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A case report of recurrent herpetic gingivostomatitis with special reference to the role of cytology in diagnosis

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Abstract:
Most of the oral ulcers are similar in clinical appearance and the recurrent ulcers may not be diagnosed solely on their clinical appearance and require systematic approach for diagnosis by the clinicians to reduce the ailments of the patient and start early treatment. Biopsy, serological tests and culture seem to be either invasive or time consuming procedures from the viewpoint of a dental outpatient. However, cytological smear at this stage is non-invasive, less time consuming and easily acceptable by the patient. A case of herpetic gingivostomatitis, with diagnosis based on cytology in correlation with clinical presentation and history is discussed.

Keywords: Herpetic gingivostomatitis, cold sores, herpetic ulcers, herpes simplex virus, oral ulcers, cytology, Tzanck test.

Introduction:
An ulcer is described as “a breach in the continuity of the epithelium that may either follow molecular death and disintegration of the surface epithelium or its traumatic removal”.1

The recurrent aphthous minor, aphthous major, herpetiform aphthous ulcers, recurrent herpes lesions, hand foot and mouth disease, herpangina, cyclic neutropenia, PFAPA syndrome (periodic fever, aphthous stomatitis, pharyngitis and adenitis syndrome), Behcet’s syndrome are the common recurrent ulcerative lesions affecting the oral mucosa. Some skin diseases like erosive lichen planus, pemphigus, erythema multiforme, ulcers of infectious origin like syphilis, acute necrotizing ulcerative gingivitis, tuberculosis and mycosis, ulcers of immune disorders like in human immunodeficiency virus (HIV) infection and diabetes, ulcers due to gastrointestinal diseases like Crohn’s disease, ulcerative colitis and malabsorption syndrome and ulcers of blood disorders like cyclic neutropenia, leukemia may also manifest as recurrent ulcers.2

If stimulus persists, radiation induced, drug-induced and traumatic ulcers can also be recurrent. Herpetic infections in particular are often encountered in dental clinic and pose occupational hazard to dentists. The diagnosis and management of these lesions is mainly based on a thorough history, clinical examination, appropriate investigations, and finally, the evaluation for modifications in the regular management based on diagnosis.3

Cytology was first used in cutaneous disorders by Tzanck in 1947 for the diagnosis of vesiculo-bullous disorders which is much forgotten.4 Diagnostic cytology is a simple, rapid, inexpensive and reliable. Various cytological methods include aspiration cytology, imprint smear, exudate smear, skin/mucosa scraping smear and Tzanck smear. In many instances cytological findings are diagnostic, but in some they are only suggestive of a disease which should be confirmed by histopathologic examination.5 Though not always confirmatory, cytology remains as an indispensable tool in the diagnosis of mucosal lesions. Cytology can give a picture of the lesion, though not very clear, which may sometimes be path changer in the diagnosis as in the present case.
Case Report:
A 65 year old male reported with a complaint of pain in the right lower front region and ulcers in the mouth since 8-10 days. Pain was localized, gradual in onset, dull aching, continuous, of moderate intensity and was associated with difficulty in eating. There was no history of aggravating or relieving factors. Patient had the habit of beedi smoking. He had fever and body ache 15 days back; following which ulcers appeared and persisted for the past 7 days. Patient also gave a history of similar ulcers one and half month back for which he consulted a local doctor and took multivitamin supplements after which the ulcers subsided.

Intraoral examination revealed multiple ulcers on the lower labial mucosa, right buccal mucosa, alveolar mucosa, floor of the mouth and posterior hard palate (Fig 1, 2 & 3). Base of the ulcers were covered by slough and was surrounded by erythematous halo. Ulcers measured 1cm X 1cm, were irregular in shape, tender on palpation and showed non-indurated soft edges on palpation. Submandibular lymphnodes were palpable. Routine blood investigation values were within the normal range except for a slight increase in erythrocyte sedimentation rate. Provisional clinical diagnosis of herpetic gingivostomatitis was made. For confirmation, Tzanck test was performed by gently scraping the base of the lesion with the spatula. Cytosmears were prepared, allowed to air dry and were stained with haematoxylin and eosin and Papanicolaou stain.

Microscopic examination of the cytosmears revealed multinucleated giant cells/syncitial cells and ballooning degeneration of cells (Fig 4). The nuclei of these cells were glassy and homogenous with pink viral inclusions. All the above features were suggestive of recurrent herpes simplex gingivostomatitis. Patient was prescribed oral acyclovir (800 mg five times per day for 10 days) and chlorhexidine mouthwash, topical mucopain for relief of symptoms. Patient was also advised to take adequate rest, liquid supplements and soft diet till symptom subsided. Patient was however lost to followup.

Discussion:
The common ulcers like traumatic ulcers and ulcers due to irritation heal faster and do not recur. However, recurrent ulcers cause considerable aliment to the patient. At the other end of the spectrum of ulcerative lesions, long standing chronic ulcers and those with systemic signs have to be biopsied compulsorily for a definitive diagnosis. Herpes simplex virus 1(HSV1) causes primary herpetic gingivostomatitis, more common in 1-5 years, sometimes affects adolescents and young adults with lesions mostly on non-keratinized mucosa. This condition is contagious, can spread through contact of body fluids like saliva and can be sexually transmitted. It is characterized by a prodromal fever, headache and myalgia, sorethroat followed by severe gingivostomatitis, inability to eat, hypersalivation, halitosis and lymphadenopathy. Primary form can be asymptomatic if it is subclinical. Recurrent HSV1 lesions in immunocompetant individuals tend to be more severe with mucocutaneous labial lesions (herpes labialis) and rarely when intraoral sites are involved, ulcers on keratinized mucosa especially on palate and attached gingiva are seen. The present case of gingivostomatitis is HSV1 reactivation/recurrent form showing multiple intraoral lesions affecting both keratinized and non-keratinized mucosa. Trigeminal ganglion is the primary site for HSV-1 latency. Reactivation occurs in up to half of infected individuals, stimulated by emotional factors like stress, hormonal changes, ultra violet light, excess of sunlight, gastrointestinal disturbances, fever, trauma, dental procedures and immunosuppression. Vesicles usually rupture within 2-3 days to form shallow, painful ulcers persisting for several days. Lesions heal within 7-14 days without scarring in otherwise healthy individuals. However, it can complicate for severe lesions in immune compromised individuals and may lead to systemic viremia. Varicella zoster virus infection, Bell’s palsy and Lyme’s disease are few complications. Herpes labialis can result in pain, paresthesia, burning sensation on lip and perioral sites like ala of nose. Herpetic whitlow is commonly seen on fingers due to autoinoculation characterized by pain, erythema and swelling of digital pulp space, nail folds or lateral aspects of finger. Sometimes pain radiates up the arm accompanied with fever, malaise and axial lymphadenopathy. Herpetic keratoconjuntivitis can also occur due to autoinoculation and is associated with pain, blurring of vision, lacrimation, photophobia and characteristic dendritic lesions of the cornea. In recurrent cases, opacification of cornea and loss of vision can occur. Most of the differential diagnosis mentioned earlier can be ruled out by the history, clinical and systemic examination. Recurrent aphthous ulcers are most often mistaken for recurrent herpes lesions (Table 1). Terri et al. gave a detailed clinical review to differentiate aphthous from herpetic ulcers.
<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Recurrent Apthous ulcers</th>
<th>Recurrent Herpetic ulcers</th>
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</thead>
<tbody>
<tr>
<td>Synonyms</td>
<td>Canker sores</td>
<td>Cold sores</td>
</tr>
<tr>
<td>Spread</td>
<td>Non-contagious</td>
<td>Contagious</td>
</tr>
<tr>
<td>Clinical presentations</td>
<td>Not preceded by vesicles, regular ulcers covered with necrotic slough. Usually with erythematous halo due to secondary bacterial infection. Three forms - minor, major and herpetiform</td>
<td>Preceded by vesicles which rupture to form irregular shape, vesicles occur in crops, later rupture leaving ulcerative lesions. Sometimes coalesce to form large lesions</td>
</tr>
<tr>
<td>Symptoms</td>
<td>Usually not associated with fever</td>
<td>Usually associated with prodromal symptoms like fever</td>
</tr>
<tr>
<td>Systemic involvement</td>
<td>No systemic disease</td>
<td>Systemic lesions can be seen like ocular and genital lesions, herpetic whitlow</td>
</tr>
<tr>
<td>Site</td>
<td>Non-keratinized mucosa, ulcers recur at different site</td>
<td>Both non-keratinized and keratinized mucosa, ulcers recur at same site, unilateral</td>
</tr>
<tr>
<td>Investigations</td>
<td>No known well established</td>
<td>Serology, viral culture,</td>
</tr>
</tbody>
</table>
Various diagnostic methods for HSV 1 include serology, viral culture, cytology, immunofluorescence and PCR. Differentiating HSV lesions from others lesions is needed for a prompt treatment. Skin lesions like pemphigus and lichen planus show oral ulcers within a diffuse or vesiculobullous lesion and also with associated lesions on the skin. The Tzanck smear in pemphigus have acantholytic cells having basophilic cytoplasm which are non-cohesive and rarely form giant cells; lichen planus and erythema multiforme show necrotic keratinocytes and lymphocytes instead of acantholytic cells. Herpangina and Foot-and-mouth disease involves oropharynx and palms/soles respectively. Foot and mouth disease shows cells with syncytial nuclei in a Tzanck smear.5 Immunologic diseases can be ruled out by proper history of organ/bone marrow transplantation or steroid medication. Serologic investigations rule out neutropenic ulcers, HIV infection and Leukemia.6 Serology is also not reliable for diagnosis of primary herpetic lesions at early stage since, immunoglobulins are not detected but can be useful in recurrent lesions.10 The sensitivity of investigations will decrease when samples are taken from older resolving herpetic lesions or lesions more than 72 hours old which will yield negative culture. Old age and poor nutritional status may also be the additional factors contributing to the ulcers.

In the present case, Tzanck test was performed. Durdu M et al. showed that the sensitivity and specificity of this cytologic test for diagnosing mucocutaneous herpetic infections was 84.7% and 100% respectively.9 The Cytological findings of Tzanck test in various dermatological lesions was reported by Gupta et.al.5 Ballooning degeneration of epithelial cells, multinucleated cells, intranuclear Lipschutz bodies, chromatin margination are observed in cytosmear.11 However, Tzanck smear cannot differentiate the lesions between herpes subspecies. HSV2 and varicella which can also infect the oral cavity and it is very difficult to differentiate clinically but DNA sequencing7 and clinical presentation (like involvement of single surface), viral culture and direct immunofluorescence in Varicella help to differentiate.5 Viral culture and PCR remain the gold standard for diagnosis of subspecies.

**Limitations of Tzanck smear Cytology:** Ideally, a vesicle less than 3 days old should be obtained since older lesions may get crusted or secondarily infected and the characteristic cytomorphology may no longer be present. Tzanck preparation shows signs of infection in only 50–70% of people with a herpes infection.12 A negative Tzanck preparation may have to be confirmed by a herpes culture or other laboratory test.13

Tzanck smear cytology does not distinguish between HSV-1 and HSV-2, nor between HSV and varicella zoster virus infection, as described by Singh A et al.14 A biopsy from the center of the ulcer without epithelium may not show viral inclusions and cannot be presumed negative for herpes simplex infection. Viral culture wherein the vesicles break and release a yellow fluid replete with live viruses that can be cultured or identified with polymerization chain reaction which has high sensitivity and specificity can be done. However these are time consuming and expensive.7,13,14

**Conclusion:**

Though presumptive diagnosis could be made at the time of clinical examination, sometimes the doubt persists in the clinician during the instances of recurrence, patient’s failure to remember history, subclinical infection and disappearance of features. Cytology provides a rapid chair side investigation for the clinician thus enabling early initiation of treatment in such situation.

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**Fig 1: Ulcers present on the lower lip.**

**Fig 2: Ulcer present on the buccal mucosa**
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1. Das S. A Concise textbook of surgery. 4th Ed. Dr. S. Das; 2006. Chapter 11, Ulcer, Sinus and Fistula; p.125

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