A study on the effectiveness of Tzanck smear to diagnose the vesiculobullous lesions in comparison with histopathology

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Abstract

Background: Tzanck smear named after Arnault Tzanck (1886-1954), to evaluate Cytopathology as a quick less invasive method for early diagnosis of bullous lesions.

Aim: To determine the diagnostic value of Tzanck smear in vesiculobullous skin lesions, to evaluate the vesiculobullous lesions and correlating the diagnosis with Tzanck smears and histopathological findings and when required with Immunoflourescence.

Materials and methods: 565 patients clinically diagnosed as vesico-bullous skin lesions were included in the present study from July 2011 to July 2018 for a period of 7 years. Tzanck smear preparation and biopsy as well as immunoflouresence tests were done in all 565 patients.

Results: Out of the 565 patients with vesiculo bullous lesions, 297 were males, and 268 were females. The concordant results between cytology and histopathology was observed in majority (92.7%) direct immunoflouresence test was done in some which also confirmed the diagnosis.

Conclusion: The Tzanck smear test is an inexpensive, useful, and an easy diagnostic tool for vesiculo-bullous lesions of skin diseases and can be recommended as a bedside first line investigation.
Key words
Tzanck smear, Vesiculobullous lesions, Immunofluorescence.

Introduction
Tzanck smear was first introduced by Arnault Tzanck (1886-1954) [1, 2]. Tzanck smear was initially used as a tool of diagnostic cytology for diagnosis of vesiculobullous conditions, especially herpes simplex [3, 4]. Later it evolved and using for several other dermatological conditions like immuno bullous disorders, genodermatosis, cutaneous infections, and cutaneous tumours [5]. The Tzanck smear test is an inexpensive, useful, and an easy diagnostic tool for vesiculo-bullous lesions of skin diseases and can be recommended as a bedside first line investigation. But it requires certain amount of skill and experience for accurate interpretation [5]. Presence of acantholytic cells or typical Tzanck-like cells in Tzanck smears can suggest a diagnosis of pemphigus group of diseases. Skin is the largest desquamating organ and though exfoliative cytology is routinely used for diagnosis in other medical and surgical specialties, studies on cutaneous cytology remain limited. Certain studies have been published in Western literature regarding the accuracy and diagnostic reliability of Tzanck smear [5]. However, there is a paucity of studies in Indian literature regarding the utility and limitations of Tzanck smear cytology for different groups of dermatological diseases [5, 6]. The aim of this study is to highlight the potential usefulness and diagnostic pitfalls of Tzanck smear for diagnosis of various vesiculobullous lesions of the skin [3, 7].

A typical Tzanck cell is a large, roundin shape with a hypertrophic nucleus, and abundant basophilic cytoplasm, and a perinuclear halo. The diagnostic value of Tzanck smear in various erosive and vesiculobullous lesions are limited, especially in Indian literature [1, 2, 7]. Although Tzanck smear can aid in establishing the clinical diagnosis with ease and rapidity and can serve as an adjunct to the diagnostic methods [8, 9, 10, 11].

Aim and objectives
• To determine the diagnostic value of Tzanck smear in vesiculobullous skin lesions.
• To evaluate the vesiculobullous lesions and correlating the diagnosis with Tzanck smears and Histopathological findings and when required with Immunofluorescence.

Materials and methods
The study was conducted at Gandhi Hospital with the involvement of both Dermatology and Pathology Departments. Five hundred and sixty five (565) patients clinically diagnosed as vesiculobullous skin lesions were included in the present study from July 2011 to July 2018 for a period of 7 years. Tzanck smear preparation and biopsy as well as immunofluorescence tests were done in all 565 patients.

All Tzanck smears and skin biopsies from the cases with vesiculobullous disorders irrespective of age, sex and associated diseases were included in the study. The exclusion criteria included: Mechanical, thermal, suction and chemical blisters, Metabolic disorders and others e.g. Irritant contact dermatitis, Eczematous dermatitis. The Tzanck smear preparation was done after obtaining consent from the patient, preferably the bulla of less than 3 days duration is preferred because of chances of secondary infections and rupture may be there in the older lesions. The intact roof of the bulla should be incised along one side with a scalpel and folded back, the fluid should be wiped off carefully with a cotton wool, then the floor of the lesion to be scrapped with the scalpel and spread on the glass slide as a thin film and fixed in methanol. Staining can be done with Giemsa stain, Hemacolory or Diff-Quiky, haematoxylin and eosin, Wright methylene blue, Papanicolaou and Toluidine blue. The stained slides are examined under a light microscopy. Acantholysis is a key

phenomenon in various bullous diseases (Figures – 1 to 18).

**Figure - 1:** Proper Tzanck preparation is scraping cells from the floor of the blister.

**Figure - 2:** Acantholytic Cells.

**Figure - 3:** Herpes simplex 1.

**Figure - 4:** HPE: H & E section studied reveal virus infected acantholytic keratinocytes, ballooned keratinocytes cells.

**Figure - 5:** Multi nucleated giant cells.

**Figure - 6:** HPE: H & E section studied show, intra epidermal vesicle.
Acantholytic cells – Chicken pox
B. Lakshminarayana, Ram Mohan, G. Saisoumya, J. Tulasi, V. Srinivaskumar. A study on the effectiveness of Tzanck smear to diagnose the vesiculobullous lesions in comparison with histopathology. IAIM, 2018; 5(11): 77-83.

**Figure - 7**: Multinucleated giant cells and inflammatory cells in necrotic background.

**Figure - 8**: HPE: Herpes zoster
Intradermal vesicle with ballooning degeneration of the cells. Intracellular edema with multinucleated giant cells.

**Figure - 9**: H & E Section shows intra epidermal bull, acantholytic cells.

**Figure - 10**: Pemphigus vulgaris- H & E section shows suprabasal bulla with acantholytic cells.

**Figure - 11**: Direct immunoflorescence - lace like intercellular spaces of keratinocytes and IgG deposition seen in the lower zones.

**Figure - 12**: Direct Immunoflorescence Ig G deposits in all cells of epidermis

**Figure - 13**: H & E section shows intact epidermis and sub epidermal bulla.

**Figure - 14**: Linear arrangement of IgG in dermo epidermal junction.
Results

The study included 565 patients, divided into four groups: infections, immunologic disorders, genodermatosis, and spongiotic dermatitis.

Two types of infections were noted; 269 viral and in 68 patients bacterial. Among viral group, Herpes simplex was the most common, seen in 119 (21%) patients, followed by Varicella zoster in 75 (13.2%) patients and herpes Zoster in 65 (11.5%) patients. Cases of bacterial infection included 68 cases of bullous impetigo.

The second group of immunologic disorders was seen in 169 (29.9%) patients. This group consisted of two classes of disorders-pemphigus and its variants diagnosed in 135 (79%) patients and pemphigoid in 34 (21%) cases (Table - 1).

Table - 1: Study groups classification.

<table>
<thead>
<tr>
<th>Vesiculo bulous lesions</th>
<th>No of patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herpes simplex</td>
<td>119</td>
</tr>
<tr>
<td>Herpes gestationis</td>
<td>10</td>
</tr>
<tr>
<td>Varicella zoster</td>
<td>75</td>
</tr>
<tr>
<td>Pemphigus vulgaris</td>
<td>61</td>
</tr>
<tr>
<td>Bullous pemphigoid</td>
<td>34</td>
</tr>
<tr>
<td>Pemphigus foliaceous</td>
<td>30</td>
</tr>
<tr>
<td>Linear IGA Dermatosis</td>
<td>10</td>
</tr>
<tr>
<td>Dermatitis herpetiformis</td>
<td>51</td>
</tr>
<tr>
<td>Sub cornual pustular dermatosis (SCPD)</td>
<td>7</td>
</tr>
<tr>
<td>Grovers disease</td>
<td>7</td>
</tr>
<tr>
<td>Drug induced bullous pemphigoid</td>
<td>7</td>
</tr>
<tr>
<td>Bullous impetigo</td>
<td>68</td>
</tr>
</tbody>
</table>

Tzanck smear findings

Cytologic examination was performed in all the 565 cutaneous infectious lesions, showing acantholytic cells and multinucleated giant cells. Tzanck smear positivity in the 269 cases of viral infections was 56.7% some showed, ballooning degeneration, and inflammatory cells. Predominently neutrophils were noted in bullous impetigo.
Acantholytic cells and few acute and chronic inflammatory cells found in 75% pemphigus lesions from 135 patients. All the lesions were subjected to Histopathological examination by staining with Hematoxylin and Eosin.

Direct Immunofluorescence was done in 105 lesions. Indirect Immunofluorescence was done in 98 lesions. Sex ratio distribution in various conditions was as per Table – 2.

Table - 2: Sex ratio distribution in various conditions.

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>Male to Female ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herpes simplex</td>
<td>1 : 1.26</td>
</tr>
<tr>
<td>Varicella zoster</td>
<td>1 : 1.152</td>
</tr>
<tr>
<td>Herpes zoster</td>
<td>1 : 0.62</td>
</tr>
<tr>
<td>Epidermolysis bullosa</td>
<td>1 : 2.15</td>
</tr>
<tr>
<td>Pemphigus vulgaris</td>
<td>1 : 1.059</td>
</tr>
<tr>
<td>Bullous pemphigoid</td>
<td>1 : 0.78</td>
</tr>
<tr>
<td>Pemphigus foliaceous</td>
<td>1 : 0.35</td>
</tr>
<tr>
<td>Linear iga Dermatosis</td>
<td>1 : 1.34</td>
</tr>
<tr>
<td>Dermatitis herpetiformis</td>
<td>1 : 0.46</td>
</tr>
<tr>
<td>Subcorneal pustular dermatosis</td>
<td>1 : 1.36</td>
</tr>
<tr>
<td>Grovers disease</td>
<td>1 : 1.34</td>
</tr>
<tr>
<td>Drug induced bullous pemphigoid</td>
<td>1 : 1.72</td>
</tr>
<tr>
<td>Bullous impetigo</td>
<td>1 : 1.63</td>
</tr>
</tbody>
</table>

Discussion

The Tzanck cells are formed due to the breakage of intercellular bridges between the epidermal cells. The cells are intact without attaching with one another. That results in round cells and formation of bullae or vesicles [1, 2, 8, 12].

The viral infections showed almost 57% positivity with Tzanck smear and confirmed by histopathology and serological tests. Similar results were noted by Durdu, et al. [3], Ozcan, et al. [11], Oranje, et al. reported Tzanck smear positivity 80% [10, 13] when compared with PCR. Sadick, et al. reported 76.9% Tzanck positivity [12]. Solomon, et al. reported 53% sensitivity [14].

All the Pemphigus lesions showed 80% positivity and confirmed by histopathology and Immunofluorescence, similar results were found in the studies of Blank, et al. [15], Ruocco, et al. [8], Shaheen, et al. [16].

Bullous pemphigoid showed plenty of eosinophills and used as exclusion criteria for pemphigus lesions without Acantholytic cells [1, 2, 3].

Bullous Impetigo lesions showed 88% sensitivity, along with tank cells, neutrophils were also found. Durdu etall also reported the similar results [3].

Over all Tzanck smear positivity observed in 81% of all the 565 vesiculobullous lesions. Which concludes cytomorphologic studies can be of use in screening of most of the vesicolo bullous lesions. The Concordant results between cytology and histopathology was seen in majority (92.7%) of lesions studied.

Conclusions

Tzanck, if performed skillfully and with perfection in making cytology a fairly sensitive 'patient compliant' technique for rapid diagnosis of bullous skin lesions. The Concordant results between cytology and histopathology was seen in majority (92.7%) of lesions studied.

Direct Immunofluorescence staining on Tzanck smear is a novel technique for the diagnosis of pemphigus. The Tzanck smear test is an inexpensive, useful, and an easy diagnostic tool for vesiculo-bullous lesions of skin diseases.

References