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## Infected Trophic Ulcers and Tenderness of Posterior Tibial Nerve in Cured Leprosy Patients

### TO THE EDITOR:

This is with reference to the letter by Dr. Anil H. Patki on the above subject in the September 1993 issue [Vol. 61 (3) pp. 473–474].

1. Very often patients with acutely inflamed plantar ulcers complain of pain (which can be quite severe) in the foot, and careful questioning reveals the site of pain as behind the tibial malleolus. The posterior tibial nerve shows exquisite tenderness in these cases. The inguinal lymph nodes are often, but not always, enlarged and tender. The nerve tenderness improves as the ulcer improves, rather slowly but steadily. Mild tenderness may persist for a while even after complete healing of the ulcer.

2. In my opinion this is not merely due to lymphangitis although that, perhaps, contributes to the clinical picture because: a) there is rarely, if ever, any associated inflammation of the superficial lymphatics in most of these cases, and b) the tenderness persists, although at a lower level, for some time even after control of the acute infection. It appears quite likely that there is a real neuritis of the posterior tibial nerve in these cases only it is not related to leprosy, as such, but to acute infection in the ulcer.

The fact that the incidence of tetanus in leprosy patients with chronic plantar ulcers appears to be very much less than one would expect has been explained on the basis of intraneural and perineural fibrosis which would also block the lymphatics (<sup>1</sup>).

3. In the old dapsone monotherapy days, this clinical phenomenon was not very much stressed and, also, it did not matter. However, the situation today is different and, as Dr. Patki says, somebody examining such a patient (who has already been treated and discharged as cured) may easily diagnose disease activity/acute neuritis/reaction/relapse just because the posterior tibial nerve is very tender. I congratulate Dr. Patki for bringing this matter to our attention and I endorse his cautionary statement.

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## Mast Cells in Lepromatous Leprosy

### TO THE EDITOR:

Leprosy is a chronic disease, caused by *Mycobacterium leprae*, which is characterized histologically by granuloma formation. The type of granuloma depends upon the cellular-immune response of the host to the organism (<sup>8</sup>). Apart from the usual cells which participate in the granuloma, such as epithelioid cells, macrophages and lympho-

cytes, mast cells also have been observed in the lesion. It has been noted that the numbers of mast cells in lesions of lepromatous leprosy are more compared to those of tuberculoid leprosy (<sup>1</sup>). The purpose of this study is to study the role of mast cells in treated and untreated cases of lepromatous leprosy.

Skin biopsies from 21 patients with lepromatous leprosy were reviewed. Classifi-

TABLE 1. Clinical data and mast cell counts in skin lesions of 11 untreated leprosy patients.

Case no.	Sex	Age (yrs.)	Duration of symptoms	Mast cell count per mm <sup>2</sup>
1	M	17	2 yrs.	19
2	F	36	2 mos.	18
3	M	28	5 mos.	27
4	F	26	Not known	35
5	M	31	6 mos.	40
6	M	36	Not known	38
7	M	34	Not known	5
8	F	61	1 yrs.	23
9	M	30	Not known	42
10	M	25	6 mos.	54
11	F	30	Not known	10

TABLE 2. Clinical data and mast cell counts in skin lesions of 10 treated leprosy patients.

Case no.	Sex	Age (yrs.)	Duration of therapy	Drugs used	Mast cells per mm <sup>2</sup>
12	M	61	8 yrs.	MDT <sup>b</sup>	24
13 <sup>a</sup>	M	36	4 mos.	MDT	33
14	M	36	4 mos.	MDT	16
15	F	50	15 yrs.	Dapsone	27
16	M	53	16 yrs.	Dapsone	13
17	F	19	6 mos.	MDT	7
18	M	50	4 mos.	MDT	51
19	F	26	4 mos.	MDT	47
20	M	42	4 mos.	MDT	43
21 <sup>c</sup>	M	31	4 mos.	MDT	35

<sup>a</sup> Same case as case no. 2.

<sup>b</sup> MDT = Multiple drug therapy.

<sup>c</sup> Same case as case no. 5.

cation of leprosy was according to the criteria of Ridley and Jopling (<sup>9</sup>). Sections cut from paraffin-embedded material were stained with hematoxylin and eosin (H&E) and Fite's stain for acid-fast bacilli (AFB). Two additional serial sections from each case were stained with the chloracetate esterase (Pararosanilin) method to detect mast cells (<sup>2</sup>). Granules of mast cells were stained pinkish red. The number of mast cells in the entire biopsy tissue was counted using a light microscope (Olympus) under a  $\times 100$  objective (oil immersion). The final mast cell count was obtained by averaging the numbers of mast cells in the two serial sections of the biopsy. The density of the mast cells was expressed as the number of cells/mm<sup>2</sup>. The Mann-Whitney U test was used to compare the mast cell numbers in two groups of lepromatous leprosy cases (untreated and treated).

The clinical data of the patients and the density of mast cells in the skin lesions are given in Tables 1 and 2. H&E sections of lesions of untreated leprosy cases revealed macrophage granulomas containing numerous solid AFB. Biopsies of treated cases showed vacuoles in the macrophages. AFB staining exhibited mainly fragmented bacilli.

There were no statistically significant differences between the mast cell counts in treated and untreated lepromatous groups ( $p = 0.83$ ).

Mast cells play an important role not only in immediate hypersensitivity reactions but

also in delayed hypersensitivity reactions (<sup>5</sup>). Mast cells are activated by cytokines such as interleukin-3 (IL-3) and IL-4 produced by the activated T cells (<sup>3</sup>). The T-cell-mediated immune response plays a major part in leprosy. A study of cytokine patterns and T-cell subsets in leprosy has been done, showing that in lepromatous lesions, typical Th-2 cytokines are high, one of which is IL-4 (<sup>11</sup>). On the other hand, reactive T cells from tuberculoid patients were of the Th-1 phenotype, and these cells produce undetectable levels of IL-4 (<sup>6</sup>). Several researchers (<sup>1,7,10</sup>) have observed the predominance of mast cells in lepromatous leprosy lesions as compared to those of tuberculoid leprosy. One would speculate that with chemotherapy the density of mast cells in lepromatous lesions would reduce. However, in the present study no significant difference in mast cell density was noted between treated and untreated lepromatous lesions. This is probably due to two reasons. Firstly, the life span of mast cells is long, ranging from weeks to months (<sup>7</sup>). In our study, skin biopsy in 7 of the 10 patients in the treated group was done within 6 months of chemotherapy. Perhaps 6 months is too short a period to expect reduction in the mast-cell density in the lesions. Secondly, it is possible that reduction of mast cell density occurs only when the lesions upgrade to become more of the tuberculoid type, with changes in the cytokine pattern. We did not note any upgrading of the lesions in the treated group.

The histology of the lesions in the treated group showed vacuolated macrophages containing fragmented bacilli, indicating only a response to chemotherapy.

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## Cell-Mediated Immunity in Skin Tuberculosis

TO THE EDITOR:

A small study was undertaken to observe the lymphoproliferative responses in the peripheral blood of patients with secondary skin tuberculosis, both before and after therapy.

Fifteen patients, 9 suffering from lupus vulgaris (LV) and 6 from scrofuloderma (SD), aged between 7 and 25 years with disease durations ranging from 3 months to 4 years, and 10 age-matched controls were studied separately for evaluation of antigen-induced lymphocyte transformation tests (LTT) to assess lymphocyte function. All of the patients and controls were clinically examined, X-rayed to rule out pulmonary tuberculosis, and blood was drawn prior to other investigations. A Mantoux test was done and read after 48 hr. The induration on the forearm measured between 16 mm and 30 mm in diameter in 13 and 10 mm in 2. Clinical diagnosis was confirmed by histopathology. The patients were then giv-

en the standard regimen (6) consisting of rifampin and isoniazid for 6–9 months supplemented during the first 2 months with pyrazinamide. The test was repeated after completion of therapy when the lesions had subsided. One patient was lost to follow up. None of the patients was taking corticosteroids during the period of study.

Soluble antigens used were purified protein derivative (PPD) of *Mycobacterium tuberculosis* at a concentration of 100 µg/ml and tetanus toxoid (1:10). Integral *M. leprae* bacilli were extracted from human lepromas as described (2), heat killed and used at an optimal concentration of  $5 \times 10^6$ /ml.

Peripheral blood mononuclear cells (PBMC) were separated from venous blood by the previous method (2) and suspended in RPMI-1640 with 10% fetal calf serum (FCS) at concentrations of  $4-5 \times 10^6$ /ml; 100 µl of these cells were then cultured in a 96-well plate in the presence of antigens (PPD, tetanus toxoid and *M. leprae*) and