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HISTOPATHOLOGICAL SPECTRUM OF LEISHMANIASIS IN PUNJAB

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Abstract:

Aim: *The aim of the study was to determine the different histological spectrums of cutaneous leishmaniasis and to discover whether there are other useful histological findings in addition to the detection of amastigotes that could assist in the diagnosis.*

Materials and methods: *The study was held in the Dermatology department of Bahawal Victoria Hospital, Bahawalpur for one-year duration from March 2019 to March 2020. The total number of patients included in the study was 36. These patients were referred from various areas of the Punjab. All but one was men. Their ages ranged from 8 to 60 years. The duration of the disease ranged from 3 to 32 weeks. A skin biopsy of the most typical lesion was performed in each patient. The biopsy slide was stained with H&E dye and viewed under a microscope by one expert. Histopathological results were recorded according to the given criteria.*

Results: *The diagnosis was confirmed by detecting amastigotes (LT bodies) in the preparations. They were detected in 11 out of 36 patients (30.55%). The other most significant finding found in all formulations was the presence of a mixed infiltration of inflammatory cells in the superficial dermis.*

Conclusion: *It was found that the chances of capturing amastigote in biopsy specimens are not high. Hence, the presence of a dense, mixed infiltration of inflammatory cells in the superficial dermis should be treated as a diagnostic feature in the absence of LT bodies.*

Key words: *Cutaneous leishmaniasis, histopathology, bodies LT*

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INTRODUCTION:

Leishmania infection must be definitively diagnosed before starting chemotherapy. Laboratory diagnosis of cutaneous leishmaniasis requires the presence of parasites in smears, biopsies or isolation of the organism in a culture medium or an experimental animal. Many other methods for demonstrating the parasite (histochemical and immunohistochemical) or for detecting antibodies against leishmania (serological) have been described. Many advances have been made in these areas, but the methodology and technology related to immunohistochemistry and serology remain beyond the reach of standard laboratories. In both developed and less developed countries, laboratories continue to rely on the demonstration of parasites in Giemsa-stained smears and on processed and stained hematoxylin and eosin (H&E) biopsies. Skin biopsy for histopathological purposes is the most frequently performed examination in Pakistan. A positive report requires the presence of amastigote in the preparations. The chances of detecting amastigote in histopathological slides are not high, so it is important to identify also other histopathological findings that may contribute to the diagnosis.

MATERIALS AND METHODS:

Study design

It was a non-interventional, descriptive study.

Research objectives

The study had two goals:

1. First, to detect various histopathological features in patients with cutaneous leishmaniasis in North West Frontier Province and see if the results are similar or different from other tests.

2. Second, to determine if there are other histopathological findings in addition to the detection of LT bodies in the final diagnosis.

Study population

The study was held in the Dermatology department of Bahawal Victoria Hospital, Bahawalpur for one-year duration from March 2019 to March 2020. The total number of patients enrolled in the study was thirty-six (n = 36). All but one of the patients were male. Their ages ranged from 8 to 60 years (average 31.97 years). Disease duration ranged from 3 to 32 weeks (mean 10 weeks).

Admission Criteria

The study included patients of all ages with one or more nodules, ulcers, or crusts on exposed areas of the body for at least 3 weeks (the presence of satellite lesions or spread of sporotrichoids further supported the diagnosis). An exposure history of the disease in a known endemic area was also included as one of the inclusion criteria.

Exclusion criteria

Very young and very old patients, patients who did not consent to participate in the study and patients with questionable clinical changes were excluded from the study. Those patients who did not travel to the endemic area and those who received some definitive treatment for their disease were also excluded.

Record keeping

A typical pro forma was prepared that was used by every patient. It included the patient's first and last name, age and sex, duration of the disease, area of contact with the disease, and histopathological results according to uniform criteria.

Histopathology

Each patient underwent a skin biopsy from the edge of the most representative lesion. The sample was sent in formalin to the histopathology department. Formalin-fixed specimens were paraffin-embedded following a number of processes, ie fixation, drainage, purification and impregnation. Sections were cut with a rotary microtome (model AS-325 Shandon) followed by staining with hematoxylin and eosin. The prepared preparations were examined under a single microscope (Model CH-20 Olympus) and by only one histopathologist in consultation with a dermatologist.

RESULTS:

The results of this study are as follows:

1. Leishmania trophozoite (LT) bodies were collected on histopathological slides from 11 out of 36 patients (30.5% yield).

2. A feature visible on all histopathological slides was the presence of a dense, mixed infiltration of inflammatory cells consisting of lymphocytes, histiocytes and plasma cells in the superficial dermis (100% efficiency).

3. Neutrophils were part of this infiltration in the histopathological preparations of 26 patients (72.2%). Of these 26 preparations, neutrophilic abscesses were found in 14 patients (38.9%).

4. Epidermal ulceration was found in histopathological slides of 28 patients (77.8%), almost always associated with the presence of neutrophils in the infiltrate.

5. Epidermal hyperplasia was visible on all slides (100%).

6. Epithelial cells were found in the histopathological slides of 20 patients (55.5%), and granulomas from epithelial cells in 16 patients (44.4%). Langhans giant cells were found to be part of the granulomas in 11 patients (30%) and necrosis of the glands within the granulomas in 7 (19.4%) patients.

Clinico-pathological correlation

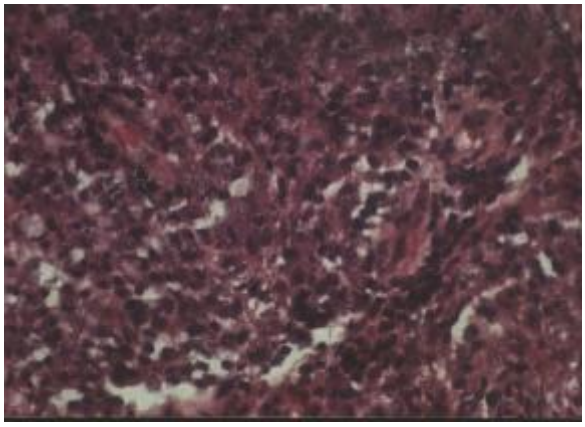
In wet and ulcerated lesions, the infiltration was usually mixed and there was a greater probability of detecting amastigote in histopathological preparations. In dry and nodular lesions, the infiltration was mixed again, but there was a greater tendency to form granulomas with less chance of surprise.

Histological patterns

In this study, four final histological patterns were identified. They were:

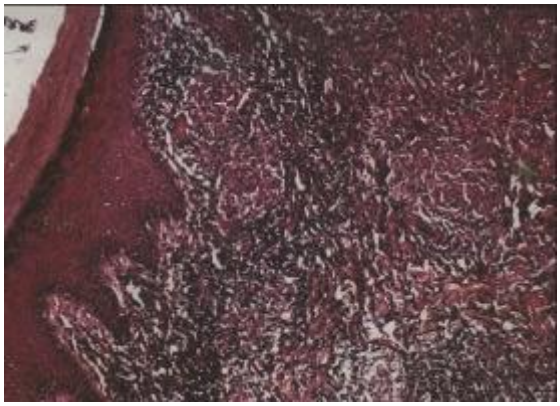
1. Mixed infiltration of inflammatory cells with LT bodies without granulomas (7 [19.5%] patients) [Fig. 1].

Figure 1: Mixed inflammatory cell infiltrate with LT bodies and no granuloma

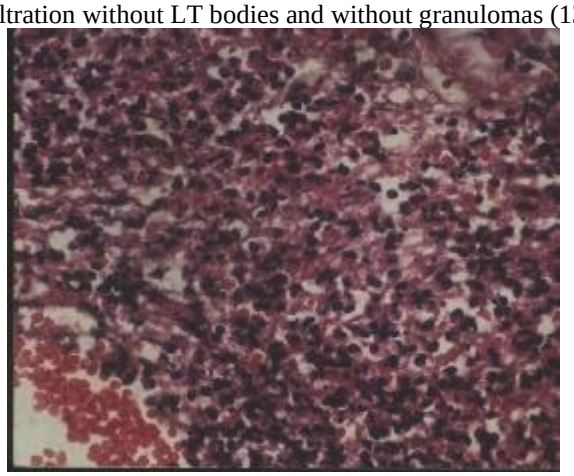


2. Mixed inflammatory cell infiltration with LT bodies with epithelial granulomas present (4 [11.1%] patients).

3. Mixed infiltration of inflammatory cells without LT bodies and with the presence of epithelial cell granulomas (12 [36.1%] patients) [Fig. 2].



4. Mixed inflammatory cell infiltration without LT bodies and without granulomas (13 [33.3%] patients) [Fig. 3].



DISCUSSION:

Four final patterns emerged from a specific area of the country. In an earlier study published in Pakistan, the histopathological results were also divided into four groups. Two groups (76% of patients) showed mixed inflammatory cell infiltration, 12% chronic inflammatory infiltration without granuloma, and 12% only tuberculosis granulomas. There was no pure tuberculosis pattern or pure mononuclear infiltration in our study. Although the histopathological examination of skin biopsy is the most frequently performed examination in patients with leishmaniasis, the chances of obtaining LT bodies are not high. In our body study, LT was detected in 11 out of 36 patients (30.5%). The results of other studies are very similar to ours. Bhutto et al. LT bodies were detected in 365 out of 1210 registered patients (diagnostic efficiency was 30.2%). Sharquie et al. amastigotes were found in 18 out of 60 patients (30% efficiency). In the study by Weigle et al. 23 slides tested positive for LT bodies out of all 165 patients (13.9% yield). Azedah et al. 6 detected amastigots in 64 of 117 slides (54.7% yield), and Simeen et al. reported a 76% yield, collecting amastigots in 38 of the 50 specimens tested. This comparison is shown graphically in Figure 4. After reviewing the above comparisons, the first question that arises is that if the percent efficiency of amastigot on skin biopsy is so low, it is such an invasive procedure, which is also costly in our environment. , reasonable diagnosis? Second, in the absence of visible LT bodies, could there be any other histological feature that could contribute to the confirmation of a diagnosis of cutaneous leishmaniasis? Features such as epidermal hypertrophy, epidermal ulceration, granuloma formation, Langhans cells, and mononuclear or polymorphonuclear infiltration are present in other related diseases, e.g. lupus vulgaris, dermatophyte infection, etc. A feature that was present on all slides in our study, regardless of time The duration of the disease was the presence of a dense and superficial mixed inflammatory cell infiltrate consisting mainly of lymphocytes, plasma cells and histiocytes. In most cases, neutrophils and epithelial cells were part of this infiltrate. Therefore, this mixed inflammatory infiltrate should be regarded as the final diagnostic feature in the absence of demonstrable LT bodies.

CONCLUSION:

The chances of detecting amastigote in histopathological preparations are not very high. A constant finding in our research was the presence of dense mixed inflammatory cells infiltrating the superficial dermis. This feature is absent from any other chronic granulomatous disease; therefore, it should be regarded as a diagnostic feature in the absence of LT bodies.

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