ORIGINAL RESEARCH PAPER

Pathology

HISTOCHEMICAL ANALYSIS OF LICHEN PLANUS AND LICHENOID LESIONS.

KEY WORDS: Lichenoid, PAS, Lichen planus.

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ABSTRACT

Lichenoid eruptions represent a heterogenous group with variable histopathological features and often present as diagnostic challenge to the pathologist. We aimed to study the histopathological changes in lichen planus and lichenoid lesions and evaluate the role of special stains in their diagnosis. A retrospective study was conducted in 42 lichenoid lesions in which the cases were divided into two groups including 22 cases of lichen planus and its variants(LP) and 20 cases of other lichenoid lesions(OLL). These cases were studied in routine Hematoxylin & Eosin(H&E) as well as Periodic acid Schiff(PAS) and Toluidine blue stain(TB). The results were analysed using Mann Whitney U test and Fischers exact test. Few histopathological features and thickened PAS positive basement membrane were significantly associated with LP and its variants and turned out to be helpful parameters in making the right diagnosis.

INTRODUCTION

Lichen Planus(LP) is an inflammatory skin condition with characteristic clinical and histopathological findings. Classic LP typically presents as pruritic, polygonal, violaceous, flat topped papules and plaques. Many variations in morphology and location also exists. The clinical presentation of the rarer variant lesions may be largely dissimilar to classic LP and therefore difficult to diagnose solely based on the clinical examination. Histopathological examination, although reveals similar features and aids in proper diagnosis.[1]Many clinically distinct inflammatory dermatoses also have in common varying elements of histopathological features and these are called as lichenoid dermatoses.[2] The clinical spectrum of lichenoid tissue reactions is wider with most of the diseases showing basal cell damage and a band like lymphocytic infiltrate that hugs the dermo-epidermal junction, except for subtle differences that define the particular variant.[3]

AIMS & OBJECTIVES

The aim of our study was to compare the histopathological features of Lichen planus(LP) and compare them with other lichenoid lesions (OLL).We also studied the utility of histochemical stains as Periodic Acid Schiff stain (PAS) and Toluidine blue(TB) stain in diagnosing LP and OLL.

MATERIAL & METHODS

A retrospective study was conducted in the department of pathology where 42 lichenoid lesions reported from August 2017 to July 2018 were taken out from the archival formalin fixed paraffin embedded tissue. The cases were divided into two groups including 22 cases of lichen planus(LP) and its variants(including Lichen planopilaris and Lichen planus pigmentosus) and 20 cases of other lichenoid lesions(OLL).

TABLE 1: Histopathological diagnosis of Lichen Planus and other lichenoid lesons.

Diagnosis	Number of patients	Percentage (%)
Lichen Planus	14	33.3
Lichen planus pigmentosus	4	9.52
Lichen planopilaris	4	9.52
Pityriasis Lichenoides chronica	5	11.9
Lichen sclerosis atrophicus	4	9.52
Lichen simplex chronicus	2	4.76
Lichen spinulosis	2	4.76
Lichen amyloidosis	3	7.14
Discoid Lupus Erythematosus	2	4.76
Lichenoid dermatitis	2	4.76
TOTAL	42	100

Age, sex and clinical presentation of patients from both the groups were recorded from the clinical requisition forms. The paraffin embedded blocks of the cases of both LP and OLL were cut into three slides each for routine Hematoxylin & Eosin(H&E) as well as Periodic acid Schiff(PAS) and Toluidine blue stain(TB). Based on the standard histopathological criteria previously diagnosed cases of LP and OLL were re-evaluated by 2 independent observer pathologists. PAS staining was used to see basement membrane thickness which as coloured magenta. The PAS stained positive basement membrane were measured using an eye piece reticule at 400X magnification. The mean of 3 values were expressed in micrometers(μm).One percent Toluidine blue in 1% Sodium chloride was used to stain mast cells in areas of basal cell degeneration of which dark reddish purple staining was taken to be positive. The mast cells in each case were counted as average number seen per 10 high power fields(HPF). The results were compared between the LP and OLL groups. Areas of basal cell degeneration were correlated with PAS stain The results were analysed using various statistical tests (Mann Whitney U test and Fischers exact test).

OBSERVATION & RESULTS

The overall age of presentation of the lichenoid lesions grouped together was 27.92 years[Standard Deviation(SD)=11.53]. In the LP group, the mean age of presentation was 26.54 years (SD=11.40) and 28.55 years (SD=1.60) in OLL group. There was no significant difference between age of presentation in both LP and OLL groups.(p=0.5892)

Overall in both groups, majority of patients were males (54.7%) with no significant correlation with any of the groups.Similarly most common site of presentation was in the leg in 66.66% cases, however it was also not associated with any of the groups.

Among the LP group, the most significant histopathological parameters were noted to be melanin incontinence ,increased vascularity in the papillary dermis along with band like inflammatory infiltrate ugging the dermo-epidermal junction. In the epidermis,hypergranulosis, hyperkeratosis,civatte bodies and liquefactive degeneration of the basal layer were signicant features of LP ($p<0.05$).

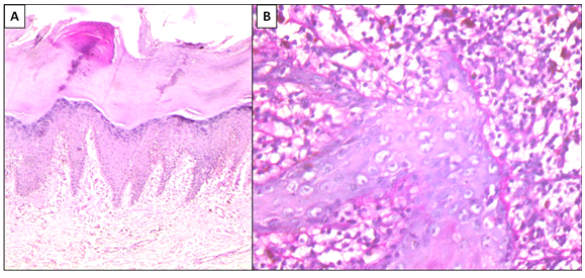
TABLE 2: Correlation of various histopathological parameters and their significance (p value) in cases of Lichen Planus(LP) and other lichenoid lesions(OLL).

Parameter	Group	Present	Absent	p value
1.Hypergranulosis	LP	13	9	p=0.0135
	OLL	4	16	
2.Parakeratosis	LP	0	22	p=0.0993
	OLL	3	17	

3.Hyperkeratosis	LP	10	12	p=0.0167
	OLL	2	18	
4.Acanthosis	LP	6	16	p=0.4595
	OLL	3	17	
5.Atrophy	LP	4	18	p=1.00
	OLL	4	16	
6.Spongiosis	LP	12	10	p=1.00
	OLL	10	10	
7.Papillomatosis	LP	2	20	p=0.4004
	OLL	4	16	
8.Saw tooth rete ridges	LP	11	11	p=0.0577
	OLL	4	16	
9.Civatte bodies	LP	6	16	p=0.0202
	OLL	0	20	
10.Liquefactive degeneration of basal layer	LP	16	6	p=0.0286
	OLL	7	13	
11.Follicular plugging	LP	5	17	p=1.00
	OLL	5	15	
12.Band like infiltrate hugging basal layer	LP	12	10	p=0.0031
	OLL	2	18	
13.Lymphocytes	LP	21	1	p=1.00
	OLL	20	0	
14.Melanin incontinence	LP	19	3	p=0.0001
	OLL	5	15	
15. Increased vascularity papillary dermis	LP	14	8	p=0.0058
	OLL	4	16	

Thickening of the PAS positive basement membrane was a significant feature of LP (p=0.0374). The mean thickness of LP and OLL groups was 12.09µm (SD=6.240) and 5.70µm(SD=2.515) respectively. Civatte bodies were also highlighted very well and could easily be identified on PAS staining.

Figure 1: (A) Classical histopathology of Lichen planus in one of the cases (H&E stain, 40X magnification)(B) Thickened magenta coloured basement membrane (H&E stain,400X magnification)



The TB stain did not show any mast cells in any of the cases in our study. However, a pitfall in screening was noted that melanophages which stained dark coloured could be easily mistaken for mast cells, but the confusion can be solved by comparing with the corresponding H&E sections.

DISCUSSION

The clinical presentation of classic LP and its variants may be largely dissimilar to each other.[1] On the other hand, many clinically distinct inflammatory dermatoses have in common varying amounts of lichenoid histological features. Lichenoid eruptions represent a heterogenous group of conditions that resemble LP in terms of their clinical appearance.[2] In the present study no significant association of age, sex and site could be identified this was similar to Sehgal et al on their study of 147 cases.[4] No significant difference was seen on comparing these characteristics among the LP and OLL group.

While no specific gender preference has been recognized in our study, LP may affect more adult female than adult males.5

It is important to distinguish LP from OLL on microscopic examination because of different treatment modalities as well as prognosis.6 The differential diagnosis can be narrowed down by focusing upon the distinctive histopathological features of the various lichenoid eruptions. Consistent histopathological features have been pointed out fo Lp such as wedge shaped hypergranulosis, marked hyperkeratosis and saw toothing like acanthosis of rete ridges. The dermo- epidermal junction typically shows signs of vacuolar degeneration with apoptotic keratinocytes, while the upper dermis shows a dense band like lymphocytic infiltrate obscuring the junction. Civatte bodies hypothesized to be apoptotic keratinocytes ready for phagocytosis can be seen in the epithelium as well as in the upper dermis. Direct immunoflourescence also shows large number of IgM staining cytoid bodies in the dermal papillae or peribasilar areas.1 In the present study , hyperkeratosis, hypergranulosis, civatte bodies and degeneration of basal layer in the epidermis as well as dermal melanin incontinence, increased vascularity and band like infiltrate of lymphocytes emerged as significant histopathological parameters of LP helpful to differentiate from OLL and other clinical diagnosis.

There was significant increase in PAS positive basement membrane thickness in cases of LP. This was similar to a study by Juneja et al[7] on oral LP. PAS is a staining method to detect polysaccharides manifested as glycogen, glycoproteins, glycolipids and mucin in tissue.[8,9] Velez et al [10]also similarly reported increased PAS positivity in the basement membrane and compared it with Direct immunoflourescence(DIF) immunoreactivity Their results showed that PAS positivity goes hand in hand with DIF immunoreactivity. staining the basememnt membrane zone. They also reported similar positivity (both PAS and DIF) on civatte bodies in LP. They speculated that these bodies may contain polysaccharides and hence are better highlighted on PAS stained sections. A recent review[11,12]postulated" The altered glycan theory of immunoreactivity" suggesting that each autoimmune disease may have an exclusive glycan signature characterized by site site specific relative protrusions of individual glycan structures on immune cells and extracellular proteins this is related to site specific glycosylation patterns of individual immunoglobulin classes and subclasses.

Unlike other studies on oral LP[7,13], however, we did not find any mast cells in cases on skin. These studies[14,15]have demonstrated role of increased mast cell density, degranulation as well as released chemokines and cytokines in regulating immune responses and activating T cells in oral lichenoid tissue reactions. We authors, recommend further large scale studies on mast cells to elucidate these findings on the skin lesions.

CONCLUSION

We conclude that histopathological parameters are very helpful in proper diagnosis of lichenoid lesions. PAS staining in each case can support the histopathological findings as well as highlight some useful details such as basement membrane thickness and civatte bodies. Proper implementation of these stains can go a long way in distinguishing various disease entities under broad category of lichenoid tissue reactions and hence improve the diagnosis for appropriate treatment.

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