

Diagnosis and Clinical Features of Pemphigus Vulgaris

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KEYWORDS

• Pemphigus vulgaris • Desmoglein 3 • Diagnosis

CLINICAL PRESENTATION OF PEMPHIGUS VULGARIS

Autoimmune bullous diseases are associated with autoimmunity against structural components that maintain cell-cell and cell-matrix adhesion in the skin and mucous membranes.¹ They include those where the skin blisters at the basement membrane zone (bullous pemphigoid, herpes gestationis, mucous membrane pemphigoid, linear immunoglobulin (Ig)A dermatosis, epidermolysis bullosa acquisita, bullous lupus, and dermatitis herpetiformis) and those where the skin blisters within the epidermis (pemphigus vulgaris [PV], pemphigus foliaceus [PF], and other subtypes of pemphigus).

Because of the considerable overlap in the clinical presentation of these conditions, diagnosis of autoimmune bullous skin conditions can be challenging. Detection of tissue-bound and circulating serum autoantibodies and characterization of their molecular specificity is an important modality for diagnosis. In the past decade, there have been several advances in diagnostic modalities for autoimmune bullous skin conditions.

Pemphigus, a word derived from the Greek word “pemphix” meaning bubble or blister, is a life-threatening autoimmune blistering disease characterized by intraepithelial blister formation.^{2–4} Damage to intercellular adhesion structures, desmogleins, are the target of circulating autoantibodies resulting in the hallmark of this condition, acantholysis.^{5,6} Acantholysis may result in the development of the Tzanck phenomenon, which is the rounding of single epidermal cells caused by the loss of cell-cell attachment.

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EPIDEMIOLOGY

The incidence of pemphigus is approximately 1 in 100,000 people. Pemphigus vulgaris is the most common variant of pemphigus with an incidence of 0.1 to 0.5 per 100,000 population and higher among Jewish patients.⁷ In India, Malaysia, China, and the Middle East, pemphigus vulgaris accounts for 70% of all pemphigus cases and may be the most common autoimmune blistering disease.^{8–10}

Various environmental and pharmacologic etiological factors have been reported in pemphigus. These factors include medications, pesticides, malignancy, ultraviolet radiation, and stress.^{11–18}

Foods containing an allium, phenol, thiol, or urushiol group have also been reported to trigger pemphigus.^{19,20}

Pathogenesis

PV is an autoimmune condition that is more likely to develop in patients with HLA types after certain triggers. PV is associated with several autoantibodies. The main autoantibodies target the desmosomal protein, desmoglein 3 (dsg3), a 130-kDa glycoprotein cadherin.⁵ Later, there is secondary development of antibodies to another desmosomal cadherin, desmogleins 1, which is a 160-kDa glycoprotein dsg1 antigen, when the skin is involved.²¹ Further potential target antigens in PV are acetylcholine receptors on keratinocytes.^{22–24} Pincelli's group has shown that apoptosis pathways are triggered early in PV and are followed by desmosomal endocytosis.^{25,26} In addition, David Rubenstein's group has shown that the signaling molecule p38 is rapidly phosphorylated when PV IgG is added to human keratinocytes and that inhibiting p38 blocks dsg3 endocytosis, keratin retraction, and actin reorganization. P38 is phosphorylated in the skin of neonatal mice tested with either PV or PF IgG. P38 inhibitors block blistering in both the PV and PF passive transfer mouse models. Similarly, p38 is phosphorylated in the skin of human patients with PV and PF.^{27,28}

PV is strongly associated with the HLA serotypes HLA-DR4 and HLA-DR6.²⁹ 48% of healthy relatives of patients with PV had low levels of autoantibody and the inheritance of autoantibody positivity was linked to the DR4 and DR6 haplotype. Although population studies report differing prevalences of alleles in various ethnic groups, greater than 95% of patients with PV possess one or both of these haplotypes.³⁰ In 2006, Lee and colleagues³⁰ concluded that in the non-Jewish population, 8 alleles were positively associated and 1 allele was negatively associated with PV. The 2 candidate alleles most likely to contribute to disease susceptibility in the Non-Jewish population included DRB1*0402 and DQB1*0503. DRB1*0402 was determined to be the sole allele likely to confer susceptibility to PV in Ashkenazi Jewish patients.

CLINICAL FEATURES

Clinical Presentation

The variants of pemphigus are determined according to the level of intraepidermal split formation. There are 5 main variants of pemphigus: pemphigus vulgaris, pemphigus foliaceus, pemphigus erythematosis, drug-induced pemphigus, and paraneoplastic pemphigus. This review focuses only on PV.

Patients suffering from PV present with oral erosions and then subsequently develop cutaneous involvement.^{31,32} Mucosal erosions usually precede the cutaneous manifestations of the disease and often result in a protracted course of misdiagnosis with conditions, such as aphthous ulceration. In some cases, oral ulceration may be the only manifestation of the disease. The mucosal surfaces that may be involved include the oropharynx, esophagus, conjunctiva, nasal, larynx, urethra, vulva, and cervix.^{33–38}

Cutaneous involvement may be localized or generalized. Skin lesions have a predilection for the trunk, groins, axillae, scalp, face, and pressure points (**Figs. 1–4**).³⁹ Flaccid blisters develop on these sites and may coalesce; these blisters eventually rupture and result in painful erosions. PV usually presents in adults and can affect anywhere in the body but predominantly affects the buccal and labial mucosa (**Fig. 5**). This condition is characterized by Nikolsky's sign; the direct Nikolsky is when the application of slight pressure on a blister results in extension of the blistering to adjacent skin and the indirect Nikolsky is when rubbing on clinically normal skin causes shearing. These signs are not always 100% reliable for the diagnosis of PV, but they are suggestive if present.⁴⁰

Other more rare clinical manifestations include nail dystrophy, paronychia, subungual hematomas, and neonatal pemphigus vulgaris.^{41,42}

DIFFERENTIAL DIAGNOSIS

Patients are often treated for multiple other blistering conditions before the diagnosis of pemphigus vulgaris is made with diagnostic investigations, in particular a skin or mucosal biopsy for DIF.

The differentials for mucosal lesions include stomatitis secondary to herpes simplex virus; aphthous ulcers; lichen planus; paraneoplastic pemphigus; or autoimmune disease, such as lupus erythematosus or dermatitis herpetiformis. Differentials for cutaneous involvement include other autoimmune blistering skin conditions, such as pemphigus foliaceus, pemphigus vegetans, IgA pemphigus, paraneoplastic pemphigus,



Fig. 1. Patient with pemphigus vulgaris presenting with crusted erosions on his scalp and face.

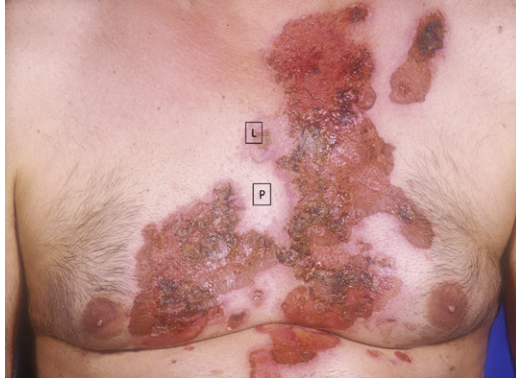


Fig. 2. Crusted, superficial erosions on the chest and abdomen of the same patient with pemphigus vulgaris. Boxed *L* indicates an appropriate site for a lesional biopsy for routine histology and boxed *P* indicates a site that is good for the perilesional biopsy for direct immunofluorescence.

bullous pemphigoid, linear IgA disease, erythema multiforme, Grover disease, and Hailey-Hailey disease.

There are a variety of blistering conditions that must be taken into consideration when suspecting that patients have pemphigus vulgaris. The etiology of blistering diseases may be broadly divided into autoimmune, infective, or inflammatory.



Fig. 3. Vegetative erosions on the nose of a patient with pemphigus vulgaris.



Fig. 4. Some tense and flaccid, well-circumscribed bullae and superficial erosions on the abdomen of a young woman with pemphigus vulgaris.

DIAGNOSIS

The diagnosis of PV can be made using 4 major criteria:

1. Clinical findings
2. Light microscopic findings
3. Direct immunofluorescence findings
4. Indirect immunofluorescence findings.^{4,43}

The clinical findings assisting in the diagnosis of PV, a potentially life-threatening autoimmune vesiculobullous disorder, include nonscarring, fragile vesicles, and bullae involving the mucosae with varying cutaneous involvement.

The biopsies should be carefully performed on lesional and perilesional skin. The most recent, untreated lesion is the most preferable site for biopsy. At the edge of a blister (lesional) is the best site for hematoxylin and eosin (H+E) analysis to discern the presence and level of blistering or acantholysis (see **Fig. 2**). The type, density, and level of inflammatory infiltrate in conjunction with the direct immunofluorescence findings assist in making the diagnosis of pemphigus vulgaris. Lesional biopsies for H+E can be transported in formalin.

Perilesional biopsies taken within 2 cm of active blistering (see **Fig. 2**) should be taken for DIF analysis and transported in normal saline if they can get to the laboratory straight away for the best results⁴⁴ or Michel's medium otherwise. These characteristically show netlike, intercellular staining with IgG, C3, IgM, or IgA (discussed in the next section).



Fig. 5. Superficial, diffuse gingival erosions in a patient with pemphigus vulgaris. (Courtesy of Dr Schifter, Skin and Cancer Foundation, Westmead, Sydney.)

Patients with isolated mucosal disease should have lesional and perilesional mucosal biopsies performed for H+E and DIF to confirm the diagnosis of pemphigus vulgaris in conjunction with biopsies of cutaneous lesions, if clinically warranted.

HISTOPATHOLOGY

PV is usually characterized by suprabasilar loss of adhesion leaving a single layer of basal keratinocytes attached to the dermoepidermal basement membrane (tomb-stone pattern). This characteristic is distinguished from PF, which is associated with a more superficial split formation in the subcorneal layer. Histologic findings in PV lesions on hematoxylin and eosin demonstrate suprabasilar acantholysis and infiltration with predominantly neutrophils and eosinophils (**Fig. 6**). The early findings of pemphigus may be subtle and include suprabasilar acantholysis associated with a mild, superficial mixed inflammatory infiltrate, including some eosinophils. These changes can also be present in more developed lesions (see **Fig. 6**).

On direct immunofluorescence, IgG or C3 binding to the intercellular cement substance in the mid-lower or entire epidermis of perilesional skin or mucosa is characteristic (**Fig. 7**).^{45–48}

Desmoglein 3⁵ and desmoglein 1²¹ are the targets for autoantibodies in PV and PF, respectively. Tissue-bound IgG, C3, IgM, or IgA in a characteristic netlike intercellular distribution pattern within the epidermis is demonstrated on direct immunofluorescence microscopy (see **Fig. 7**). Anti-dsg1 and anti-dsg3 antibodies predominantly belong to the IgG4 subclass.^{49–52} Anti-IgA and IgE subclass antibodies have also been detected. Patients with predominantly mucosal involvement have antibodies only against dsg3; however, a significant proportion of mucosal-dominant disease will also have dsg1 autoantibodies.^{53,54} The phenotypic presentation of pemphigus vulgaris with respect to mucosal and generalized cutaneous involvement has been attributed to the presence of desmoglein 3 and desmoglein 1 autoantibodies, respectively.^{55–62}

Indirect immunofluorescence microscopy reveals the presence of serum autoantibodies against desmosomal antigens. Pemphigus sera show a characteristic netlike intercellular staining of IgG with human skin as a substrate. Other substrates, such as monkey esophagus, guinea pig esophagus or rat bladder epithelium, may be used in the diagnosis of paraneoplastic pemphigus.⁶³

Enzyme-linked immunosorbent assay (ELISA) has provided higher sensitivity and specificity in making the diagnosis of pemphigus subtypes.⁶⁴ ELISA results can be used to determine pemphigus vulgaris disease activity and be used for longitudinal

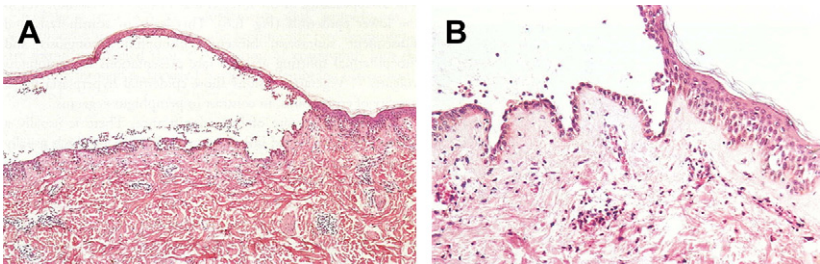


Fig. 6. Hematoxylin and eosin findings of an acute lesion of pemphigus vulgaris showing suprabasilar acantholysis (A, B). (From Weedon D, Strutton G, Rubin AI. The vesiculobullous reaction pattern. In: Weedon D, editor. *Weedon's Skin Pathology*. 3rd edition. Philadelphia: Churchill-Livingston; 2010; with permission.)

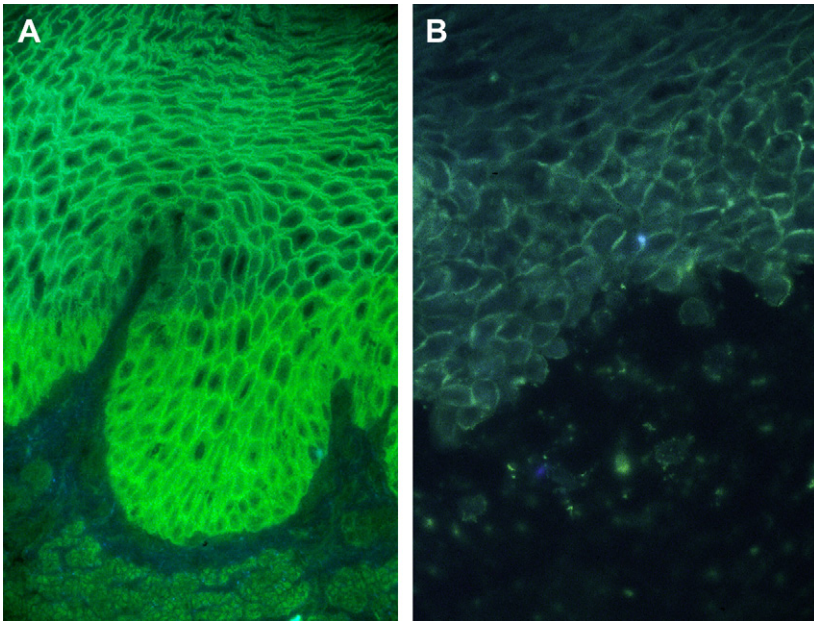


Fig. 7. Direct immunofluorescence results in a patient with pemphigus vulgaris showing intercellular staining with IgG (A) and C3 (B). (Courtesy of Professor Kossard, Skin and Cancer Foundation, Darlinghurst, Sydney.)

monitoring of treatment response. Antibodies can be detected in patients without clinical signs of pemphigus and can be found in patients with staphylococcal scalded skin,⁶⁵ penicillin adverse drug reactions,⁶⁶ toxic epidermolysis necrosis,⁶⁷ and burns.⁶⁸ In addition, patients with blood group O who have antibodies to blood groups A and B may give low false positives on indirect immunofluorescence testing,⁶⁹ which is avoided by preabsorbing their sera with these blood group antigens.

In active PV, immunoblot analysis with recombinant dsg3 demonstrated that anti-dsg3 of the IgG4, IgA, and IgE subtypes predominate; however, chronic remittent PV is characterized by IgG1 and IgG4 autoantibodies.^{50,53}

DISEASE ACTIVITY CATEGORIZATION

The Consensus Statement on Disease Endpoints and Therapeutic Response for Pemphigus⁷⁰ divides pemphigus disease activity into the following categories:

- Early endpoints
 - Baseline
 - Control of disease activity
 - End of consolidation phase
- Late endpoints
 - Complete remission off therapy
 - Complete remission on therapy
 - Minimal therapy
 - Minimal adjuvant therapy
 - Partial remission off therapy
 - Partial remission on minimal therapy

- Relapse/flare
- Treatment failure.

Early endpoints provide a useful clinical indicator for clinicians regarding the commencement of differing treatment regimes. The *baseline* is classified as the day that the treating practitioner initiates treatment. *Control of disease activity* is defined as the time at which there is cessation of new lesions in conjunction with the healing of preexisting lesions. In the majority of cases the expected time period in this stage is weeks. The *end of the consolidation phase* is the time period in which no new lesions have developed over a minimum period of 2 weeks. This phase is also characterized by the healing of most lesions, and most medical practitioners would consider the weaning of steroids.

Late endpoints of disease activity may be reached with or without therapy. *Complete remission off therapy* is characterized by the absence of new lesions over a 2-month period after cessation of therapy. *Minimal therapy* constitutes treatment with less than or equal to 10 mg/day of prednisone or the equivalent or the use of minimal adjuvant therapy for a duration of at least 2 months. *Minimal adjuvant therapy* comprises of half the dose required to be defined as treatment failure. *Partial remission off therapy* is classified as development of lesions after cessation of treatment that heal within 1 week without treatment. Patients must be off systemic therapy for 2 months to be classified in this category. Patients may suffer a partial remission on minimal therapy when they develop new lesions that heal within 1 week while receiving minimal therapy. Topical steroids also constitute minimal therapy.

A *relapse/flare* is defined by the development of 3 or more new lesions that persist without healing for greater than 1 week or by the extension of preexisting established lesions. Treatment failure results when there is no change in disease activity despite treatment on therapeutic doses of systemic steroids and other agents whose doses and durations were agreed by international consensus.⁷⁰

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