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Chapter

Gingival Nikolsky’s Sign: A Valuable Tool in Identifying Oral Manifestations of Mucous Membrane Pemphigoid and Pemphigus Vulgaris

Hiroyasu Endo, Terry D. Rees, Hideo Niwa, Kayo Kuyama, Maya Oshima, Tae Serizawa, Shigeo Tanaka, Morio Iijima, Masamichi Komiya and Takanori Ito

Abstract

Autoimmune bullous diseases are a group of rare, chronic blistering diseases that affect the skin and mucous membranes. Mucous membrane pemphigoid (MMP) is the most frequently occurring autoimmune bullous disease in the oral cavity, followed by pemphigus vulgaris (PV). Early diagnosis of MMP or PV is critical for proper management and prevention of potential serious complications. This study was based on a retrospective review of 39 cases that were classified as MMP (25 patients) or PV (14 patients). Nikolsky’s sign characterized by epithelial detachment as a result of slight pressure or rubbing the oral mucosa is a simple test that can confirm the existence of gingival desquamation. A positive reaction was confirmed in 38 patients (97.4%) at their first visit. This result indicates that patients showing positive Nikolsky’s sign should include MMP or PV in the differential diagnosis and, in that case, histopathological examination and direct immunofluorescence testing are critical to establish the final diagnosis. For the early diagnosis of autoimmune bullous disorders, oral healthcare providers should consider the use of the test for Nikolsky’s sign that may ultimately lead to the early diagnosis of MMP and PV or other diseases or disorders.

Keywords: gingival diseases, pemphigoid benign mucous membrane, pemphigus, oral medicine, autoimmune diseases

1. Introduction

Autoimmune bullous diseases are a group of rare, chronic blistering diseases that affect the skin and mucous membranes. Mucous membrane pemphigoid (MMP) is the most frequently occurring autoimmune bullous disease in the oral cavity, followed by pemphigus vulgaris (PV) [1–3]. Other diseases include bullous pemphigoid, lichen planus pemphigoides, paraneoplastic pemphigus, and chronic
ulcerative stomatitis [4, 5]. The primary lesions of MMP or PV often develop in
the oral cavity, and patients may complain of oral symptoms and visit their dental
clinic first before seeking medical consultation [6, 7]. Therefore, oral healthcare
providers need to have some current knowledge about autoimmune bullous diseases
and have a great responsibility to achieve early detection, diagnosis, and treatment
of the diseases or to refer the patients to other medical or dental specialists as soon
as possible.

The gingiva is one of the target tissues of autoimmune bullous diseases. Patients
often complain of uncomfortable or painful gingiva or other oral pathologic tissues
and usually seek care from their general dentist or periodontist. Desquamative
gingivitis (DG) characterized by gingival desquamation, erosion, ulceration,
erythematous gingiva, and hemorrhage is a clinical term used to describe some
pathologic changes that are common to a variety of gingival diseases or disorders
[1–3, 8, 9]. Table 1 summarizes the clinical appearance of DG. It is important to
remember that DG is a general descriptive term rather than a diagnosis (Table 2).
Therefore, diagnosis of the specific disease or disorder causing DG is important to
provide proper treatment. Biopsy evaluation is often required for definitive diag-
nosis. Especially, histopathological examination and direct immunofluorescence
(DIF) testing are critical to establish the final diagnosis for MMP or PV [3, 10–12].

MMP is a group of rare, autoimmune bullous disease that can primarily affect
mucous membranes. Various components in the basement membrane zone (BMZ)
have been recognized as the target antigens of MMP [13–16]. The major autoanti-
gens in MMP are BP180 C-terminal domain and laminin-332 [15, 16]. In more than
90% of MMP patients, lesions are found in the oral mucosa [14, 17, 18]. DG lesions
are usually present. Most MMP patients are in their fifth decade of life, and majority
of them are females [13, 14, 17, 18]. Scar formation and an associated loss of func-
tion are the most serious complications of some forms of MMP. Sight-threatening
ocular scarring and life-threatening airway obstruction have been reported
although the scarring is rarely seen in the oral mucosa [17, 19–22]. Early diagnosis of
MMP is critical, and immunosuppressive therapy may prevent scarring in mucous
membranes. Histopathologically, subepithelial blister formation is characteristic,
but it is not always seen in biopsy specimens [3, 10, 12, 13]. However, this is a
nondiagnostic finding since it is also found in other vesiculobullous diseases. In DIF
testing, a linear pattern of C3, IgG, or other immunoglobulin, fibrin, or fibrinogen
is present along the BMZ [3, 10, 12, 13].

PV is a rare, autoimmune bullous disease that is characterized by intraepithelial
acantholysis. PV can develop at any age but most commonly occurs in middle-aged and
elderly patients [2, 23, 24]. PV affects both males and females equally [2, 23, 24]. PV
is a rare, but serious and potentially life-threatening condition if left untreated [25].
Oral lesions are the first site of PV involvement in most patients. The oral lesions of PV
are usually multiple, typically involving the buccal mucosa and soft palate [24, 26]. On

| Painful gingiva |
| Burning sensation |
| Gingival bleeding |
| Gingival erythema not resulting from dental plaque accumulation |
| Desquamation, erosion, and ulceration of the gingiva |
| Blister formation on the gingiva |
| Other intraoral and/or extraoral lesions |
| Possible positive Nikolsky’s sign of the gingiva |

Modified from Endo et al. [6, 17], Rees & Burkhart [3].

Table 1.
Clinical appearance of desquamative gingivitis.
occasion, the gingiva is the only site involved, and DG is a relatively common clinical manifestation of the disease [27, 28]. It has been determined that the principal autoantigens in pemphigus patients are desmogleins (Dsgs), which are the components of desmosomes in the epidermis and mucous membranes [29, 30]. The main target antigen of PV is Dsg 3 [29, 30]. Most patients with PV lesions limited to the oral mucosa have only anti-Dsg 3 antibody in the serum, whereas patients involving both the oral mucosa and skin may have both anti-Dsg 3 and anti-Dsg 1 antibodies [28, 31]. In a histopathologic examination, PV is characterized by acantholysis and suprabasilar blister formation in the epithelium [3, 10, 12]. In the DIF testing of PV patients, deposition of IgG and C3 is often found between the epithelial cells and is characterized by a “fishnet” or “chicken-wire” pattern [3, 10, 12].

In addition to the classic DG lesions, clinical diagnosis for MMP or PV may be supported by the presence of extragingival lesions including the buccal mucosa, the soft palate or tongue, or the presence of extraoral lesions including the eyes, upper respiratory tract, genitals, anus, or skin [3, 17, 31]. However, the patients often had lesions confined only to the gingiva [27]. In such a case, early diagnosis of autoimmune bullous diseases in the oral cavity may become more difficult. Diagnosis delays of more than 6 months were experienced by 30.8% of this group of PV patients and 54.2% of the MMP patients [27]. 16.7% of patients with MMP were delayed for more than 12 months from onset to diagnosis [27].

Epithelial desquamation of the gingiva is a prominent clinical feature that supports early clinical diagnosis of autoimmune bullous diseases in the oral cavity [6]. Some of the patients with MMP or PV were aware of painful epithelial desquamation of the gingiva during meals or oral hygiene practices, and the patients complained it to the dental practitioners. However, due to the limited understanding of oral healthcare providers for autoimmune bullous diseases, MMP or PV was not included in the differential diagnosis [6]. For that reason, many patients are not diagnosed until lesions have become severe. Early diagnosis of MMP or PV is critical for proper management and prevention of potential serious complications. Nikolsky’s sign is a phenomenon characterized by epithelial desquamation as a result of slight pressure or rubbing the skin or oral mucosa [32]. This sign is a simple test that can confirm the existence of gingival desquamation. In the dental clinic, the presence of Nikolsky’s sign can be evaluated by the application of a firm sliding or rubbing force to the mucosal surface using a dental instrument [3, 32].

In an attempt to facilitate the recognition of the early symptoms of autoimmune bullous diseases, the purpose of this study was to examine the frequency of positive Nikolsky’s sign at the first visit in patients with MMP or PV. Results of this study may expedite the diagnosis of autoimmune bullous diseases developing in the oral cavity.

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Table 2.
Diseases or disorders that are associated with desquamative gingivitis.

<table>
<thead>
<tr>
<th>Most frequent diseases or disorders</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Oral lichen planus</td>
<td></td>
</tr>
<tr>
<td>Mucous membrane pemphigoid</td>
<td></td>
</tr>
<tr>
<td>Pemphigus vulgaris</td>
<td></td>
</tr>
<tr>
<td>Hypersensitivity reactions to dental hygiene products, food flavorings, or preservatives</td>
<td></td>
</tr>
</tbody>
</table>

* A variety of other potential causes such as lupus erythematosus, mixed connective tissue disease, graft versus host disease, erythema multiforme, epidermolysis bullosa, epidermolysis bullosa acquisita, Kindler syndrome, chronic ulcerative stomatitis, lichen planus pemphigoides, plasmacytosis, plasma cell gingivitis, orofacial granulomatosis, foreign body granulomas, candidal infection and linear IgA disease, factitious injury of the gingiva, Crohn’s disease, panniculitis, sarcoidosis, and adverse drug reactions may possess some but usually not all of the clinical features of desquamative gingivitis. Modified from Endo et al. [2, 6], Rees & Burkhart [3].

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The table lists the most frequent diseases or disorders associated with desquamative gingivitis.
2. Materials and methods

The present study was based on a retrospective review of 39 cases that were classified as MMP (25 patients) or PV (14 patients) at Nihon University, School of Dentistry at Matsudo, from 2001 to 2018. The protocol of this study was approved by an institutional review board (Ethics Committee Approval No. EC14-011-1). The summary of the 39 patients are shown in Table 3. Some of the 39 patients presented in this study have been previously reported [6, 10, 11, 14, 27, 28, 31]. All 39 patients described gingival lesions consistent with DG (Figures 1 and 2). The oral lesions were confined to the gingiva in 27 patients (69.2%), although other 12 patients (30.8%) also had extragingival involvements (the buccal mucosa, soft palate, or tongue). Eleven of the 39 patients (28.2%) confirmed the existence of extraoral involvements (nose, pharynx, larynx, ocular mucosa, or skin). Gingival biopsies were performed in all 39 patients. Patients were diagnosed with MMP or PV through clinical examination supported by histopathologic diagnosis and DIF testing for each patient (Figures 3 and 4). The current study examined the clinical records of each

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>MMP (n = 25)</th>
<th>PV (n = 14)</th>
<th>Total (n = 39)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age at diagnosis</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean (years)</td>
<td>65.8</td>
<td>46.9</td>
<td>59.0</td>
</tr>
<tr>
<td>Range (years)</td>
<td>36–80</td>
<td>24–73</td>
<td>24–80</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>9</td>
<td>1</td>
<td>10 (25.6%)</td>
</tr>
<tr>
<td>Female</td>
<td>16</td>
<td>13</td>
<td>29 (74.4%)</td>
</tr>
<tr>
<td><strong>Clinical findings</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Desquamative gingivitis</td>
<td>25</td>
<td>14</td>
<td>39 (100%)</td>
</tr>
<tr>
<td>Intraoral site involvement</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Restricted to the gingiva</td>
<td>18</td>
<td>9</td>
<td>27 (69.2%)</td>
</tr>
<tr>
<td>Gingiva + extragingiva</td>
<td>7</td>
<td>5</td>
<td>12 (30.8%)</td>
</tr>
<tr>
<td>Extraoral site involvement*</td>
<td>8</td>
<td>3</td>
<td>11 (28.2%)</td>
</tr>
<tr>
<td><strong>Biopsy findings</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Histopathological examination</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Subepithelial blisters</td>
<td>21</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Acantholysis and suprabasilar blisters</td>
<td>—</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>Nonspecific</td>
<td>3</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Nondiagnostic</td>
<td>1</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>DIF examination</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMZ deposition**</td>
<td>25</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>ICS deposition**</td>
<td>—</td>
<td>14</td>
<td></td>
</tr>
</tbody>
</table>

MMP = mucous membrane pemphigoid; PV = pemphigus vulgaris; DIF = direct immunofluorescence; BMZ = basement membrane zone; ISC = intercellular space.

*After a diagnosis of MMP or PV, patients were advised to confirm the presence or absence of extraoral lesions by a dermatologist, an otorhinolaryngologist, and an ophthalmologist.

**Deposition of varying combinations of IgG, IgA, fibrinogen, and complement C3.

Table 3. Characteristics of 39 patients with autoimmune bullous diseases.
Gingival Nikolsky’s Sign: A Valuable Tool in Identifying Oral Manifestations of Mucous…
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Figure 1.
Desquamative gingivitis in mucous membrane pemphigoid. The intensity of the gingival erythema or erosion is variable, and the involvement may be diffuse or patchy distribution.

Figure 2.
Desquamative gingivitis in pemphigus vulgaris. The attached gingiva presents as friable nature of the tissue. Bullae develop quickly and then rupture, leaving eroded painful surfaces with ragged borders.

Figure 3.
Biopsy confirmation of mucous membrane pemphigoid. A subepithelial blister formation was found in hematoxylin-eosin-stained section. Direct immunofluorescence showed a linear deposition of IgG at the basement membrane zone.

Figure 4.
Biopsy confirmation of pemphigus vulgaris. Acantholysis and suprabasilar blister formation were recognized in hematoxylin-eosin-stained section. Direct immunofluorescence showed an intercellular deposition of IgG.
individual which provided information on each patient's gingival symptoms, gingival site involvement, and the presence of gingival epithelial desquamation based on a test for Nikolsky's sign. At the initial dental appointment, a test for Nikolsky's sign was performed in all 39 patients by a single examiner (HE) using the “marginal” method and the “direct” method (Figures 5 and 6) [32, 33]. Briefly, a positive gingival Nikolsky's sign described the extension of the erosion on the surrounding normal-appearing tissue by rubbing the edge of the affected area with a periodontal probe (the “marginal” method), or the ease of inducing erosion by rubbing apparently unaffected the gingiva distant from the lesions (the “direct” method). All 39 patients were evaluated using the “marginal” method. In addition, in some patients we also used the “direct” method. When a positive Nikolsky’s sign was identified, the presence of gingival bleeding was also evaluated (Figure 6).

3. Results

Table 4 summarizes the gingival symptoms in the 39 patients. Clinical symptoms described were soreness (31 patients, 79.5%), bleeding (21 patients, 53.8%), and swelling (18 patients, 46.2%). The results summarizing the gingival site involvement are shown in Table 5. The sites where DG lesions were most frequently found were the anterior areas (35 patients, 89.7%). In contrast, only four patients (10.3%) had DG lesions confined to the molar areas. Most of the gingival involvement was observed in the labial/buccal area (37 patients, 94.7%). In 22 patients (56.4%), gingival involvement was also observed in the palatal/lingual area. A positive Nikolsky's sign was demonstrated in 38 of the 39 patients (97.4%) at the first visit (Table 6). In 16 of the 38 patients (42.1%) in association with positive
Nikolsky’s sign, gingival bleeding was induced by gentle pressure (Table 7). All 16 patients were subsequently diagnosed as having MMP (Table 7).

4. Discussion

In this study, 39 DG patients with autoimmune bullous diseases diagnosed as MMP or PV participated. All the patients complained of gingival soreness, bleeding, and/or swelling (Table 4). A positive reaction showing Nikolsky’s sign was confirmed in 38 patients (97.4%) at their first visit (Table 6). This result indicates that it is important to evaluate the presence of gingival Nikolsky’s sign in DG patients. Patients showing positive Nikolsky’s sign should have MMP or PV included in the differential diagnosis when DG is identified. However, it should be noted that it is critical to conduct DIF biopsy testing in addition to histopathological examination. By doing this, the oral healthcare providers can contribute to the early diagnosis and treatment for MMP or PV lesions in the oral cavity. It is
important to note, however, that DG sites should not be selected for biopsy diagnosis since intact epithelium is necessary to confirm the diagnosis of these autoimmune disorders [3, 10]. This retrospective study was limited to those who exhibited autoimmune bullous diseases, and consequently we do not know the nature or number of diseases causing DG in individuals with other disorders. Another limitation of this study is that it does not include a control group. Future controlled clinical studies, including the test in non-autoimmune diseases groups, are needed to establish the validity of the results of the present study.

This study found that the gingiva is a preferable site for performing a test for Nikolsky’s sign. The site where DG lesions were frequently found was the anterior area of 35 patients (89.7%), while 37 patients (94.7%) were identified to have DG on either labial or buccal gingiva (Table 5). This indicates that direct access to the gingival surface to be examined is easy. The most suitable site for a test for Nikolsky’s sign would be the labial gingiva of the anterior area of the upper and lower jaws. In evaluating Nikolsky’s sign, the presence of bleeding from the gingiva roughly guess which epithelial cleavage level is occurring (subepithelial separation or intraepithelial separation). If the gingival bleeding occurred after performing a Nikolsky’s sign, this would imply a subepithelial separation such as MMP. In contrast, if the gingival bleeding was unlikely to occur, this would imply an intraepithelial separation such as PV. In this study, 16 patients (42.1%) had bleeding after application of a sliding or rubbing force on the gingiva, and all of 16 patients diagnosed as having MMP (Table 7). It should be noted, however, that the presence of gingival bleeding is also affected by the magnitude of the sliding or rubbing force to the gingival surface and the degree of gingival inflammation caused by concomitant dental plaque-induced gingivitis.

The classic Nikolsky’s sign seen on the skin was first described by Piotr Vasilyevich Nikolskiy who was a Russian dermatologist [33]. Presently, “Nikolskiy” and “Nikolsky” are synonyms in the English literature [32, 33]. Nikolsky’s sign that was originally defined by Nikolskiy is a characteristic of skin lesions in pemphigus foliaceus [34]. Many experts, however, now agree that Nikolsky's sign is elicited by several mucocutaneous disorders, as well as the pemphigus group [32–37]. Grando et al. [33] described two modifications of Nikolsky’s sign, the “marginal” method that is performed on the edge of an active skin lesion and the “direct” method that is on an area of apparently unaffected skin distant from the lesions. The “direct” Nikolsky’s sign is a phenomenon that occurs when an immunological disorder has been implicated such as in pemphigus [38]. This finding supports the concept that immune deposits in autoimmune bullous diseases may be present in outwardly normal-appearing tissue. Sheklakov, another Russian dermatologist, first reported the ability to elicit Nikolsky’s sign in the oral mucosa [33]. This phenomenon is very common in MMP or PV patients with lesions in the oral mucosa as shown in this study. Other autoimmune bullous diseases such as bullous pemphigoid, lichen planus pemphigoides, and paraneoplastic pemphigus show a positive Nikolsky’s sign in the mouth although the number of patients is small [32, 39]. In addition, there are a number of other non-autoimmune diseases or disorders associated with positive Nikolsky’s sign on the oral mucosa [32, 35]. Oral lichen planus is a chronic inflammatory mucocutaneous disease caused by an unknown etiology. A possible autoimmune etiology has been suggested but not yet confirmed in lichen planus. Nonetheless, a positive Nikolsky’s sign sometimes was identified in patients with erosive oral lichen planus (Figure 7) [40, 41]. Histopathologically, oral lichen planus is characterized by band-like lymphocyte infiltration below the epithelium and basal cell liquefaction [40, 42]. The basal cell liquefaction may cause the epithelial separation from underlying connective tissue, especially if traumatic forces are present [3]. Positive DIF findings are only considered to be
supportive but not diagnostic for oral lichen planus [3]. Erythema multiforme is a rare, acute reactive disorder that can affect the skin and mucous membranes. The clinical appearance of oral lesions may present as diffuse erythema, bulla, erosions, and ulcerations with or without pseudomembrane [43, 44]. The vermilion border of the lips is often involved. Nikolsky’s sign of the gingiva has occasionally been described (Figure 8) [32]. The diagnosis of oral erythema multiforme is often difficult because the clinical features may mimic other oral inflammatory and vesiculobullous diseases or disorders. The diagnosis is usually supported by biopsy and exclusion of other causes [43, 44]. On rare occasions, gingival lesions caused by

Figure 7.
Desquamative gingivitis in erosive oral lichen planus. Localized erythematous lesions were found in the attached gingiva. The “marginal” Nikolsky’s sign showed a positive reaction. The histopathological findings indicated band-like lymphocyte infiltration below the epithelium and basal cell liquefaction.

Figure 8.
Erythema multiforme with epithelial desquamation. The clinical manifestations of severe oral ulceration can be difficult to differentiate from autoimmune bullous diseases. Histopathological and direct immunofluorescence findings were nonspecific. The “marginal” Nikolsky’s sign of the gingiva showed a positive reaction.

Figure 9.
Gingival injuries caused by excessive toothbrushing. Sharply demarcated abrasions of the gingiva were seen and may mimic the “marginal” Nikolsky’s sign elicited by autoimmune bullous diseases. The gingival trauma was arrested quickly by making the patients aware that it was caused by incorrect toothbrushing and that it could be alleviated by learning correct oral hygiene practices. Their gingival trauma has not recurred since their treatment.
excessive or improper oral hygiene practices or by hypersensitivity reactions to oral hygiene products such as toothpaste or mouth rinses may mimic positive Nikolsky’s sign elicited by autoimmune bullous diseases (Figure 9) [45–47]. Biopsy may provide histopathologic evidence supporting the diagnosis, but DIF is often not indicated because it is routinely negative since intact epithelium may be required to validate the diagnosis. Eliminating causative agents leads to disappearance of gingival involvement in most patients with hypersensitivity reactions to dental or dental hygiene products.

After the diagnosis of MMP or PV, patients often require an extraoral examination by medical specialists including a dermatologist, an ophthalmologist, and an otolaryngologist. All patients with extraoral involvement should be managed by medical specialists using systemic treatment with or without hospitalization. Patients with exclusively oral lesions may be managed using moderate to very-high-potency topical corticosteroid therapy often combined with effective dental plaque control. The therapeutic goal for DG lesions is the remission or suppression of the clinical signs and symptoms such as gingival soreness, bleeding, and swelling as shown in Table 4. Response to therapy can be assessed to determine whether or not the patient exhibits a positive Nikolsky’s sign or other evidence of ongoing disease. The disappearance of lesions and of Nikolsky’s sign may indicate a favorable treatment outcome.

5. Conclusions

Nikolsky’s sign is a simple nondiagnostic test that may suggest a need for biopsy diagnosis of autoimmune or other diseases in the oral mucosa. The gingiva is often a preferable site for performing the test for Nikolsky’s sign especially if DG is present. A positive reaction of this sign is the basis for suspecting autoimmune bullous diseases such as MMP and PV. In that case, it is critical to conduct DIF testing in conjunction with histopathological examination to establish the final diagnosis. It is also important to remember that DG is a general descriptive term rather than a diagnosis. Oral healthcare providers have a great responsibility to remain suspicious of unexplained oral manifestations of systemic or unusual intraoral diseases and disorders. The presence of a positive oral Nikolsky’s sign serves as a warning that careful evaluation is needed in search of the etiology of the sign. Once other causes have been eliminated, the clinician must remain aware that biopsy or referral for biopsy may be necessary to determine the current diagnosis. It is important to remember, however, that biopsy of tissue sites that feature Nikolsky’s sign is not indicated because a positive Nikolsky’s sign is indicative of friable epithelium and proper diagnosis is predicated on obtaining a biopsy from a site with intact epithelial surfaces. Nonetheless, this simple test for Nikolsky’s sign may serve as a valuable indicator of underlying autoimmune or other diseases and lead to obtaining a correct diagnosis. In many instances patients with diseases or disorders featuring gingival Nikolsky’s sign may require appropriate referral to other dental or medical specialists after identifying suspicious lesions. For the early diagnosis of autoimmune bullous disorders, oral healthcare providers should consider the use of this test that may ultimately lead to the early diagnosis of MMP and PV or other diseases or disorders.

Conflict of interest

The authors report no conflicts of interest related to this study.
Gingival Nikolsky’s Sign: A Valuable Tool in Identifying Oral Manifestations of Mucous...  
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