ORIGINAL ARTICLE

STUDY OF TZANCK SMEARS OVER A PERIOD OF SIX MONTHS

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ABSTRACT: INTRODUCTION: Cytology of cutaneous lesions can be used for preliminary and rapid diagnosis of many skin diseases. Though the gold standard is a biopsy with or without immunofluorescent studies, the use of simple Tzanck test gives reliable information in many varieties of skin lesions. It is especially useful for cutaneous herpetic lesions and also for early presumptive diagnosis of pemphigus vulgaris. **AIMS:** The study was carried out to determine the usefulness of Tzanck test in herpetic lesions, pemphigus vulgaris and other cutaneous lesions. **MATERIALS AND METHODS:** The present study was a prospective one, carried out over a six month period in the department of pathology at Kamineni Academy of Medical Sciences and Research Centre, Hyderabad, from June 2014 to December 2014. A total of twenty-one Tzanck smears were studied. The material was collected in the Dermatology out-patient department by the dermatologist and sent to the laboratory. A Giemsa stain was used for all the smears and examined under the microscope. **RESULTS:** There were total 21 cases ranging in age from 3 years to 62 years. There were 11 male and 10 female patients, the male to female ratio being 1.1:1. There were 8 cases of herpes zoster and Tzanck smear was reported positive in 5 cases (62.5%). For pemphigus vulgaris, out of 4 cases, 2 (50%) were reported positive on Tzanck smear. As biopsy was not available for all the cases the histopathological correlation could not be done. **CONCLUSION:** Tzanck smear is a rapid, simple, inexpensive and reliable test which is useful for the diagnosis of cutaneous viral infections like herpes zoster, varicella and molluscum contagiosum. It is also helpful for the presumptive diagnosis of pemphigus vulgaris thereby facilitating early treatment of the patients.

KEYWORDS: Tzanck smear, herpes infection, pemphigus vulgaris.

INTRODUCTION: The Tzanck smear was introduced for the first time by the Frenchman Arnault Tzanck[1] in 1947 for diagnosing vesiculo-bullous disorders especially cutaneous herpetic lesions. Theskin is the largest desquamating organ, but not much attention has been paid to cutaneous cytology. As the skin is easily accessible for biopsy and also as special studies like immunofluorescence can be carried out on biopsy material, it becomes the method of choice to study skin lesions. However, cytology of cutaneous lesions can be used for preliminary and rapid diagnosis of many cutaneous disorders like genetic disorders (Hailey-Hailey disease), viral and non-viral infections, vesiculo-bullous disorders like pemphigus vulgaris, bullous pemphigoid, paraneoplastic pemphigus and even tumors.[2]

MATERIALS AND METHODS: The present study was a prospective one, carried out over a six month period in the department of pathology at Kamineni Academy of Medical Sciences and Research Centre, Hyderabad, from June 2014 to December 2014. A total of twenty-one Tzanck

smears were received during this period. Brief clinical details and the provisional diagnosis were noted for all the cases.

Procedure for collection of the material: The samples were collected in the dermatology out-patient department. A fresh blister usually within 48 hours of onset was selected. After de roofing the blister with a scalpel, the fluid was discarded and the base of the blister was scraped gently but firmly. The scraped material was spread on a slide, air dried and sent to the laboratory where a Giemsa stain was carried out. The smears were reported on the same day.

RESULTS: There were a total of 21 cases ranging in age from 3 years to 62 years. There were 11 male and 10 female patients, the male to female ratio being 1.1:1. (Table 1 and Table 2) Herpes zoster was suspected clinically in eight cases and Tzanck smear from these cases showed positive result in the form of multinucleated giant cells in five cases. (Figure 1). Varicella was suspected in three cases and Tzanck smear came positive in one case. Pemphigus vulgaris was suspected clinically in five cases out of which the typical acantholytic cells (Figure 2) were present in two cases which were confirmed by histopathology and direct immunofluorescence. Out of 5 cases of pemphigus vulgaris, biopsy along with immunofluorescence study was available in three cases. On biopsy and direct immunofluorescence study (DIF), two were given as pemphigus vulgaris; one was reported as cutaneous small vessel vasculitis. For both biopsy proven cases of pemphigus vulgaris, the Tzanck smears had shown 'acantholytic cells'. Histopathological examination was done in six out of twenty-one cases. One case of suspected bullous pemphigoid showed nonspecific findings on Tzanck smear but the subsequent biopsy and direct immunofluorescence study showed features of epidermolysis bullosa aquisita. Two cases of Molluscum contagiosum were subjected to Tzanck smear of which one case (50%) was reported positive for the 'molluscum bodies' and the other was given as nonspecific findings. In this second case biopsy showed the typical features of molluscum contagiosum.

DISCUSSION: Cytology is not used widely for diagnosis of cutaneous lesions the reason being skin lesions are easily accessible, and amenable to biopsy, and hence, the latter becomes the preferred diagnostic method. Clinical identification of viral infections like herpes simplex, usually do not pose any diagnostic difficulty. But problems may arise when the features overlap with those of aphthous ulcers, with other venereal diseases or with insect bites.[2] For such cases, a Tzanck preparation can reveal the pathognomonic, large, multinucleated keratinocytes and works as a reliable diagnostic tool. Sometimes atypical presentation or generalized presentation of herpes zoster or varicella in adults may be misdiagnosed as bacterial folliculitis. In such situations also the Tzanck smear helps in correct diagnosis.[3] In cases of outbreaks of Kaposi's varicelliform eruption also the Tzanck smears can be carried out easily on a larger scale and help in rapiddiagnosis.[4] Durdu et al in their study found a sensitivity of Tzanck smear for multinucleated giant cells in herpetic infections as 84.7%.[5] Oranje et al have also reported similar sensitivity and a specificity of 90% for multinucleated giant cells for herpetic lesions,[6] while Nahass et al have reported positive Tzanck smears in 60 % and 75 % of herpes simplex and varicella infections.[7]In our study, five out of eight cases (62.5 %) showed positive Tzanck results for herpes zoster. The

reason for this slightly low sensitivity could be crusting of the lesions causing difficulty in sampling.

Tzanck smears are especially useful in the diagnosis of vesiculo-bullous lesions of pemphigus vulgaris, where the typical acantholytic cells are seen on the smear and are also called as 'Tzanck cells'[8] whereas, the presence of numerous eosinophils points to a diagnosis of bullous pemphigoid. As the Tzanck smear takes less time than the biopsy, a positive report enables the clinician to start treatment early thereby improving the disease outcome.[9] In our study also in five suspected cases of pemphigus vulgaris, one on biopsy was found to be small vessel vasculitis and two out of the remaining four cases (50%) had shown the typical acantholytic cells. For these patients, the treatment for pemphigus vulgaris was started immediately pending the biopsy and immunofluorescence studies. Subsequently, biopsy and immunofluorescence studies of both these cases were reported as pemphigus vulgaris. For the other two cases biopsy was not available and we assume them to be pemphigus vulgaris. In a study by Durdu et al, the sensitivity of 'acantholytic cells' for pemphigus vulgaris was reported as high as 100%.[5]Direct immunofluorescence on the cytology smear can also be used to demonstrate IgG between the acantholytic cells which supports a diagnosis of pemphigus vulgaris.[10]Molluscumcontagiosum can sometimes be difficult to diagnose in adults especially when it presents as isolated or umbilicated lesion and at times it may be misdiagnosed as milia. Tzanck smear if done, shows the 'molluscum bodies' which appear as large hyperbasophilic ovoid anucleate masses.[2]Tzanck smear is also helpful to differentiate between toxic epidermal necrolysis (TEN) and staphylococcal scalded skin syndrome (SSSS) where early diagnosis directs the correct treatment plan. SSSS is a life threatening blistering skin disease caused by certain strains of Staphylococcus aureus and may clinically resemble TEN which is a severe form of cutaneous drug reaction. In SSSS, the Tzanck smear shows numerous viable, acantholytic keratinocytes without inflammatory cells, whereas, TEN shows few necrotic keratinocytes, fibroblasts and inflammatory cells.[11] Other tests that can be used for cutaneous lesions depending on the clinical presentation and provisional diagnosis are methylene blue, Gram staining, potassium hydroxide examination, viral serology, bacterial and fungal cultures.[5] Viral cultures can confirm the diagnosis of varicella or herpes zoster in only 60 % to 64 % cases, whereas, Tzanck smears can confirm in 80% to 100% cases.[12]

The limitations of Tzanck smear are that when slides are improperly prepared from a vesicle that is crusted or when the base of the lesion is not scraped, the representative material will be missed. Also the diagnostic yield is most efficient within 2 to 3 days after the onset of symptoms. Sometimes poorly preserved cells may resemble neoplastic cells of squamous cell carcinoma. Another disadvantage is that it cannot differentiate between herpes simplex virus infection and varicella zoster virus infection.[6] To have a definitive diagnosis, either immunohistochemistry has to be performed on the sections as even the routine hematoxylin and eosin biopsy sections show similar features and cannot differentiate between the two.[13] Employing a polymerase chain reaction (PCR) assay can also be helpful in such situations.[3]

The advantages of Tzanck smear test are that it is easy to perform, simple, inexpensive procedure which is suitable for office diagnosis and does not require a specialized laboratory.[6,14] It causes negligible trauma and discomfort to the patient, and is well-tolerated. It can be performed in highly anxious patients and children and is especially useful for sites which are

difficult to biopsy like the eyelids, lips, oral cavity or genital area. In the study reported by Folkers et al the sensitivity of Tzanck smears for mucous membrane lesions was found to be 81%.[14] It is a better choice over biopsy for cosmetically sensitive areas like face, as biopsy may leave a scar.[2] The Tzanck smear test can also be used as a diagnostic adjunct to dermatoscopy for cutaneous pigmented lesions.[15] In addition, the cytology test can also be used for cutaneous malignancies as reported by Naraghi et al who found a sensitivity and specificity of 87.3% and 95.7% for cutaneous basal cell carcinoma.[16]

CONCLUSION: Tzanck smear is a rapid, simple, inexpensive and reliable test which is useful for the diagnosis of cutaneous viral infections like herpes zoster, varicella and molluscum contagiosum. It is also helpful for the presumptive diagnosis of pemphigus vulgaris thereby facilitating early treatment for the patients. It should be used as an adjunct to histopathological and immunofluorescent studies.

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Clinical diagnosis	Number of cases	Percentage
Herpes zoster	8	38.09%
Varicella zoster	3	14.28%
Pemphigus vulgaris	5	23.80 %
Bullous pemphigoid	1	4.76 %
Subcorneal pustular dermatosis	1	4.76 %
Molluscumcontagiosum	2	9.52 %
Bacterial folliculitis	1	4.76 %
Total	**21**	**100 %**

Table 1: Clinical Diagnosis

Tzanck smear findings	Number of cases	Percentage
Nonspecific findings	11	52.38 %
Herpes zoster	5	23.80 %
Varicella zoster	1	4.76 %
Pemphigus vulgaris	2	9.52 %
Subcorneal pustular dermatosis	1	4.76 %
Molluscumcontagiosum	1	4.76 %
Total	**21**	**100 %**

Table 2: Tzanck smears

Fig. 1: Tzanck smear from herpes zoster lesion showing multinucleated keratinocytes and numerous acantholytic cells. (Hematoxylin and eosin stain, 400X).

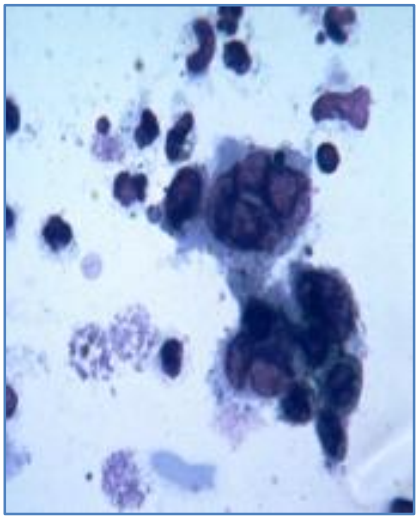

Fig. 1

Fig. 2: Tzanck smear from pemphigus vulgaris lesion showing typical 'acantholytic cells' (Hematoxylin and eosin stain, 400X).

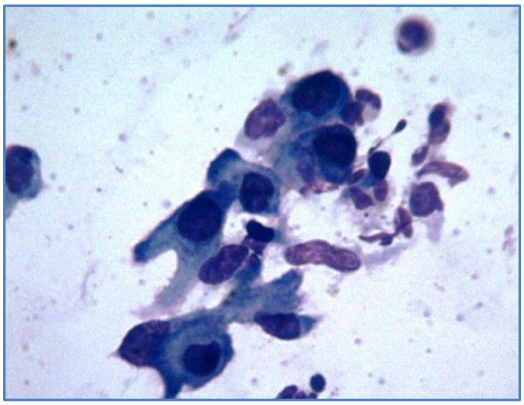

Fig. 2

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