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The Diagnostic Value of Tzanck Test in Vesiculobullous, Pustular, and Oral Lesions in The Dermatology Clinic

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ABSTRACT

Background: Vesiculobullous disorders represent a heterogeneous group of dermatoses with protean manifestations. The accurate diagnosis of bullous disorders of the skin and mucous membrane requires evaluation of clinical, histological, and immunofluorescence findings.

Objective: The aim of this study was to investigate Tzanck smear findings and to determine the diagnostic value of this test in moist (erosive, vesicular, bullous, and pustular) skin and oral lesions.

Patients and Methods: This cross sectional study was conducted on one hundred patients presented with vesicular, bullous, and pustular skin lesions and/or oral lesions. All patients were included from Dermatology Outpatient Clinic at Al-Zahraa University Hospital over the period from March 2018 to March 2019.

Results: Positivity of multinucleated giant cells in herpetic infection, acantholytic cells in pemphigus, dyskeratotic acantholytic cells and cocci in bullous impetigo were useful landmarks in Tzanck smear. A decrease in viral shedding from herpetic lesions of longer duration may correlate with the lower sensitivity of the Tzanck smear. Therefore, in this study we excluded the lesions older than 3 days. The percentage of positivity in vesicles was much higher than that of pustules.

Conclusion: According to this study, Tzanck smear was an inexpensive, simple, and rapid test and did not require any specialized laboratory equipment for evaluation of various vesiculobullous and pustular lesions either skin or mucosal one.

Keywords: Tzanck test, Vesiculobullous, Pustular, Oral lesions, Dermatology.

INTRODUCTION

Vesiculobullous disorders represent a heterogeneous group of dermatoses with protean manifestations. The accurate diagnosis of bullous disorders of the skin and mucous membrane requires evaluation of clinical, histological, and immunofluorescence findings **(1).** Cytology may be used as a diagnostic tool in a wide range of dermatological conditions. It enables rapid diagnosis at low cost and swift referral of the patient for appropriate treatment. The procedures are well tolerated by the patient and complications are rare. In addition to its role in the diagnosis of disease, cytology is useful in monitoring disease progress and detecting relapse **(2).** In developing countries, clinical information supported by cytodiagnosis is valuable in the investigation of various infectious diseases. Furthermore, cytology can provide highly reliable information concerning a variety of skin tumours in situations where biopsy is to be avoided **(2)**. Cytological sampling techniques result in minimal tissue injury compared with biopsy. With the increasing use of new, non-invasive topical treatment modalities for non-melanoma skin, cancer cytology may become the diagnostic method of choice. A comprehensive knowledge of the cytological features of primary skin neoplasms is particularly important in establishing the precise origin of a skin tumour and to distinguish it from metastasis from another site **(2)**. As a method for the diagnosis of cutaneous disorders, cytology was first used by Arnault Tzanck in 1947. Although it was suggested as a simple, rapid, and reliable technique to be used in the diagnosis of many diseases during the following 6 decades, the practice of cytodiagnosis has been limited to a few diseases **(3)**.

To date, only a few studies have examined the dermatological use and diagnostic value of this method. The majority of these studies have related to herpetic infections, pemphigus, cutaneous leishmaniasis, and cutaneous neoplasms, especially basal cell carcinoma **(4).**

AIM OF THE WORK

The aim of this study is to investigate Tzanck smear findings and to determine the diagnostic value of this test in moist (erosive, vesicular, bullous, and pustular) skin and oral lesions.

PATIENTS AND METHODS

Patients: This cross sectional study was conducted on one hundred patients presented with vesicular, bullous, and pustular skin lesions and/or oral lesions. All patients were included from Dermatology Outpatient Clinic at Al-Zahraa University Hospital over the period from March 2018 to March 2019.

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Consent: Informed written consent was obtained from the patients and/ or their relatives. The approval from the Research Ethics Committee of the Faculty of Medicine for Girls, Al-Azhar University, was also obtained on the beginning of this study.

Inclusion criteria:

- Both sexes.
- Any age.
- Patients with vesicular, bullous, and pustular skin lesions who were previously diagnosed by clinical, histopathology +/- immunofluorescence
- Patients with oral lesions.

Exclusion criteria:

- Patients who refused the procedure.
- Patients whose diagnosis were not confirmed.

No patients were excluded from this study.

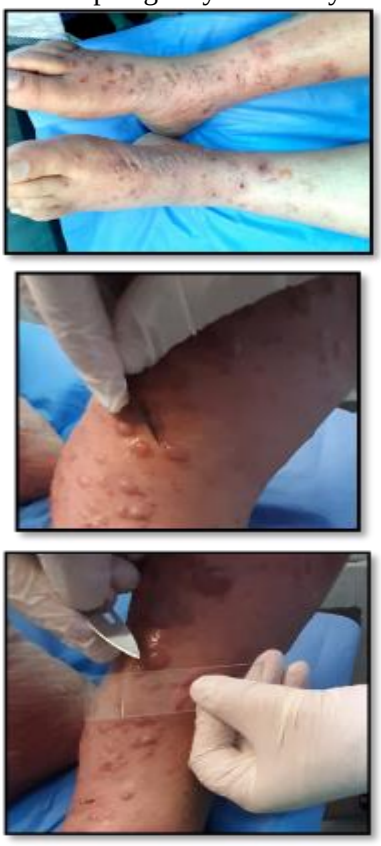
All patients were subjected to the following:

a) Complete history taking.
b) Clinical examination.
c) Photographic documentation.
d) Steps of the procedure:

- *Tzanck smear was obtained from each lesion as follows:*

1. A fresh blister usually within 48 hours of onset was selected;

2. After deroofing the blister with a scalpel blade number 15 (Figure 1), the fluid was discarded and the base of the blister was scraped gently but firmly.



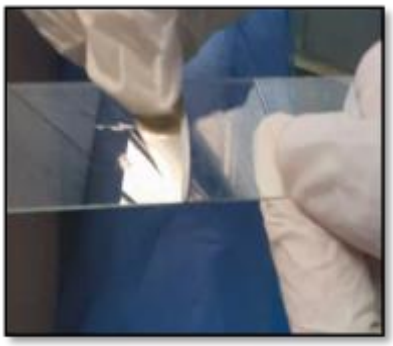


Figure (1): A case of vesiculobullous lesion in 63 years old man. Diagnosed as bullous pemphigoid. De-roofing and scrapping of vesicular lesion using scalpel blade, and spreading of material obtained on microscopic slide. Diagnosed after that as bullous pemphigoid.

3. The obtained cellular material was spread as a thin layer onto a clean microscopic slide, then fixed in methanol for 2–3 min;

4. Staining was accomplished using 2–3 drops of a stock solution of *May–Grunwald–Giemsa* stain, prepared by diluting 1 part of stain with 3 parts of distilled water**,** for another 5-10 min;

5. The stain–water mixture was then poured off and the slide was quickly washed off and allowed to dry.

6. The slide was covered by a cover slip to protect it.

7. The slide was examined by cytopathologist under a light microscope *(Olympus CX 41 Binocular Microscope)*

8. Cytological findings on smears were photographed using *Olympus* camera.

9. Hemostasis could be achieved by pressure with gauze pad.

N.B. As Papanicolaou stain shows intranuclear properties and intranuclear inclusion bodies in more detail than MGG stain and can detect koilocytes. The stain was used in this study.

Statistical analysis of the collected results.

Data were collected, revised, coded and entered to the Statistical Package for Social Science (IBM SPSS) version 23. The quantitative data were presented as mean, standard deviations and ranges when parametric. Also, qualitative variables were presented as number and percentages.

The comparison between groups regarding qualitative data was done by using ***Chi-square test*** and/or ***Fisher exact test*** when the expected count in any cell found less than 5.

Receiver operating characteristic curve (ROC) was used in the qualitative form to assess the sensitivity, specificity, positive predictive value, negative predictive value and accuracy of Tzanck smears test on the standard test.

The comparison between more than two independent groups with quantitative data and parametric distribution was done by using ***One Way ANOVA.***

The confidence interval was set to 95% and the margin of error accepted was set to 5%. So, the p-value was considered significant as the following:

- P-value > 0.05: Non-significant (NS)
- P-value < 0.05: Significant (S)
- P-value < 0.01: Highly significant (HS)

RESULTS

Age of the patients ranged from 3-78 years old with mean (35.14 years). Family history of a similar lesion was positive in 26 cases (26%). Risk factors including drugs in pustular lesion, immunocompromised state in viral lesion, and Niklosky sign in immune bullous lesion were positive in 59 cases (59%). Male to female ratio was approximately equal to 1: 1 in all lesions (Table 1).

Table (1): Description of patient's data.

			No. = 100
Age	Mean ± SD		35.14 ± 19.76
	Range		3 – 78
Sex	Female		50 (50.0%)
	Male		50 (50.0%)
Family history	Negative		74 (74.0%)
	Positive		26 (26.0%)
Risk factor	Negative		41 (41.0%)
	Positive		59 (59.0%)

The patients were classified according to nature of the lesion into 3 groups.

- ***Group 1*** (vesiculobullous group) included 73 patients with mean age 35.14 ± 19.76 years. Lesion duration ranged from 1-3 days with the mean 1.51 ± 0.65 days. 28.8% had positive family history of similar lesions. 46.6% had risk factors. This group was subdivided into 2 subgroups according to mechanism of vesicle formation
- *Group 1a:* immune vesiculobullous, which included 31 patients.
- *Group 1b:* viral vesiculobullous, which included 42 patients.
- ***Group 2*** (miscellaneous vesiculobullous group) included 8 cases (8.0%). With the same mean age as previous groups. Mean duration of lesions 1.25 ± 0.46 days. Risk factors were positive in (75%)
- ***Group 3*** (pustular group) included 19 patients (19%) with the mean age of the patients 35.14 ± 19.76 years and duration of lesion 2.58 ± 0.61 days. All patients of this group with positive risk factors (100%).
- High statistically significant difference was found between the three studied groups as regard duration & risk factors (p value =0.000), however no statically significant difference was found regarding sex, family history, size of lesion between the three studied groups (Table 2).

Table (2): Demographic and descriptive data of the lesions in three groups

		Vesiculobullous group	**Pustular group**	**Miscellaneous group**	**Test value**	**P-value**	**Sig.**
		No. = 73	**No. = 19**	**No. = 8**			
Sex	Female	37 (50.7%)	10 (52.6%)	3 (37.5%)	0.566*	0.753	NS
	Male	36 (49.3%)	9 (47.4%)	5 (62.5%)			
Family history	Negative	52 (71.2%)	16 (84.2%)	6 (75.0%)	1.324*	0.516	NS
	Positive	21 (28.8%)	3 (15.8%)	2 (25.0%)			
Duration	Mean±SD	1.51 ± 0.65	2.58 ± 0.61	1.25 ± 0.46	24.026•	0.001	HS
	Range	1 – 3	1 – 3	1 – 2			
Size/cm	Mean±SD	0.51 ± 0.35	0.46 ± 0.29	0.73 ± 0.61	1.514•	0.225	NS
	Range	0.1 – 2	0.1 – 1	0.2 – 2			
Risk factor/s	Negative	39 (53.4%)	0 (0.0%)	2 (25.0%)	18.709*	0.001	HS
	Positive	34 (46.6%)	19 (100.0%)	6 (75.0%)			
Complication/s	Negative	61 (83.6%)	15 (78.9%)	8 (100.0%)	1.895*	0.388	NS
	Positive	12 (16.4%)	4 (21.1%)	0 (0.0%)			

:Chi-square test; •: One Way ANOVA test P-value <0.05: Significant (S); P-value< 0.01: highly significant (HS)

As regard, cytological findings in the studied groups showed various immune vesiculobullous diseases that were encountered in this study and percentage of each type of cells identified after tzanck smear in each of them (Table 3).

Table (3): Immune vesiculobullous diseses

		Pemphigus vulgaris	Pemhigus vegetans	Chronic bullous disease	Bullous pem-phigoid	Test value	P-value	Sig
Superfacial squmaous	No	0 (0.0%)	0 (0.0%)	1 (11.1%)	5 (100.0%)	25.305	0.001	HS
	Yes	13 (100.0%)	4 (100.0%)	8 (88.9%)	0 (0.0%)			
Acantholytic cells	No	0 (0.0%)	0 (0.0%)	9 (100.0%)	5 (100.0%)	31.089	0.001	HS
	Few	2 (15.4%)	1 (25.0%)	0 (0.0%)	0 (0.0%)			
	Plenty	11 (84.6 %)	3 (75.0%)	0 (0.0%)	0 (0.0%)			
Squamous cells with basophilic bodies	No	13 (100.0%)	4 (100.0%)	9 (100.0%)	5 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Dyskeratotic cells	No	5 (38.5%)	3 (75.0%)	5 (55.6%)	3 (60.0%)	1.973	0.578	NS
	Yes	8 (61.5%)	1 (25.0%)	4 (44.4%)	2 (40.0%)			
Tadpole cell	No	12 (92.3%)	4 (100.0%)	9 (100.0%)	3 (60.0%)	6.711	0.082	NS
	Few	1 (7.7%)	0 (0.0%)	0 (0.0%)	2 (40.0%)			
	Plenty	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Neutrophil	No	2 (15.4%)	0 (0.0%)	0 (0.0%)	2 (40.0%)	19.223	0.004	HS
	Few	10 (76.9%)	1 (25.0%)	2 (22.2%)	3 (60.0%)			
	Many	1 (7.7%)	3 (75.0%)	7 (77.8%)	0 (0.0%)			
Eosinophil	No	1 (7.7%)	0 (0.0%)	3 (33.3%)	0 (0.0%)	7.816	0.252	NA
	Few	3 (23.1%)	1 (25.0%)	4 (44.4%)	2 (40.0%)			
	Many	9 (69.2%)	3 (75.0%)	2 (22.2%)	3 (60.0%)			
Lymphocytes	No	2 (15.4%)	0 (0.0%)	2 (22.2%)	0 (0.0%)	2.100	0.552	NS
	Few	11 (84.6%)	4 (100.0%)	7 (77.8%)	5 (100.0%)			
	Many	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Multinucleated giant cells	No	13 (100.0%)	4 (100.0%)	9 (100.0%)	5 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Cocci	No	13 (100.0%)	3 (75.0%)	6 (66.7%)	4 (80.0%)	4.757	0.190	NS
	Yes	0 (0.0%)	1 (25.0%)	3 (33.3%)	1 (20.0%)			
Bacilli	No	13 (100.0%)	4 (100.0%)	9 (100.0%)	5 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			

In Pemphigus vulgaris there were high statistically significant of superficial squamous cell (100 %), acantholytic cell (100), then eosinophil (92.30%), then lymphocyte (84.6%), neutrophil (84.6%), followed by dyskeratotic cells (61.5%) and tadpole cell (7.70%) (Table 3). The main type of cells in cytology of pemphigus vegetans cases were as follow: superfacial squamous cells, acantholytic cells, neutrophil, eosinophil and lymphocyte (100%), then dyskeratotic cell (25%). Cocci was identified in (25%) (Table 3). In Chronic bullous disease of childhood there were high statistically significant of neutrophil (100%), then superfacial squamous cells (89%), then lymphocyte (78%), then eosinophil (66.6%), followed by dyskeratotic cells (44.40%). Cocci was present in (33%) (Table 3). As regard bullous pemphigoid cases there were high statistically significant of eosinophil, lymphocyte (100%), then neutrophil (60%), followed by tadpole, dyskeratotic cell (40%) (Table 3).

As regards cytological findings in the studied groups showed various viral vesiculobullous diseases that were encountered in this study and percentage of each type of cells identified in Tzanck smear in each of them.

Table (4): Viral vesiculobullous diseases.

Viral		Herpes zoster	Herpes simplex	Chicken pox	Molluscum contagisum	HFM	Test value	P-value	Sig.
Superfacial squmaous	No	1 (5.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	1.366	0.850	NS
	Yes	17 (94.4%)	7 (100.0%)	6 (100.0%)	8 (100.0%)	3 (100.0%)			
Acantholytic cells	No	9 (50.0%)	7 (100.0%)	5 (83.3%)	7 (87.5%)	2 (66.7%)	8.313	0.081	NS
	Few	9 (50.0%)	0 (0.0%)	1 (16.7%)	1 (12.5%)	1 (33.3%)			
	Plenty	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Squamous cells with basophylic bodies	No	18 (100.0%)	7 (100.0%)	6 (100.0%)	1 (12.5%)	3 (100.0%)	35.700	0.001	HS
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	7 (87.5%)	0 (0.0%)			
Dyskeratotic cells	No	10 (55.6%)	7 (100.0%)	2 (33.3%)	7 (87.5%)	2 (66.7%)	9.063	0.060	NS
	Yes	8 (44.4%)	0 (0.0%)	4 (66.7%)	1 (12.5%)	1 (33.3%)			
Tadpole cell	No	15 (83.3%)	7 (100.0%)	6 (100.0%)	8 (100.0%)	3 (100.0%)	4.308	0.366	NS
	Few	3 (16.7%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
	Plenty	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Neutrophil	No	8 (44.4%)	3 (42.9%)	3 (50.0%)	6 (75.0%)	2 (66.7%)	5.242	0.731	NS
	Few	9 (50.0%)	4 (57.1%)	2 (33.3%)	2 (25.0%)	1 (33.3%)			
	Many	1 (5.6%)	0 (0.0%)	1 (16.7%)	0 (0.0%)	0 (0.0%)			
Eosinophil	No	18 (100.0%)	6 (85.7%)	5 (83.3%)	8 (100.0%)	2 (66.7%)	6.462	0.167	NS
	Few	0 (0.0%)	1 (14.3%)	1 (16.7%)	0 (0.0%)	1 (33.3%)			
	Many	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Lymphocytes	No	3 (16.7%)	2 (28.6%)	5 (83.3%)	3 (37.5%)	0 (0.0%)	13.172	0.106	NS
	Few	13 (72.2%)	5 (71.4%)	1 (16.7%)	5 (62.5%)	3 (100.0%)			
	Many	2 (11.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Multinucleated giant cells	No	4 (22.2%)	1 (14.3%)	2 (33.3%)	8 (100.0%)	1 (33.3%)	16.692	0.002	HS
	Yes	14 (77.8%)	6 (85.7%)	4 (66.7%)	0 (0.0%)	2 (66.7%)			
Cocci	No	14 (77.8%)	7 (100.0%)	6 (100.0%)	8 (100.0%)	3 (100.0%)	5.895	0.207	NS
	Yes	4 (22.2%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Bacilli	No	18 (100.0%	7 (100.0%)	5 (83.3%)	8 (100.0%)	3 (100.0%)	6.146	0.188	NS
	Yes	0 (0.0%)	0 (0.0%)	1 (16.7%)	0 (0.0%)	0 (0.0%)			

In herpes zoster, superfacial squamous cell was identified in 94% of cases, then lymphocyte (83.3%), then multinucleated giant cell (78%), then neutrophil, acantholytic cells (50%), followed by dyskeratotic cell (44.40%), and tadpole cell (16.7%) (Table 4). In herpes simplex, there were high statistically significant of superficial squamous cell (100%), then multinucleated giant cell (86%), then lymphocyte (71%), then neutrophil (57.1%), followed by eosinophil (14.3%) (Table 4). In chicken pox, there were high statistically significant of superficial squamous cell (100%), then multinucleated giant cell (67%), then dyskeratotic cell (66.7%), then neutrophil (57.1%), followed by eosinophil (14.3%) (Table 4).

While in molluscum contagiosum, there were high statistically significant of superfacial squamous cell (100%), then squamous cell with inclusion bodies (88%), followed by lymphocyte (63%), then neutrophil, acantholytic cell, and dyskeratotic cell (Table 4).

Table (5): Pustular diseases, various pustular diseases and percentage of different types of cells in each of them

Pustular group		Pustular psoriasis	AGEP	Acneiform eruption	Folliculitis	Bullous impetigo	Folliculitis decalvans	Test value	P-value	Sig.
Superfacial squmaous	No	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	NA	NA	NA
	Yes	4 (100.0%)	4 (100.0%)	3 (100.0%)	4 (100.0%)	3 (100.0%)	1 (100.0%)			
Acantholytic cells	No	1 (25.0%)	3 (75.0%)	2 (66.7%)	1 (25.0%)	0 (0.0%)	1 (100.0%)	12.706	0.241	NS
	Few	3 (75.0%)	0 (0.0%)	1 (33.3%)	3 (75.0%)	3 (100.0%)	0 (0.0%)			
	Plenty	0 (0.0%)	1 (25.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Squamous cells with basophylic bodies	No	4 (100.0%)	4 (100.0%)	3 (100.0%)	4 (100.0%)	3 (100.0%)	1 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Dyskeratotic cells	No	0 (0.0%)	3 (75.0%)	0 (0.0%)	1 (25.0%)	1 (33.3%)	0 (0.0%)	7.826	0.166	NS
	Yes	4 (100.0%)	1 (25.0%)	3 (100.0%)	3 (75.0%)	2 (66.7%)	1 (100.0%)			
Tadpole cell	No	3 (75.0%)	3 (75.0%)	3 (100.0%)	4 (100.0%)	3 (100.0%)	1 (100.0%)	3.074	0.689	NS
	Few	1 (25.0%)	1 (25.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
	Plenty	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Neutrophil	No	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	13.359	0.02	S
	Few	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (75.0%)	0 (0.0%)	0 (0.0%)			
	Many	4 (100.0%)	4 (100.0%)	3 (100.0%)	1 (25.0%)	3 (100.0%)	1 (100.0%)			
Eosinophil	No	3 (75.0%)	3 (75.0%)	2 (66.7%)	3 (75.0%)	2 (66.7%)	0 (0.0%)	8.617	0.569	NS
	Few	1 (25.0%)	0 (0.0%)	1 (33.3%)	0 (0.0%)	1 (33.3%)	1 (100.0%)			
	Many	0 (0.0%)	1 (25.0%)	0 (0.0%)	1 (25.0%)	0 (0.0%)	0 (0.0%)			
Lymphocytes	No	0 (0.0%)	1 (25.0%)	0 (0.0%)	2 (50.0%)	2 (66.7%)	0 (0.0%)	17.601	0.062	NS
	Few	4 (100.0%)	3 (75.0%)	2 (66.7%)	2 (50.0%)	1 (33.3%)	0 (0.0%)			
	Many	0 (0.0%)	0 (0.0%)	1 (33.3%)	0 (0.0%)	0 (0.0%)	1 (100.0%)			
Multinucleated giant cells	No	4 (100.0%)	4 (100.0%)	3 (100.0%)	4 (100.0%)	3 (100.0%)	1 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			
Cocci	No	4 (100.0%)	3 (75.0%)	3 (100.0%)	1 (25.0%)	1 (33.3%)	1 (100.0%)	8.972	0.11	NS
	Yes	0 (0.0%)	1 (25.0%)	0 (0.0%)	3 (75.0%)	2 (66.7%)	0 (0.0%)			
Bacilli	No	4 (100.0%)	4 (100.0%)	3 (100.0%)	4 (100.0%)	3 (100.0%)	1 (100.0%)	NA	NA	NA
	Yes	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)			

In Pustular psoriasis, there were high statistically significant of superficial squamous cell, dyskeratotic cell, neutrophil, lymphocyte (100%), then acantholytic cell (75%) (Table 5).

Table (6): Complication of the procedure.

Complication/s of procedure	Negative	84 (84.0%)
	Positive	16 (16.0%)

Complications of the procedure occurred during taking the smear, including mild discomfort and pin point bleeding. All were temporary for minutes, and all spontaneously resolved (Table 6).

DISCUSSION

Tzanck smear has a widespread use in substantiating the diagnosis of vesiculobullous diseases especially the pemphigus groups as well as herpetic infections. It is however, important to interpret tzanck smear findings, along with adequate clinical details, in order to maximally utilize this technique [5]. Tzanck smear test is particularly helpful in providing a provisional diagnosis of pemphigus vulgaris when the site of the lesion is not amenable for biopsy or the disease is in a very early stage. Moreover some patients may develop viral infection on top of longstanding pemphigus lesions due to prolonged immunosuppression so presumptive diagnosis facilitating early & proper treatment of the patients.

In this study cytological features of pemphigus group of lesions displayed typical acantholytic cells 'Tzanck cells'. While other bullous lesions showed scarcity of keratinocytes, absence of acantholytic cells and relative predominance of inflammatory cells. Eosinophils were seen in abundance in cases of bullous pemphigoid**.** Several authors reported the same cytological findings; **Panwar *et al.*** [5] proved the value of Tzanck smear in the initial diagnosis of Pemphigus. Positivity of acantholytic cells in Tzanck smear was 100% for pemphigus. This is in accordance with **Ruocco *et al.*** [6] who reported that positivity of acantholytic cells in Tzanck preparations of pemphigus was between 93.3% and 100%. Similar findings were also reported by **Shaheen *et al.*** [7] in their study on 37 cases of active pemphigus. Moreover, **Yaeen *et al.*** [8] reported the positivity rate of 71.4% in pemphigus.

In our study, sensitivity of Tzanck smear in diagnosis of pemphigus and bullous pemphigoid, in comparison to histopathology was 84.6% and 60%, respectively. This is in agreement with the results reported by **Yaeen *et al.*** [8] who reported that Tzanck smear was 81.8% sensitive when compared to histopathology. **Durdu *et al.*** [9] reported a Tzanck smear sensitivity of 100% in pemphigus. **Shaheen *et al.*** [7] reported the overall sensitivity of Tzanck smear in pemphigus to be 73%.

Accordingly, Tzanck test is considered highly sensitive test in diagnosis of such diseases. Furthermore, as Tzanck smear takes less time than the biopsy, a positive report enables the clinician to start treatment early for improving outcome.

As regard specificity, it was 100% in pemphigus and bullous pemphigoid in this study. This is in disagreement with other studies. In **Yaeen *et al.*** [8] study was 83.33% and 75.0%, respectively and in **Durdu *et al.*** [9] study was 43.4%. This discrepancy may be due to absence of false positive results in our study, as we select the patients who were previously diagnosed.

Although the hallmark for the diagnosis of pemphigus is histopathology and immunofluorescence, Tzanck test is particularly useful in the provisional diagnosis and or differentiating pemphigus flares from herpetic infection especially in oral erosions. It is very helpful and convenient for the patient who is already in pain and distress.

Regarding viral infections, in this study, the presence of inflammatory cells in addition to multinucleation and crowding of the nuclei, nuclear molding, peripheral margination of the nuclear chromatin and ground glass appearance of the nuclei, intranuclear inclusions with a prominent halo and ballooning degeneration and inflammatory cells were considered as typical herpetic changes in cases of clinically suspected viral infections. These findings were encountered in herpes simplex, herpes zoster and chicken pox cases. These findings were similar to those reported by **Yaeen *et al.*** [8]**.**

In this study, the sensitivity and specificity of Tzanck smear were evaluated in contrast with the clinical features of the viral lesions. They were in herpes simplex 85.7% and 100%, respectively. A decrease in viral shedding from herpetic lesions of longer duration may correlate with the lower sensitivity of the Tzanck smear in these cases. Moreover, we depend on sure viral cytopathic effects as multinucleation, crowding of nuclei, intranuclear inclusions. Hence very early lesions may display only nucleomegaly and squamous cell nucleation these features may overlap with other diseases.

The diagnostic value of Tzanck smear when compared to viral serology was investigated by **Yaeen *et al.*** [8]. He reported for herpetic infections, the sensitivity and specificity of the Tzanck smear were 86.33% and 91.30%, respectively. **Durdu *et al.*** [9] noted 84.7% sensitivity and 100% specificity of the test. **Ozcan *et al.*** [10] reported a Tzanck smear sensitivity and specificity of 76.9% and 100% respectively in herpetic infections.

In molluscum contagiosum, the clinical features may sometimes overlap with other infections or milia. In such cases, identification of typical molluscum bodies, which appear as large hyperbasophilic ovoid anucleated masses on Tzanck smear, can be a useful diagnostic clue.

In this study, 7 out of 8 cases clinically diagnosed as molluscum contagiosum showed positivity for squamous cells with basophilic inclusion bodies by Tzanck smear and were diagnosed as molluscum contagiosum. This is in agreement with the findings reported by **Ruocco *et al.*** [6], who reported that sensitivity of Tzanck smear in comparison with clinical features was 87.5% for molluscum. Specificity was 100%. **Krishnamurthy and Nagappa** [11] were among very few authors who reported about the cytological diagnosis of molluscum contagiosum.

To sum up from our study, we found that positivity of multinucleated giant cells in herpetic

infection, acantholytic cells in pemphigus, dyskeratotic acantholytic cells and cocci in bullous impetigo, all were useful landmarks in Tzanck smear.

Numerous studies investigated Tzanck smear in other different diseases as in the study by **Durdu *et al.* [(9)]**. They used Tzanck smears to aid in the diagnosis of cutaneous infections like leishmaniasis, genodermatoses such as Hailey-Hailey disease and tumors (basal and squamous cells carcinomas). However, these groups of diseases were out of scope of this study, hence reliability of tzanck smear cytology for diagnosis of such diseases could not be assessed. Larger studies over a longer period are required to establish the diagnostic value of Tzanck smear in these disorders.

In this study, the limitations of Tzanck smear test were encountered when the slides were improperly prepared from the vesicle that was crusted or when the base of the lesion was not scraped well. Both factors resulted in that representative material would be missed.

CONCLUSION

According to this study, Tzanck smear was an inexpensive, simple, and rapid test and did not require any specialized laboratory equipment for evaluation of various vesiculobullous and pustular lesions either skin or mucosal one.

Considering the advantages and disadvantages of diagnostic tests, the best diagnostic method for vesiculobullous erosive lesions and papular, pustular, nodular, and tumoral lesions is cytology

Tzanck test are used to support the clinical diagnosis especially in early stage of viral infection and its routine use in addition to adequate clinical history can help sharpen the diverse skin lesions.

RECOMMENDATIONS

- Larger studies over a longer period are required to establish the diagnostic value of Tzanck smear in other groups of lesion.
- We recommend the use of Tzanck smear as a first-line investigation for vesiculobullous and pustular lesions.
- The utility of Tzanck smear is no longer restricted to corroborate the diagnosis of pemphigus group of lesions and herpetic infections as before.
- Dermatologists are able to use the Tzanck preparation effectively for diagnosis in office practice with the help of supplemental screening by a cytopathologist.

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