Original Research Article

Clinico-histopathological Study of Vesiculobullous Lesions of the Skin at Tertiary Care Centre

Authors

Shekhawat Pratibha¹, Yadav Ajay²*, Udawat Hema³, Ranawat Chandraseet Singh⁴, Kotia Suman⁵

¹,⁵Resident, ²Senior Professor, ³Associate Professor
Department of Pathology, SMS Medical College, Jaipur, Rajasthan, India
⁴Senior Resident, Department of Neurology, SMS Medical College, Jaipur, Rajasthan, India

*Corresponding Author

Yadav Ajay

Abstract

Introduction: Vesiculobullous eruptions are encountered in various dermatosis, which include various inflammatory, infective, autoimmune, drug induced as well as genetic. Primary lesion in vesiculobullous diseases is a vesicle or a bulla involving skin and/or mucous membrane.

Aim: To study the histomorphological features of various vesiculobullous disorders of skin with clinical correlation.

Materials and Methods: A laboratory based cross-sectional observational study was conducted over a period of 10 months from June 2018 to March 2019 in the Department of Pathology, S.M.S Medical College, Jaipur. 55 clinically diagnosed cases of vesiculobullous lesions of the skin included in the study.

Results: Out of 55 cases of vesiculobullous disorders Pemphigus vulgaris constituted the most common disease with 49.09% followed by Bullous pemphigoid 12.72%. Out of the 55 cases, 50 cases were clinically diagnosed as one of the varieties of vesiculobullous lesions, which were confirmed on histology.

Conclusion: Pemphigus vulgaris constituted the most common subtype of vesiculobullous disorder in this study followed by Bullous pemphigoid. Clinicopathological correlation is very important in the diagnosis of vesiculobullous disorders rather than considering either of them alone.

Keywords: Vesiculobullous disorders, Pemphigus vulgaris, Bullous pemphigoid.

Introduction

Skin is the largest organ of the body and variety of diseases commonly involve the skin, out of which vesiculobullous lesions form one of predominant group.

Vesicles (blister less than 0.5 cm in diameter) and bullae (blister greater than 0.5 cm in diameter) occur in several skin disease in all the layers of epidermis from stratum corneum to basal layers and subepidermally. Each entity in this group has different clinical features and lesions share number of histologic features but only some extent have common pathogenic mechanism e.g.; Bullous pemphigoid and Pemphigus vulgaris are autoimmune in nature, whereas Epidermolysis bullosa is inherited disease caused by non-immunologic mechanism.¹,²,³
Vesiculobullous diseases are one of the most significant primary morphological patterns of skin reaction to various external and internal pathologic stimuli. Wide range of pathologic processes can lead to development of vesiculobullous eruptions over body. They may occur in many dermatosis, which include various inflammatory, infective, autoimmune, drug induced as well as genetic. Vesiculobullous diseases are a group of disorders in which primary lesion is a vesicle or a bulla, on the skin or mucous membrane or both. Histopathological study is helpful in knowing the level of cleavage in the bulla, mechanism of blister formation, character of the inflammatory infiltrate, including its presence or absence, its pattern, the specific cell types involved and the condition of the epidermis and dermis. Over the last two decades, great advances have been made in understanding the clinical behaviour and molecular nature of autoimmune diseases. Histological study is one of the most valuable means of diagnosis in Dermatology. The greatest diagnostic accuracy is obtained by correlating the clinical and histological findings.

Materials and Methods
A Laboratory based cross-sectional observational study was conducted over a period of 10 months from June 2018 to March 2019 in the Department of Pathology, S.M.S Medical College, Jaipur. After taking approval from institutional ethics committee, 55 clinically diagnosed cases of vesiculobullous lesions of the skin were included. Vesiculobullous lesions due to radiation, burn and drug induced skin eruption were excluded from the study. Clinical data like age, sex, duration of the lesion; site of the lesion, significant family and personal history, history of associated diseases and other relevant history like any drug intake history were collected in a predesigned and semi-structured performa. The punch biopsy of intact blister is obtained and fixed in 10% formalin solution. Gross examination of the skin biopsy included measurement of size, shape of the biopsy then submitted for routine processing and embedded in paraffin wax, the sections were cut at 3-4 microns thickness and stained with Hematoxylin and Eosin. Histopathological examination of sections included plane of separation, whether subcorneal, intraepidermal, suprabasilar or subepidermal was noted. Mechanism of blister formation whether by acantholysis, spongiosis, basement membrane zone destruction, cytolysis, presence or absence of inflammation and type of inflammatory cell infiltrate were noted.

Results
A total of 920 (8.69%) skin punch biopsies were received and vesiculobullous lesion biopsies were 55 in number which constituted 0.51% of overall biopsy specimen and 5.97% of total skin punch biopsies (Figure-1).

In the present study, Twenty-seven cases of Pemphigus vulgaris (49.09%) followed by 7 cases of Bullous pemphigoid (12.72%) formed the majority of the cases. Four cases each of Pemphigus foliaceus and Dermatitis herpetiformis, three cases each of Subcorneal pustular dermatosis and Darier’s disease, two cases of Linear IgA dermatosis were obtained. Less common lesions included Erythema multiforme and Hailey-Hailey disease one case each (1.81%) as indicated in table-1.

Most of the vesiculobullous lesions were in the age group of 10-60 years. The youngest patient was 11 years old and the oldest was 80 years old. Pemphigus vulgaris was detected most commonly between 41 to 60 years. Bullous pemphigoid was detected most commonly between 51-70 years, slightly higher than in Pemphigus vulgaris.

In the present study, vesiculobullous lesion had a female preponderance with a male:female ratio of 0.89:1. Female predominance was noted in Pemphigus vulgaris, Bullous pemphigoid, Pemphigus foliaceus and Darier’s disease. Male predominance was noted in Subcorneal pustular dermatosis, Erythema multiforme and Hailey-Hailey disease.
Vesicles, bullae, erosions and crusted lesions were the predominant lesions in all the patients. Mucous membrane involvement was predominantly seen in Pemphigus vulgaris. In the present study of 55 cases clinical diagnosis correlated with the histological diagnosis in 50 cases and in 2 cases, the diagnosis was done only on histology, where in 3 cases it was difficult to offer a conclusive histological diagnosis (Figure-2). The cases which were diagnosed mainly on histology included Bullous pemphigoid and Pemphigus foliaceus which were clinically diagnosed as Pemphigus foliaceus and Linear IgA dermatosis respectively.

The histopathological features of Pemphigus vulgaris (Figure-3) and Bullous pemphigoid (Figure-4) are indicated in table-2 and table-3 respectively. In Pemphigus foliaceus (Figure-5), subcorneal bulla was seen in all 4 cases. Blister cavity filled with acantholytic cells, neutrophils and RBC’s. Dermis showed perivascular infiltration by neutrophils and lymphocytes. In Dermatitis herpetiformis (Figure-6), all 4 cases showed the presence of subepidermal bulla formation filled with neutrophils and few eosinophils in a pink eosinophilic background. Dermis was infiltrated by mixed inflammatory infiltrate. In Subcorneal pustular dermatosis (Figure-7), all 3 cases showed an intraepidermal subcorneal bulla containing only neutrophils in one case, neutrophils, few lymphocytes and fibrin in the other, neutrophils, eosinophils and few acantholytic cells in third case. Dermis showed mixed perivascular inflammatory infiltrate in 2 cases and one case showed perivascular infiltration with neutrophils and eosinophils. In Darier’s disease (Figure-8), all cases showed hyperkeratosis, acanthosis. Dyskeratotic changes with corps ronds and grains, and suprabasal bullae along with acantholytic cells seen. Dermis showed perivascular lymphocytic inflammatory infiltrate. In Linear IgA dermatosis (Figure-9), one case of Linear IgA dermatosis, showed a subepidermal vesicle filled with fluid, neutrophils and lymphocytes. Epidermis showed hyperkeratosis and acanthosis. Dermis showed perivascular infiltration by neutrophils and lymphocytes. The other case showed subepidermal vesicle containing neutrophils. Focal thinning of epidermis present. Dermal papillae showed neutrophilic papillary microabscesses. In Erythema multiforme (Figure-10), case revealed epidermis showing necrotic keratinocytes, subepidermal bulla with neutrophils, eosinophilic debris and necrosis. Dermis showed perivascular lymphohistiocytic inflammation with necrosis of vessel wall and accumulation of fibrinoid material. Subcutaneous tissue showed lymphohistiocytic inflammation. In Hailey-Hailey disease (Figure-11), there was a suprabasal bulla containing partially acantholytic cells giving the appearance of “dilapidated brick wall” appearance. At places downward elongation of rete ridges was seen and the superficial dermis contains chronic inflammatory cell infiltrates.
Table 1: Distribution of the cases

| S. No. | Lesions                              | No. of cases | Percentage |
|-------|--------------------------------------|--------------|------------|
| 1.    | Pemphigus vulgaris                    | 27           | 49.09      |
| 2.    | Bullous pemphigoid                    | 7            | 12.72      |
| 3.    | Pemphigus foliaceus                   | 4            | 7.27       |
| 4.    | Dermatitis herpetiformis              | 4            | 7.27       |
| 5.    | Subcorneal pustular dermatosis        | 3            | 5.45       |
| 6.    | Darier’s disease                      | 3            | 5.45       |
| 7.    | Linear IgA dermatosis                 | 2            | 3.63       |
| 8.    | Erythema multiforme                   | 1            | 1.81       |
| 9.    | Hailey-Hailey disease                 | 1            | 1.81       |
| 10.   | Inconclusive                          | 3            | 5.45       |
| Total |                                      | 55           | 100        |

Table 2: Histopathological features in Pemphigus vulgaris

| S. No. | Histopathological features          | No. of cases | Percentage |
|-------|-------------------------------------|--------------|------------|
| 1.    | Suprabasal bulla                    | 27           | 100        |
| 2.    | Acantholysis                         | 26           | 96.29      |
| 3.    | Row of tombstone appearance          | 23           | 85.18      |
| 4.    | Inflammatory infiltrate in the bullous cavity | 12   | 44.44      |

Table 3: Histopathological features in Bullous pemphigoid

| S. No. | Histopathological features          | No. of cases | Percentage |
|-------|-------------------------------------|--------------|------------|
| 1.    | Subepidermal bullae                 | 7            | 100        |
| 2.    | Bullae with inflammatory cell rich infiltration | 4   | 57.14      |
| 3.    | Bullae with inflammatory cell poor infiltration | 3   | 42.85      |
| 4.    | Perivascular infiltration           | 7            | 100        |

Figure 3: H & E stained section (100X) showing suprabasal bulla in Pemphigus vulgaris.

Figure 4: H & E stained section (100X) of Bullous pemphigoid showing subepidermal blister formation and an inflammatory infiltrate composed predominantly of eosinophils in the bullous cavity.

Figure 5: H & E stained section (100X) of Pemphigus foliaceus showing subcorneal blister with acantholytic cells and neutrophils in the cavity.

Figure 6: H & E stained section (100X) of Dermatitis herpetiformis showing neutrophilic micro abscesses with subepidermal bulla.
Figure-7: H & E stained section (100X) of Subcorneal pustular dermatosis showing subcorneal vesicle filled with neutrophils.

Figure-8: H & E stained section (100X) of Darier’s disease showing hyperkeratosis, suprabasal intraepidermal vesicle and corps ronds.

Figure-9: H & E stained section (100X) of Linear IgA dermatosis showing subepidermal blister with inflammatory cells composed predominantly of neutrophils.

Figure-10: H & E stained section (100X) of Erythema multiforme showing subepidermal bullae filled with edema and inflammatory cells.

Figure-11: H & E stained section (100X) of Hailey-Hailey disease showing Dilapidated brick wall appearance with suprabasal clefting.

Discussion
The vesiculobullous skin lesions consist of a group of eruptions of different etiology and prognosis, where blister formation occurs in different levels and clinically present as blisters. Histopathological examination is an important tool in the diagnosis of blistering diseases.

Spectrum of vesiculobullous disorders
In the present study, Pemphigus vulgaris was the most common vesiculobullous disorder constituting 49.09% (27 out of 55 cases) followed by Bullous pemphigoid 12.72% (7 cases). Pemphigus foliaceus and Dermatitis herpetiformis being 7.27% each (4 cases), Subcorneal pustular dermatosis and Darier’s disease being 5.45% each (3 cases), Linear IgA dermatosis being 3.63% (2 cases), Erythema
multiforme and Hailey-Hailey disease being 1.81% each (1 case).

Pemphigus vulgaris being the most common is similar to Arundhati et al\textsuperscript{7}, Pavithra et al\textsuperscript{8}, Patel et al\textsuperscript{9} and Punneshetty et al\textsuperscript{11} studies. Bullous pemphigoid being the second most common is similar to Arundhati et al\textsuperscript{7}, Pavithra et al\textsuperscript{8} and Punneshetty et al\textsuperscript{11} studies.

**Age and sex distribution**

In the present study, Male: Female ratio was 0.89:1. Predominance of females was noted in this study similar to Arundhati et al\textsuperscript{7}, Pavithra et al\textsuperscript{8}, Patel et al\textsuperscript{9}, Punneshetty et al\textsuperscript{11}, Kudligi et al\textsuperscript{13}, Mittal et al\textsuperscript{14} and Karattuthazhathu et al\textsuperscript{15}.

In the present study, the age range in Bullous pemphigoid (51-70 years) was slightly higher than in Pemphigus vulgaris (41-60 years). In another study by Punneshetty et al\textsuperscript{11} and Mittal et al\textsuperscript{14} also, it was found that the age range in Bullous pemphigoid was 60-80 years and 19-88 years respectively (a little higher than in Pemphigus vulgaris).

In the present study, the age range in Pemphigus foliaceus was 21-40 years similar to Punneshetty et al\textsuperscript{11} study.

**Clinical and histopathological correlation**

In the present study, Out of the 55 cases 50 cases (90.90%) were clinically diagnosed as one of the varieties of vesiculobullous lesions, which were confirmed on histology. 2 cases (3.63%) which were diagnosed mainly on histology included Bullous pemphigoid and Pemphigus foliaceus. Histologically ambiguous cases included the 3 cases (5.45%) in which diagnosis was inconclusive whereas in Pavithra et al\textsuperscript{8} and Krishnamurthy et al\textsuperscript{10} study histological confirmed cases comprised of 77.14% and 64.8% respectively, cases diagnosed only on histology comprised of 11.42% and 25.6% respectively and histologically ambiguous cases comprised of 11.42% and 9.4% respectively.

**Pemphigus vulgaris**

In the present study, Pemphigus vulgaris constituted 49.09% of the total number of cases of vesiculobullous disorders, which is higher that of Pavithra et al\textsuperscript{8}, Punneshetty et al\textsuperscript{11}, Kushtagi et al\textsuperscript{12} and Mittal et al\textsuperscript{14} study. There was predominance of females in this study (M: F ratio was 0.92:1), similar to Punneshetty et al\textsuperscript{11} and Mittal et al\textsuperscript{14} study.

Suprabasal bulla was seen in 100% cases which is higher that observed by Arya et al\textsuperscript{6}, Pavithra et al\textsuperscript{8} and Punneshetty et al\textsuperscript{11} study. Acantholysis was seen in 26 cases (96.29%) which is higher than that observed by Arya et al\textsuperscript{6}, Pavithra et al\textsuperscript{8} and Punneshetty et al\textsuperscript{11} study. Row of tombstone appearance seen in 23 cases (85.18%) which is higher that observed by Arya et al\textsuperscript{6}, Pavithra et al\textsuperscript{8} and Punneshetty et al\textsuperscript{11} study. Inflammatory infiltrate in the bullous cavity seen in 12 cases (44.44%) which is higher than that observed by Pavithra et al\textsuperscript{8} study and lower than that observed by Arya et al\textsuperscript{6} and Punneshetty et al\textsuperscript{11} study.

**Bullous pemphigoid**

In the present study, Bullous pemphigoid constituted 12.72% of the total number of cases of vesiculobullous disorders, which is lower that of Pavithra et al\textsuperscript{8}, Punneshetty et al\textsuperscript{11}, Kushtagi et al\textsuperscript{12} and Mittal et al\textsuperscript{14} study. There was predominance of females in this study (M: F ratio was 0.75:1), similar to Pavithra et al\textsuperscript{8}, Punneshetty et al\textsuperscript{11} and Mittal et al\textsuperscript{14} study.

All cases (100%) showed subepidermal bullae which is similar to Pavithra et al\textsuperscript{8} study and higher than that observed by Punneshetty et al\textsuperscript{11} study. Bullae with inflammatory cell rich infiltration was seen in 4 cases (57.14%) which is higher than that observed by Pavithra et al\textsuperscript{8} and lower than that observed by Punneshetty et al\textsuperscript{11} study. Bullae with inflammatory cell poor infiltration was seen in 3 cases (42.85%) which is lower than that observed by Pavithra et al\textsuperscript{8} study and higher than that observed by Punneshetty et al\textsuperscript{11} study. Perivascular infiltration was seen in all cases.
(100%) which is higher than that observed by Pavithra et al and Punneshetty et al study.

**Pemphigus foliaceus**

In the present study, Pemphigus foliaceus constituted 7.27% of the total number of cases of vesiculobullous disorders, which is lower that of Punneshetty et al, Kushtagi et al and Basu et al study. There was predominance of females in this study (M: F ratio was 0.33:1), similar to Punneshetty et al and Kushtagi et al study. Subcorneal bulla and bulla cavity filled with acantholytic cells was seen in 100% cases of Pemphigus foliaceus which is similar to study by Punneshetty et al and Kushtagi et al study whereas in Mittal et al study only 50% cases of Pemphigus foliaceus showed subcorneal plane of separation. Rest 50% cases of Pemphigus foliaceus showed intraepidermal plane of separation. In Mittal et al study bulla cavity filled with acantholytic cells was seen in 100% cases which is similar to present study.

**Dermatitis herpetiformis**

In the present study, Dermatitis herpetiformis constituted 7.27% of the total number of cases of vesiculobullous disorders, which is lower that of Krishnamurthy et al, Kushtagi et al and Mittal et al study. There was equal number of cases in both sex in this study (M: F ratio was 1:1) whereas Krishnamurthy et al, Kushtagi et al and Mittal et al study showed predominance of males. Kudligi et al study showed predominance of females.

All cases (100%) showed the presence of subepidermal blister formation filled with neutrophils which is similar to study by Krishnamurthy et al and Kudligi et al study whereas Mittal et al study showed similar finding in 88.9% cases.

**Subcorneal pustular dermatosis**

In the present study, Subcorneal pustular dermatosis constituted 5.45% of the total number of cases of vesiculobullous disorders, which is higher that of Krishnamurthy et al, Kushtagi et al, Kudligi et al and Mittal et al study. There was predominance of males in this study (M: F ratio is 3:0), similar to Mittal et al study.

All 3 cases showed an intraepidermal subcorneal bulla containing only neutrophils in one case, neutrophils, few lymphocytes and fibrin in the other, neutrophils, eosinophils and few acantholytic cells in third case. In Kudligi et al study 100% cases showed subcorneal bulla containing only neutrophils whereas in Mittal et al study 100% cases showed subcorneal bulla containing only acantholytic cells.

**Darier’s disease**

In the present study, Darier’s disease constituted 5.45% of the total number of cases of vesiculobullous disorders, which is higher that of Krishnamurthy et al and Kushtagi et al study. There was predominance of females in this study (M: F ratio is 1:2) whereas Krishnamurthy et al showed predominance of males.

All cases showed suprabasal bullae along with acantholytic cells and dyskeratotic changes with corps ronds and grains seen. Similar observations were made by Krishnamurthy et al and Kushtagi et al study.

**Linear Ig A Dermatosis**

In the present study, Linear IgA dermatosis constituted 3.63% of the total number of cases of vesiculobullous disorders, which is higher that of Krishnamurthy et al (2.7%) and Kudligi et al (2%) study. There was equal number of cases in both sex in this study (M: F ratio was 1:1) similar to Krishnamurthy et al study.

50% cases showed a subepidermal vesicle containing neutrophils and lymphocytes and 50% cases showed a subepidermal vesicle containing neutrophils only whereas in Krishnamurthy et al study 100% cases showed a subepidermal vesicle containing mixed inflammatory infiltrate.
Erythema multiforme
In the present study, Erythema multiforme constituted 1.81% of the total number of cases of vesiculobullous disorders, which is lower than that of Krishnamurthy et al.\textsuperscript{10} (8.1%) and Kushtagi et al.\textsuperscript{12} (7.5%) study. There was 40 year old male patient in this study. Krishnamurthy et al.\textsuperscript{10} and Kushtagi et al.\textsuperscript{12} study showed predominance of males. A case revealed epidermis showing necrotic keratinocytes, subepidermal bulla with inflammatory infiltrate. Krishnamurthy et al.\textsuperscript{10} and Kushtagi et al.\textsuperscript{12} study showed similar findings.

Hailey-Hailey disease
In the present study, Hailey-Hailey disease constituted 1.81% of the total number of cases of vesiculobullous disorders, which is higher than that of Krishnamurthy et al.\textsuperscript{10} (1.3%) and Mittal et al.\textsuperscript{14} (0.90%) study. There was 40 year old male patient in this study. In Krishnamurthy et al.\textsuperscript{10} study there was also male patient whereas Mittal et al.\textsuperscript{14} study there was female patient. There was a suprabasal bulla containing partially acantholytic cells giving the appearance of “dilapidated brick wall” appearance. Krishnamurthy et al.\textsuperscript{10} study showed similar findings where as in Mittal et al.\textsuperscript{14} study intraepidermal bulla was present.

Conclusion
Vesiculobullous diseases are a group of disorders in which primary lesion is a vesicle or a bulla, on the skin or mucous membrane or both. They form a bulla either in the epidermis (subcorneal or suprabasal) or beneath the epidermis. Pemphigus vulgaris constituted the most common subtype of vesiculobullous disorder in this study followed by Bullous pemphigoid. Histopathological examination is helpful for arriving at a definitive diagnosis in majority of vesiculobullous disorders. Clinicopathological correlation is very important in the diagnosis of vesiculobullous disorders rather than considering either of them alone. It becomes important to differentiate each of these entities for appropriate management, failure of which in some cases is associated with significant morbidity and mortality.

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