Indirect immunofluorescence to demonstrate lichen planus specific antigen (LPSA) in lichen planus

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ABSTRACT

Background: Current evidence suggests that lichen planus is an immunological disease. Cytotoxic CD8+ cells in the lesional epidermis recognize a unique antigen called lichen planus specific antigen. This antigen could be demonstrated by indirect immunofluorescence using the patient's serum and autologous lesional skin. **Aim:** To study indirect immunofluorescence pattern in lichen planus, among Indian patients. **Methods:** Twenty-five consecutive patients with the clinical diagnosis of lichen planus were enrolled in the study. Direct immunofluorescence was done in all patients. Indirect immunofluorescence using lesional skin as substrate was done in all 25 patients and five patients with other dermatoses. **Results:** A specific fluorescence pattern corresponding to the distribution of lichen planus specific antigen was observed in the stratum spinosum and granulosum in 22 (88%) patients. It was absent from other parts of the epidermis, dermis and in patients with other dermatoses. **Conclusion:** Indirect immunofluorescence is a useful adjuvant test in lichen planus, particularly in atypical cases.

Key Words: Indirect immunofluorescence, Lichen planus, Lichen planus specific antigen

INTRODUCTION

The histopathological criteria for the diagnosis of lichen planus are well established in textbooks of dermatopathology. Briefly, they consist of hyperkeratosis, wedge-shaped hypergranulosis, irregular acanthosis, vacuolar degeneration of basal layer with the presence of intraepidermal or subepidermal colloid bodies, saw-toothing of rete-ridges and papillary dermal band-like infiltrate.[1] Direct immunofluorescence (DIF) of lesional skin further supports the diagnosis. It shows ragged fibrin band at basement membrane zone (BMZ) and clusters of colloid bodies (with IgM and C_3; to a lesser extent with other classes of immunoglobulin) in almost all patients.[2] Though histopathological and DIF findings are highly characteristic of lichen planus, they are not specific. Transitional histopathological and DIF features could be seen in various other dermatoses such as lupus erythematosus and dermatomyositis.[3] Previous studies have reported the usefulness of indirect immunofluorescence findings in lichen planus.[4,5] Our aim was to establish its role in Indian patients.

METHODS

Twenty-five patients with the clinical diagnosis of lichen planus were enrolled in the study. A 4-mm punch biopsy was taken from the lesional skin, snap

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frozen and stored in the deep freezer at -70°C. Using cryostat, 4 μm sections were taken. Patient's serum was obtained after collecting 5 ml of blood. Serum (1:10 and 1: 80 dilutions) was incubated with lesional skin sections for one hour. Later it was washed in phosphate buffer saline (PBS) and incubated with various fluorescein isothiocyanate conjugates for one hour. It was once again washed in PBS, mounted in buffered glycerol and examined under fluorescent microscope. DIF of lesional skin was done in all patients. Five patients with other dermatoses were used as controls (two patients with SLE, one case each of scleroderma, MCTD and atopic dermatitis) and indirect immunofluorescence was done using the same technique.

RESULTS

Among 25 patients 13 were males and 12 females. Average age of the patients in the study group was 39.72 years (range 13 - 63 years). Mean duration of illness was 2.72 years (range 15 days - 5 years). Various clinical types of lichen planus encountered are shown in Table 1. Oral lesion in the form of lacy reticular net-like pattern was seen in three patients. Results of DIF are shown in Table 2.

Indirect immunofluorescence using autologous lesional skin showed characteristic fluorescent IgG deposits in the upper epidermis, at the level of the stratum granulosum and stratum spinosum in 22 (88%) patients [Figure 1]. Such findings were not seen in other parts of the epidermis, BMZ or in the dermis.

Table 1: Variants of lichen planus

Type	No. of cases
Classical	16
Hypertrophic	04
Generalized	02
Vesicular	01
Linear	01
Actinic	01

Table 2: Direct Immunofluorescence in lichen planus

DIF finding	No. of patients (%)
Ragged fibrin band	25 (100)
IgM colloid body	21 (84)
C3 colloid body	15 (60)
IgA colloid body	11 (44)

DIF: Direct immunofluorescence

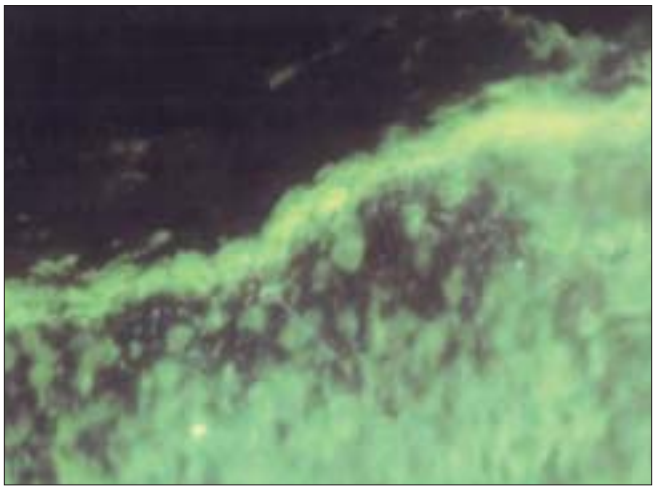

Figure 1: IgG band at the level of stratum granulosum (IIF, 200x)

Direct immunofluorescence did not reveal any such finding. The test was negative in one patient who was on oral steroid therapy; however it was positive in two others, who were also on oral steroids at the time of the test. Among the patients with negative test, one had classical lichen planus (LP) lesions and was on steroid while the other two were not on any medication and had hypertrophic and generalized type of LP respectively. The test was negative in the control group.

DISCUSSION

LP is a papulo-squamous disease of the skin and mucous membrane of worldwide distribution. The exact etiology of LP remains unknown. There is little evidence to support the older hypothesis of viral origin, neurological changes or psychological stress as sole causative factors.[6] Recent studies have shown that LP represents a cell-mediated immune response to an induced antigenic change in the epidermal cells in a genetically predisposed individual.[7] CD8+ infiltrates in the lesional skin recognize a MHC Class I antigen called lichen planus specific antigen (LPSA). The exact nature of this antigen is unknown. It may be an auto-reactive peptide or exogenous antigen such as altered protein, drug, contact allergen and viral or other infectious agent.[8] In 1983, Olsen *et al*. demonstrated LPSA by indirect immunofluorescence technique using autologous lesional skin.[4] It was present only in the stratum granulosum and stratum spinosum and absent from other parts of the

epidermis, BMZ and dermis. Lichen planus specific antigen (LPSA) is specific for LP and found in about 80% of the patients with or without oral lesions.[5] In our study, we found LPSA in 88% of patients. Though the test was negative in one patient who was being treated with systemic corticosteroids, it was positive in the other two who were also receiving steroid therapy. We feel that corticosteroid therapy was not responsible for the negative finding. Olsen *et al*. made a similar observation. They suggest that LPSA is not produced in all LP lesions or absent in the lesion during some stages of the disease or that it may not always be detectable by this method.[5]

Earlier studies have demonstrated LPSA in patients belonging to diverse ethnic backgrounds such as Caucasians, Negroes and Koreans.[5,6] Our study revealed LPSA among Indian patients with LP. LPSA can be demonstrated in various clinical types of LP such as hypertrophic, vesicular, actinic and eruptive, as shown in our study. Camisa *et al*. have shown LPSA in bullous LP,[9] while Olsen *et al*. have demonstrated the usefulness of this technique in LP / LE overlap syndrome. They concluded that this technique could help to differentiate atypical cases of LP from other dermatoses such as lupus erythematosus where DIF shows equivocal results.[10]

LPSA is unique to lichen planus and is expressed in the stratum granulosum and stratum spinosum of patients with LP. It can be demonstrated by indirect immunofluorescence using the patient's serum and autologous lesional skin. This test is of particular help to differentiate atypical cases of lichen planus from other dermatoses.

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