Section: Pathology

Original Research Article

QUANTITATIVE COMPARISION OF MICROVESSEL DENSITY IN LICHEN PLANUS LESION AND ADJOINING NORMAL SKIN BY CD34

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ABSTRACT

Background: Angiogenesis is a key component in both neoplastic and inflammatory disorders. But whether it occurs in inflammatory skin disorders such as Lichen Planus is yet to be established. This can be done by evaluating Microvascular density and comparing it to the normal skin. **Aims and Objectives:** Quantitative comparison of Microvessel density in Lichen Planus lesion and adjoining normal skin by CD34 Immunohistochemical marker and to study characteristic histological features in favor of Lichen Planus diagnosis.
Materials and Methods: A case control study was carried out on Quantitative comparison of microvessel density in lichen planus and adjoining normal skin by CD34 immunohistochemical marker in the department of Pathology.
Results: Total 30 cases of Lichen Planus were evaluated. M: F ratio was 1.73: 1 In histological features, band of infiltrates and basal cell vacuolation are seen in all the cases. Microvascular density Mean in the lesion was 9.94 compared to 4.84 in the adjacent normal skin.
Conclusion: Pathogenesis of Lichen Planus mainly is due to chronic inflammatory disease with probable autoimmune component. MVD is not a characteristic histological feature of Lichen Planus in differentiating from its mimickers. Further studies are recommended based on molecular and genetic pathogenesis of Lichen Planus using ancillary techniques so that targeted therapies can be administered.
Keywords: Lichen Planus, Microvascular density, histopathology.

INTRODUCTION

Lichen Planus is an idiopathic subacute or chronic inflammatory disease of the skin, mucous membranes and nails.[1] Exact pathogenesis of Lichen Planus is still unclear. Several hypotheses have been made regarding its etiology, including genetic, infective, psychogenic and autoimmune factors. Recent studies provide evidence that autoreactive cytotoxic T lymphocytes are the effector cells which cause degeneration and destruction of keratinocytes.[2] Cutaneous Lichen Planus is characterized by polygonal flat-topped, violaceous papules and plaques, which in some cases can be intensely itchy. The lesions may result in long- standing residual hyperpigmentation, especially in dark-skinned patients. Lichen Planus has characteristic histopathological features which make the diagnosis relatively easy. Cutaneous Lichen Planus has worldwide distribution with its incidence varying from 0.22% to 1% depending upon geographic location.1 According to one study Lichen Planus represents 0.38% of all dermatology out- patients in India.[3] However, there is a paucity of Indian literature describing the role of angiogenesis in the development of chronic inflammatory diseases including Lichen Planus is of great concern. The most representative method for the assessment of angiogenesis is the quantification of Microvessel density (MVD) using specific markers such as CD 105, VEGF and CD34.[4]

This study was undertaken in a tertiary care centre where the cases of Lichen Planus are frequent and to understand the pathogenesis of Lichen Planus expecting a significant increase in microvascular density using CD34 marker compared to normal because there are only a handful of studies which used CD34 to quantify microvascular density. This may also be helpful in diagnosing Lichen Planus from lichenoid lesions.

MATERIALS AND METHODS

The study design was a hospital based prospective case control study done in Department of Pathology conducted for the period of 2 years from August 2020 to July 2022. All the clinical and histopathological cases diagnosed as Lichen Planus received during the study period in the Department of Pathology, MediCiti Institute of Medical Sciences, Ghanpur, Telangana were studied. A total of 30 samples were included in the study.

Inclusion Criteria: All the cases diagnosed as Lichen Planus clinically and confirmed by histopathology

Exclusion Criteria: Treated cases of Lichen Planus, Cases without adjacent normal skin and Clinically diagnosed Lichen Planus which didn't correlate on histopathology.

Prior to the commencement, the study was approved by the Ethical and Research Committee. The received samples were fixed in 10% formalin and routinely embedded in paraffin. Sections of 4-µm thickness were cut from the paraffin blocks and were used for immunohistochemical staining.

Immunohistochemical staining was performed for CD34 antibody (Dako). Briefly, endogenous peroxidase activity was blocked with 0.6% H2 O2 and sections were incubated at room temperature with 100 µl of prediluted primary CD34 antibody (Dako). After washing in phosphate-buffered saline (PBS), secondary antibody was applied on the section and then washed with PBS. Diaminobenzidine Hyrochloride was applied till a brown color appeared. After counterstaining with hematoxylin, the sections were mounted with dibutyl phthalate xylol (DPX) and the slides were observed under a Nikon® research microscope. Negative and positive controls were used simultaneously to assess the specificity and reliability of the staining process. PBS was substituted for the primary antibodies of CD34 as a negative control, and placental tissue sections with known CD34 positivity were used as the positive control.

Quantification of microvessel density

Specimens were assessed blinded using a light microscope, and brown staining of the cytoplasm was considered as positive. Under 40× magnification, the sites of the greatest vascularity (hotspots) were selected, and then, the number of microvessels were counted under 400× magnification from five fields of view.

Statistical analysis: The data obtained was coded and entered into Microsoft Excel Worksheet. The data was analysed using statistical software SPSS Version 20.0. The categorical data was expressed as rates, ratios and proportions and continuous data was expressed as mean ± standard deviation (SD).

RESULTS

In present study out of 30 cases, 19 cases were male and 11 cases were females, showing male predominance with M:F ratio 1.73:1. Out of the 30 cases, the most common age group is 31 – 40 years with 9 cases accounting for 30% of the total cases. The mean age is 34.6 years.

Table 1: Age and gender distribution of cases

Age	Male	Female	Total
11-20yrs	2(10.5%)	2(18.18%)	4(13.3%)
21-30yrs	6(31.60%)	2(18.18%)	8(26.7%)
31-40yrs	5(26.3%)	4(36.36%)	9(30%)
41-50yrs	2(10.5%)	1(9.1%)	3(10%)
51-60yrs	3(15.80%)	2(18.18%)	5(16.7%)
>61yrs	1(5.3%)	0(0%)	1(3.3%)
Total	19(100%)	11(100%)	30(100%)

Out of the 30 cases, the most common histopathological feature was Basal cell vacuolation and band of infiltrate which were present in all cases (100%).

Table 2: Histopathological features seen in the cases

Histopathological feature	No. of cases(%)
Hyperkeratosis	21(70%)
Hypergranulosis	19(63.33%)
Acanthosis	24(80%)
Sawtoothing of rete ridges	22(73.3%)
Basal cell vacuolation	30(100%)
Band of Infiltrates	30(100%)
Predominant cell type(lymphocytes)	28(93.3%)

Out of the 30 cases, Hyperkeratosis was seen in 21 cases making up 70% of the total cases.

Figure 3: Color Plates

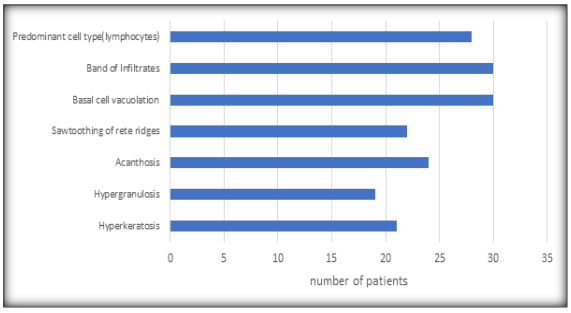


Figure 1: Figure 1: Bar diagram showing histological features in present study

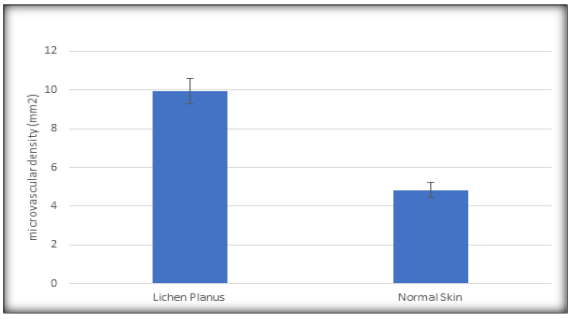
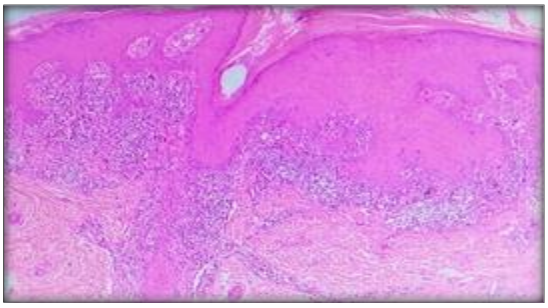
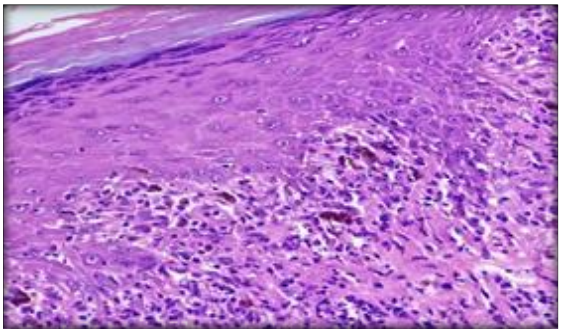


Figure 2: Microvascular density in the lesion in comparison with adjacent normal skin

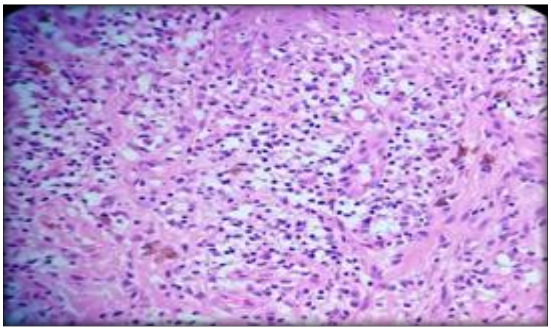
Clinical picture of histologically diagnosed lichen planus



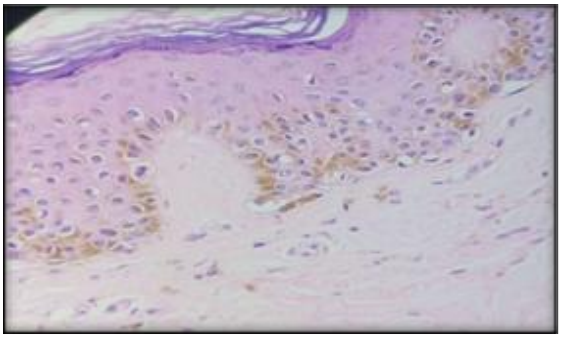
Showing irregular acanthosis and saw toothing of rete ridges and band of inflammatory infiltrates.



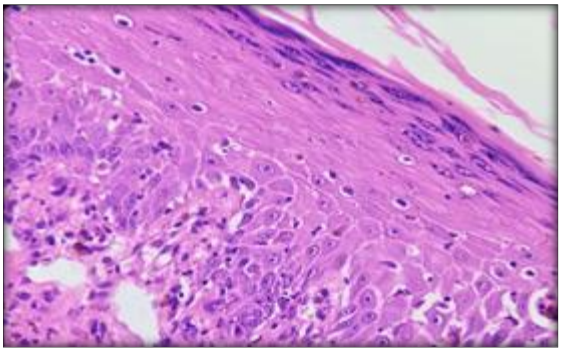
Showing pigment incontinence, basal cell vacuolation and lymphocytic infiltration



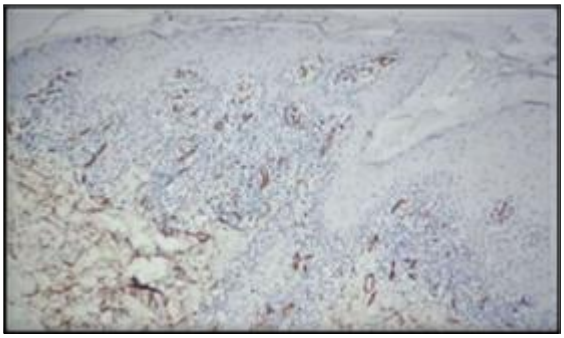
Proliferating capillaries in upper dermis



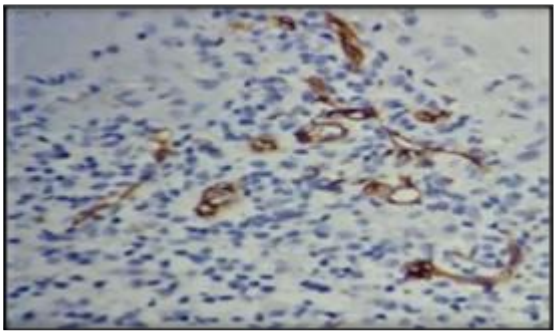
Less number of capillaries in adjoining normal skin



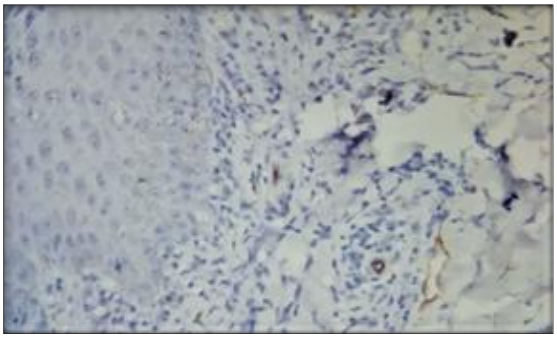
Hypergranulosis in epidermis



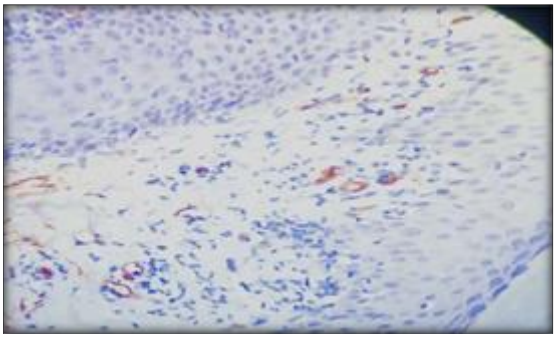
Showing capillaries in papillary dermis highlighted by cd34 (low power)



Increased capillaries in papillary dermis (high power view)



Decreased capillaries in adjoining normal skin



Less number of capillaried in adjoining normal skin

DISCUSSIONS

In present study out of 30 cases, 19 cases were male and 11 cases were females, showing male predominance with M:F ratio 1.73:1. In a study done Srivani N et al5 showed male predominance in which out of 50 cases 27 cases were male and 23 cases were female with a male: female ratio of 1.17:1. This study is in concordance with the present study

In present study out of 30 cases maximum no. of cases (9/30) were between 31-40 years of age. Next common age group was 21-30 years with 8 cases.

Hyperkeratosis was present in 21 cases. Hypergranulosis was present in 19 cases. Out of 19 cases of hypergranulosis, 3 showed wedge shaped hypergranulosis.

Acanthosis was present in 24 cases. In all positive cases the acanthosis was irregular. Saw toothing of rete ridges was present in 22 cases. Basal cell vacuolation and band of inflammatory infiltrates were present in all 30 cases. The predominant cell type in these infiltrates were lymphocytes seen in 28 cases. Only 2 cases showed eosinophilic infiltrate.

Table 3: Comparison of histopathological features with other studies

Histopathological feature	Srivani N et al[5]	Fernandez Gonzalez F et al[6]	PRESENT STUDY
Hyperkeratosis	38 (75%)	33 (66%)	21 (70%)
Hypergranulosis	Not Applicable	Not Applicable	19 (63.33%)
Acanthosis	35 (70%)	24 (48%)	24 (80%)
Saw toothing	27 (53%)	Not Applicable	22 (73.33%)
Basal cell vacuolation	50 (100%)	50 (100%)	30 (100%)
Band of infiltrates	43 (86%)	50 (100%)	30 (100%)
Lymphocyte as predominant cell type	50 (100%)	50 (100%)	28 (93.33%)

The differential diagnosis of cutaneous Lichen Planus is broad and includes graft-vs. host-disease, psoriasis vulgaris, guttate psoriasis, secondary syphilis, pityriasis lichenoides, pityriasis rosea, lichen nitidus, lichen simplex chronicus, lichen sclerosus, lichen striatus, linear epidermal naevus, eczema, prurigo nodularis, erythema dyschromicum perstans, eczematid-like purpura, drug eruption, granuloma annulare, lichen amyloidosus, Kaposi sarcoma and lupus erythematosus.

Among all histological features, basal cell vacuolar degeneration and band of lymphocyte infiltration are the most characteristic features of Lichen Planus. Different types of histological features are helpful in diagnosing Lichen Planus from its mimickers. For example, differentiating from Psoriasis vulgaris, it's the hypergranulosis which is the most helpful feature of Lichen Planus.

Microvessel density which is assessed using CD34 showed increased MVD in cases of lichen planus when compared to adjoining normal skin. The mean of MVD in lichen planus is 9.94 which is higher than the mean of adjoining normal skin 4.84 with the respective standard deviations of 0.67 and 0.38. This

is in concordance with study done by Mahmoud. R. Hussein et al,[7] in which Lesion showed more MVD when compared to the adjacent normal skin.

Table 4: Comparison of microvascular density (mean) with other studies

Studies	MVD (Mean) in Lichen Planus Lesion	MVD (Mean) in adjacent normal skin
Mahmoud R. Hussein et al[7]	2.5	1.39
Present Study	9.94	4.84

Other studies done by Poornima Parvathala et al8 took CD 105 (Endoglin) as a marker of Microvascular density showing a mean MVD of 1.68 in Lichen Planus lesion and 1.31 in adjacent normal. A hypothesis to be tested is that the increased MVD at the dermo-epidermal junction is associated with increased recruitment of the lymphocytes and hence the level of this adhesion molecule.

Angiogenesis and lymphangiogenesis—the growth of new vessels from existing ones—have received tremendous interest because of their role in promoting cancer spread. However, there is increasing evidence that both vessel types also have a major role in acute and chronic inflammatory disorders. Vessels change their phenotype during inflammation (vascular remodelling). In inflamed skin, vascular remodelling consists of a hyperpermeable, enlarged network of vessels with increased blood flow, and influx of inflammatory cells. During chronic inflammation, the activated endothelium expresses adhesion molecules, cytokines, and other molecules that lead to leukocyte rolling, attachment, and migration into the skin. Recent studies reveal that inhibition of blood vessel activation exerts potent anti- inflammatory properties.[10,11]

It can be argued that MVD is more important in Papillary dermis as it is the most topmost layer of the dermis and provides nutrition to the epidermis, which is critical for maintaining the overall health and integrity of the skin. Additionally, MVD in the Papillary dermis plays a key role in wound healing. However, the reticular dermis is also important in the MVD as it provides structural support to the skin, helps to maintain its shape, elasticity and temperature of the skin.

CONCLUSION

In the present study, using IHC marker of CD 34 for microvascular density (MVD) concluded that the pathogenesis of Lichen Planus mainly is due to chronic inflammatory disease with probable autoimmune component. MVD is not a characteristic histological feature of Lichen Planus in differentiating from its mimickers. Further studies are recommended based on molecular and genetic pathogenesis of Lichen Planus using ancillary techniques so that targeted therapies can be administered.

REFERENCES

1. Boyd AS, Neldner KH. Lichen planus. Journal of the American Academy of Dermatology. 1991 Oct 1;25(4):593-619.
2. Sontheimer RD. Lichenoid tissue reaction/interface dermatitis: clinical and histological perspectives. Journal of Investigative Dermatology. 2009 May 1;129(5):1088-99.
3. Bhattacharya M, Kaur I, Kumar B. Lichen planus: a clinical and epidemiological study. The Journal of dermatology. 2000 Sep;27(9):576-82.
4. Mysore V. Fundamentals of pathology of skin. Wolters kluwer india Pvt Ltd; 2015.(6-19)
5. Srivani N, Sravani B, Srujana S, Kumar O. A study of clinical and histopathological correlation of lichen planus. International Archives Of Integrated Medicine. 2017;4(9):136-44.
6. Fernández-González F, Vázquez-Álvarez R, Reboiras-López D, Gándara-Vila P, García-García A, Gándara-Rey JM. Histopathological findings in oral lichen planus and their correlation with the clinical manifestations. Med Oral Patol Oral Cir Bucal. 2011 Aug 1;16(5):e641-6.
7. Hussein MR. Evaluation of angiogenesis in normal and lichen planus skin by CD34 protein immunohistochemistry: preliminary findings. Cell biology international. 2007 Oct 1;31(10):1292-5.
8. Parvathala P, Baghirath PV, Reddy CN, Vinay BH, Krishna AB, Naishadham PP. Horoscopic role of CD105 (Endoglin) in progression of oral lichen planus: An immunohistochemical study. Journal of Oral and Maxillofacial Pathology: JOMFP. 2021 Jan;25(1):37.
9. Huggenberger R, Detmar M. The cutaneous vascular system in chronic skin inflammation. InJournal of Investigative Dermatology Symposium Proceedings 2011 Dec 1 (Vol. 15, No. 1, pp. 24-32). Elsevier.
10. Boch K, Langan EA, Kridin K, Zillikens D, Ludwig RJ, Bieber K. Lichen planus. Frontiers in Medicine. 2021:2089.
11. Gisondi P, Bellinato F, Girolomoni G. Topographic differential diagnosis of chronic plaque psoriasis: challenges and tricks. Journal of Clinical Medicine. 2020 Nov 8;9(11):3594.