis	No.	%
Lepromatous leprosy	25	20.66%
Tuberculoid leprosy	12	9.91%
Boderline Tuberculoid leprosy	39	32.23%
Borderline lepromatous leprosy	20	16.52%
Histioid leprosy	15	12.39%
ENL	8	6.61%
Neuritic leprosy	1	0.82%
Mid border line leprosy	1	0.82%
Total	121	100%

Fite-Faraco staining to identify acid-fast bacilli (AFB) was done in all 121 cases, which was positive in 64 (52.8%) of cases. No bacilli were noted in all cases of TT leprosy, whereas all cases of Histoid types showed the presence of acid-fast bacilli.

When it came to the bacteriological index (Ridley scale) in the patients, it was 4+ and 5+ in 16% and 12% of the patients, respectively. In comparison, it was 1+, 2+, and 3+ in 12%, 7%, and 4% of the patients, respectively, regarding (Ridley Scale). It was +6 in 3% of the patients. Moreover,47% of the patients showed negative findings (Table 4).

Table 4. Distribution of Patients According to Bacteriological Index

Bacteriological Index	**Number of cases**	**%**
0	57	47%
+1	14	12%
+2	8	7%
+3	5	4%
+4	19	16%
+5	14	12%
+6	4	3%
Total	121	100%

As shown in Table 5, the correlation of histopathological diagnosis and bacteriological index was seen in 52.8% of the cases. The highest correlation was seen in Histoid Hansen's disease (100%), LL (80%), ENL (75%), BL (70%) followed by BT (23%), TT (0%), Neuritic Leprosy (0%), and BB (0%) (Table 5).

Table 5. Correlation of Histopathological Diagnosis and Bacteriological Index

HPE	0		1+		2+		3+		4+		5+		6+		Total	CORRELATION
	No	%	No	%	No	%	No	%	No	%	No	%	No	%		
LL	5	4.13	2	1.65	3	2.47	-	-	9	7.5	5	4.13	1	0.82	25	
TT	12	9.93	-	-	-	-	-	-	-	-	-	-	-	-	12	
BT	30	24.9	7	5.78	-	-	1	0.82	1	0.82	-	-	-	-	39	
BL	6	4.95	2	1.65	4	3.30	3	2.47	5	4.13	-	-	-	-	20	52.8%
Histioid Leprosy	-	-	2	1.65	1	0.82	-	-	2	1.65	7	5.78	3	2.47	15	
ENL	2	1.65	1	0.82	-	-	1	0.82	2	1.65	2	1.65	-	-	8	
Neuritic	1	0.82	-	-	-	-	-	-	-	-	-	-	-	-	1	
Midborderline	1	0.82	-	-	-	-	-	-	-	-	-	-	-	-	1	
Total	57	47.2%	14	11.5%	8	6.59%	5	4.11%	19	15.75 %	14	11.56 %	4	3.29%	121	

Histological patterns observed in our study were epidermal changes in the form of thinning and atrophy, followed by normal epidermis and ulcerative changes. Epitheloid cell granuloma and giant cells were more common towards tuberculoid pole, whereas foamy macrophages with clear subepidermal grenz zone were more common towards lepromatous pole.

Discussion

Maximum number of cases was seen in the age group of 31-40 years (28.09%), followed by 21-30 years (23.14%), and then 41-50 years' age group (19%). Fewer numbers of cases were seen in 51-60 years (12.39%), 11-20 years (8.26%), and 61-70 years' age group (7.43%). These findings are comparable with those of Naik et al. (8), Mehta et al. (9), Singh et al. (10), Namrata et al. (11), Baddamet al. (12), and Susmitha et al. (13). These authors found that the most common age group affected was 21-40 years of age group. Although exact reason cannot be given for this age distribution, but variable and long incubation period may be considered as a possible mechanism. Disease occurrence in leprosy is often related to age at detection rather than age at the onset of disease. It is known to occur at all ages ranging from early infancy to very old ages.

In the current study, the majority of patients (58.67%) were male, while the female patients accounted for (41.32%) of the study population. This finding corroborated with the studies conducted by Singh et al. (10), Thamilselvi et al. (14), Kakkadet al. (15), Baddamet al. (12), and Susmitha et al. (13), who found that men were more commonly affected compared to the women. Male predominance may be because of many factors like heavy industrialization, urbanization, and more opportunities for contact in males. Social customs and taboos may account for the

number of females reporting for treatment to the hospital.

In the present study, the most common histopathological diagnosis was related to the BT patients (32.23%). Similar observations were noted in the studies of Naik et al. (52%) (18), Tekwani et al. (57.77%) (16), Nadia et al. (34.7%) (17), Kadam et al. (35.7%) (18), Singh et al. (31.7%)) (10), and Mehta et al. (26%) (9).

The second most common histopathological diagnosis in the current study was related to the patients with LL (20.66%). Similar observations were noted in the studies of Naik et al. (20%) (8), Nadia et al. (21.2%) (17), Singh et al. (13.3%) (10), and Mehta et al. (20%) (9), while only 5.18% of the cases and 9.5% of the cases were identified in the studies conducted by Tekwani et al. (16), and Kadam et al. (18), respectively.

BL was detected in 20 cases (16.52%) in the present study. This result is in line with the results achieved by Naik et al. (13%) (8), Tekwani et al. (14.81%) (16), Singh et al. (10), (21.7%), and Mehta et al. (25%) (9). Only 5.9% and 4.8% of the cases were identified in the studies conducted by Nadia et al. (17), and Kadam et al. (18), respectively.

Histoid leprosy was detected in 15 cases (12.39%) of the cases in the present study. This result is similar to that of the studies conducted by Naik et al. (4%) (8), Tekwani et al. (2.22%) (16), Nadia et al. (3.4%) (17), Kadam et al. (4.8%) (18), and Singh et al. (4.2%) (10). No case of histoid leprosy was found in the study of Mehta et al. (9).

TT was detected in 12 cases (9.91%) in the present study. This is consistent with the studies of Naik et al. (8%) (8), Nadia et al. (14.4%) (17), and Singh et al. (10%) (10), while most TT cases were found in the studies of Tekwani et al. (19.25%) (16), Kadam et al. (19%) (18), and Mehta et al. (26%) (9).

Erythema nodosum leprosum (ENL) was detected in 8 cases (6.61%) in the present study. This is consistent with the studies Semwal et al. (4.1%) (19) and Krutika et al. (0.88%) (20).

Furthermore, BB and Neuritic Leprosies were detected each 1 case (0.82%) in the present study. This result is consistent with the results of Tekwani et al. (0.74%) (16), Kadam et al. (2.4%) (18), Naik et al. (3%) (8), and Mehta et al. (3%) (9). The majority of BB cases were found in the studies conducted by Nadia et al. (16.1%) (17), and Singh et al. (13.3%) (10).

An immunological instability was seen in the borderline cases, which with its treatment, moves towards the tuberculoid pole and without treatment towards the lepromatous pole (13, 14). If the disease is recognized at an early stage, the biopsies taken will have features of the BT stage and if recognized late they have the features of BL stage. Increased awareness of the people due to many national programs makes them to present them at an earlier stage to clinicians, which may contribute to an increased number of borderline leprosy (16).

In the present study, the bacteriological index was 0 in 47% of the cases. This result lends support to the results of Naik et al. (37%) (8), Rahul et al. (80%) (20), Tiwari et al. (22.6%) (21), Giridhar et al. (43.9%) (22), and Kakkad et al. (30%) (15).

It was observed in a recent study that the bacteriological index (Ridley scale) was 4+ and 5+ in 16% and 12% of the patients, respectively. In comparison, it was 1+, 2+, and 3+ in 12%, 7%, and 4% of the patients, respectively, regarding (Ridley Scale). It was 6+ in 3% of the patients. The bacteriological index in the paucibacillary patients was also seen in the studies of Susmitha et al. (21.6%) (13), Tiwari et al. (33.8%) (21), and Kakkad et al. (50%) (15).

The bacteriological index was 3+ in 5 (4%) and 4+ in 19 patients (16%), respectively. It was 5+ and 6+ in 14 (12%) and 4 patients (3%), respectively. The multibacillary cases in the present study were 42 (35%). The bacteriological index in the multibacillary patients was comparable with that index in the studies of Tiwari et al. (66.2%) (21), Giridhar et al. (24.5%) (22), and Kakkad et al. (50%) (15). High bacteriological index (5+ and 6+) was seen in HL and LL types. Our findings were similar to those of Chintal et al. and Anusha et al. (23, 24).

Classification of leprosy patients into multibacillary and paucibacillary determines the duration of their treatment. Misclassification leads to increased risk of relapse due to insufficient treatment if a multibacillary patient is classified as paucibacillary. This also prolongs the time the patient is infective. The cell mediated immune response and bacterial load were determined by the bacteriological index. Generally, paucibacillary leprosy is equivalent to BT and TT disease, and Multibacillary leprosy is equivalent to BB, BL, and LL disease in the Ridley Jopling classification. However, the diagnosis cannot be made only on the basis of bacteriological index, as it can vary in various types of leprosy. In the present study, we used skin biopsies for accurate histopathological classification in all the patients.

In the current study, there was a 52.8% correlation between the histopathological diagnosis and the bacteriological index. The highest correlation was seen in Histoid Hansen's disease (100%), LL (80%), ENL (75%), BL (70%) followed by BT (23%), TT (0%), Neuritic Leprosy (0%) and BB (0%). These finding were in line with the results of Premalatha et al. (25), Tekwani et al. (16), Tiwari et al. (21), Pashupathy et al. (26), and Giridhar et al. (22).

Conclusion

Leprosy though considered to be eliminated from India, is still prevalent in many areas. Thus, in attempting to eradicate the disease, there is still a necessity to study and research this disease for better understanding the pattern of the disease occurrence and prevalence, transmission of disease, diagnosis, prophylactic intervention, and management.

The range of leprosy manifestations is very wide and there is a great variation between different types of leprosy; hence both clinical and histopathological factors and bacteriological indicators are more useful than any single parameter in achieving a definitive diagnosis and classification of the disease.

The histopathological examination should be performed in all cases for the proper diagnosis of leprosy; this may assist in better provision of the patients with the appropriate treatment. As nerve damage is irreversible, early detection and treatment is important to prevent disabilities.

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Data availability

The raw data supporting the conclusions of this article are available from the authors upon reasonable request.

Conflict of interest

This being Government college and government affiliated hospital, there are no conflicts of interest.

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Medical College Vadodara being a Government Institute affiliated to Government Hospital, Funding not required

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