Original article:

Cytopathological and clinical evaluation of various Skin Lesions at a Tertiary Care Hospital

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Abstract:

Aim: Cyto-pathology of skin has been documented to be useful in the diagnosis of several skin lesions. This study aims to evaluate cytopathology as a quick non-invasive method for early diagnosis of bullous lesions, neoplastic and pre-neoplastic skin lesions and to correlate the clinical, cytological and histopathological findings of various skin lesions.

Materials and Methods: 85 patients of skin lesions were included in our study. Skin scraping, slit smears, Tzanck smears, and fine needle aspiration cytology (FNAC) were done to obtain material for cytological examination. Excisional biopsy, punch biopsy and incisional biopsy were done to obtain tissue for histopathologically.The slides were stained with routine stains and special stains as and when required.

Results: Of the 85 patients, 45 were males and 40 females. The non-neoplastic lesions observed were vesico-bullous lesions which comprised of 41 cases followed by neoplastic lesions which consisted of 24 cases, of which six were benign and 18 malignant. Concordant results between cytology and histopathology was seen in majority (91.7%) of lesions studied.

Conclusion:

Cytology performed skillfully and with perfection, leads to an early diagnosis in majority of the lesions, as the observed cyto-morphological features of various skin lesions were fairly distinctive making cytology a fairly sensitive 'patient compliant' technique for rapid diagnosis of skin lesions.

Keywords: cytopathology, Bullous lesions, skin lesions, histopathology.

Introduction

Various lesions affecting the skin range from non-specific dermatoses and various inflammatory diseases to neoplastic changes. Cytology and skin biopsy form the basis of differential diagnosis in clinically similar dermatosis, thereby yielding important information to the pathologist and dermatologist. Though cytopathology was an excellent diagnostic tool in routine dermatologic practice, [1] studies relating to histopathological and cytological correlation are few. The objective of our this study was to evaluate cytology as a quick non-invasive method for early diagnosis of bullous lesions, and to correlate the clinical, cytological and

histopathological findings of various other skin lesions.

Materials and Methods

85 patients with various skin lesions were included in the study. A detailed history of the patients was taken and physical examination findings recorded. Skin scraping was done for superficial lesions such as those with superficial ulcers and ulcerated tumors. Tzanck smears were made for bullous lesions. [2] A fresh vesicle or bulla with no signs of secondary infection was selected for making the smears. After clearing the peripheral portion, the bulla was incised with scalpel and the roof of bulla reflected. The base of blister was scraped gently and material spread on a clean glass slide. Slit smears were made in cases suspicious of leprosy. [3] Fine needle aspiration cytology (FNAC) was done in cases of suspected malignant tumors. Excisional, incisional and punch biopsy were done to obtain tissues for histopathological examination. Cytological smears were fixed in 95% ethanol and stained with Papanicolaou stain (PaP). Sections were routinely stained with hematoxylin and eosin (H and E) and special stains (VanGieson, reticulin and Ziehl Neelsen stain etc.) were employed wherever required. Concordance rate between cytological and histopathological diagnosis was analyzed.

Results

Of the total 85 finally diagnosed cases, there were 41 cases of vesicobullous lesion, 24 neoplastic lesions and 20 granulomatous skin lesions. Histopathological confirmation was available in 60 cases.

Vesicobullous Lesions

Forty one cases of vesicobullous lesions comprised of three broad categories (A) viral infection (B) pemphigus (C) bullous impetigo.

(A) Viral infections

There were nine cases of herpes simplex (HS), 11 cases of herpes zoster (HZ) and five cases of molluscum contagiosum. Mean age was 22.5 years for HS and 38 years for HZ/ varicella zoster (VZ). Patients from both HS, HZ/VZ infections presented with painful vesiculobullous eruptions with an erythematous base of short duration (two to five days). Smears from patients of herpes showed ballooning degeneration, multi nucleated giant cells and bland inclusion containing

nuclei, molded against each other [Figure 1] and [Figure 2]. Distinction between HS and HZ was not significant in cytology. Biopsy from these cases showed intraepidermal bulla filled with serous fluid containing degenerating epithelial cells, ballooned cells and inclusion bodies. Five patients of molluscum contagiosum were observed. Discrete waxy papules with umblicated centers were seen on clinical examination. Cytology and histopathological examination showed

presence of squamous cells with basophilic or eosinophilic round, regular cytoplasmic inclusion bodies and nuclei pushed to the periphery.

(B) Pemphigus

In nine of the 12 cases of pemphigus, Tzanck smears revealed acantholytic cells lying singly and in small clusters [Figure 3] and [Figure 4]. Distinction between Pemphigus vulgaris and Pemphigus foliaceous was not significant in cytology. Histopathology revealed suprabasal intraepidermal blister in P.vulgaris (10 cases) and subcorneal intraepidermal blister in and P. foliaceous (two cases).

(C) Bullous impetigo

Four cases of bullous impetigo were included in the study, all seen in children. Cytological examination revealed acantholyic cells with a large number of

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acute inflammatory infiltrate [Figure 5]. Histopathological examination showed subcorneal pustules with superficial perivascular neutrophilic infiltrate.

Granulomatous Skin Lesions

Nine of the 20 cases were of lupus vulgaris. Clinically they presented with nodules and ulcerated plaques of long duration. Imprint smears in seven cases and histopathological examination showed epithelioid granulomas with or without caseation. Three of the seven cases showed positivity for acid fast bacilli (AFB). Eleven cases of leprosy were studied and cases were categorized according to the Ridley Jopling classification. [4] Of the 11 cases of leprosy, cytohistological correlation was available in 10; eight were concordant and two discordant. One case which showed features of borderline tuberculoid lepsory on cytological examination, exhibited feature of tuberculoid leprosy on histopathology. One case diagnosed as mid-borderline on cytology revealed feature of borderline leprosy on histological examination.

Benign Neoplastic Lesions

Six of the 24 neoplastic lesions studied were epidermal inclusion cysts .Clinically, patients presented with subcutaneous round nodules, soft to firms measuring one to three cms. On cytology, smears showed keratinous material with anucleate and nucleated squamous with cell debris in the background. Histology showed cysts lined by stratified squamous epithelial with a distinct granular layer and lumen filled with keratinous material.

Malignant Lesions

Of the 18 malignant lesions there were eight cases of squamous cell carcinoma (SCC), five of basal cell carcinoma (BCC), three of malignant melanoma and one case each of Merckel cell carcinoma (MCC) and sebaceous gland carcinoma (SGC) .Clinically the lesions were ulcerated nodular growth with crusting and indurated margins. Cytological and histopathological examination revealed malignant squamous cells with pleomorphic, hyperchromatic nucleus with clumped chromatin.

Five cases of BCC presented clinically with single or multiple nodules and ulcerated swelling either on the cheek, or eyelid or forehead. Imprint smears revealed cohesive sheets of round to oval tumor cells having basophilic cytoplasm, uniform dark, oval nuclei with evenly distributed chromatin. Histology revealed closely packed oval cells of uniform size with scant cytoplasm, hyperchromatic nuclei with peripheral palisading. Three cases of malignant melanoma were seen, all in males. Lesions showed mottled appearance due to pigmentation with irregular borders. Abundant melanin pigment was seen along with poorly cohesive, variably sized round cells with eccentric round to oval hyperchromatic nuclei on cytology. Histology revealed atypical spindle shaped melanocytes at right angles to the epidermal surface. Abundant melanin pigment was found.

A single case of MCC was seen in a 55-year-old male who presented with multiple nodules over the head and neck region. Cytology showed inadequate cellularity. Histological examination showed small round cells in the dermis in a diffuse pattern. The cells had scanty cytoplasm, round and vesicular nuclei with fine granular dusty chromatin and multiple nucleoli. A single case of sebaceous gland carcinoma was seen in a 40-year-old female who presented with a large mass on the back with multiple nodules having both solid and cystic areas. The lesion was infected and bloody discharge was present. Cytology showed clusters of epithelial cells with enlarged nucleus, prominent nucleolus and

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eosinophilic cytoplasm having clear droplets . Occasional mitotic figures were also seen. Histology revealed sheets and trabeculae of malignant cells in lower dermis with variation in size, eosinophilic and clear vacuolated cytoplasm with enlarged hyperchromatic nucleus, prominent nucleoli and abnormal mitotic figures.

Discussion :

In this study we extensively studied the role of cytology in early and quick diagnosis of various skin lesions with particular emphasis on vesicobullous lesions.

Vesicobullous Lesions

(a) Herpes Simplex / Zoster

Tzanck smears were prepared from fresh bullous lesions as described by Tzanck and his coworkers and Blank et al. [2], [5] The average age incidence of herpes simplex in our study was 22.5 years whereas Crumpacker reported peak years of incidence one to five years. [6] Cytological features observed were in accordance with the findings of Blank et al. [5] Graham et al. [7] Sehgal and Dube. [8] No differentiation was seen between herpes simplex and herpes zoster on cytological grounds. Findings of histological examination were similar to those seen on cytology in all the 9 cases i.e., concordant result were obtained in nine cases (100%). Distinction between the two types of herpes infection was made purely on clinical grounds.

(b) Pemphigus

Findings in pemphigus were similar to those observed by Sehgal and Dube and Naib. [8],[9] In three cases cytological examination was inadequate for diagnosis. Distinction between Pemphigus vulgaris and Pemphigus foliaceous was not possible on cytology. On histology, 10 cases were diagnosed as Pemphigus vulgaris and two cases as Pemphigus foliaceous. Findings on histopathological examination of Pemphigus vulgaris were similar to those seen by Camacho-

Alanso et al. [10] Histological findings of Pemphigus foliaceous were similar to those observed by Park et al. [11] Concordant results were obtained in all the nine cases (100%).

(c) Molluscum Contagiosum

In all the five cases of molluscum contagiosum, scrape smears were made. Cytology was inadequate in one case. Concordant result was seen in all the four cases (100%) similar to Patil et al. [12] who also showed cytology to be a rapid diagnostic method with high sensitivity and specificity.

(d) Bullous Impetigo

Tzanck smears were made in all four cases of bullous impetigo. Histological examination was done in three cases as one case refused biopsy. One case (33.3%) diagnosed as bullous impetigo showed features of herpes zoster on histological examination. The sensitivity of cytological examination (Tzanck smear) for the diagnosis of vesicobullous lesion was found to be as high as 96%. Histopathology, though a reliable and more specific means of diagnosis, had a lower patient compliance as 16 patients did not comply with a request for biopsy.

Granulomatous Lesions

(a) Lupus Vulgaris

In nine cases of lupus vulgaris the mean age of presentation was 22 years, though Bhambhani et al. [13] described maximum number of cases in the 0 - 10 years age group. Four cases were seen on the face, similar to Bhambani et al. [13] who also showed lesions on the face in 40% of cases, whereas the study by Savin [14] revealed 80% lesions on the face. Raghuveer et al. [15] in their study of 167 cases showed epithelioid granulomas without necrosis in

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17.4% cases, epithelioid granulomas with necrosis in 68.2% of the cases and necrosis alone in 14.4% cases. We also observed similar finding with well defined to ill defined granulomas with caseation in five cases (60%) and epithelioid granulomas without caseation in two cases (40%). Cytology was helpful in the diagnosis of seven cases of lupus vulgaris as two cases gave inconclusive results. Histopathological examination in all seven cases revealed features of lupus vulgaris (100% concordance).

(b) Leprosy

Eleven cases of leprosy were included in the study and categorized according to the Ridley Jopling classification. [4] The average age incidence in our study was 32.5 years. Farshcian and Kheirandish [16] showed the mean age of presentation to be 48.5 plus/minus 16.2 years. Out of 11 cases, cytological diagnosis of leprosy was made in eight cases, while one case was inconclusive. Histopatholoical examination confirmed the cytological diagnosis in eight of the 10 cases (80% concordance) whereas in two cases discordant results were seen. These two cases with cytological features of borderline tuberculoid (BT) and mid-borderline leprosy on histopathological examination showed features of tuberculoid leprosy and BT leprosy respectively. The sensitivity of cytology (slit smear, imprints, FNAC) for the diagnosis of various granulomatous lesions was found to be 88.2%. In the five cases, biopsy was not available.

Neoplastic Lesions

Of the six benign lesions studied, accurate cytological diagnosis was made in four, and two were hemorrhagic.Histopathological correlation was seen in all four cases (100% concordance). Observation was similar to those observed by Layfield and Glasgow. [17] SCC was the most common malignant skin lesion seen in our study comprising eight cases of a total of 18 malignant lesions studied. In all the eight cases the lesion was seen on the face similar to findings

reported by Dracopoulou et al. [18] and Allen. [19] Cytological correlation of the eight cases confirmed as SCC in seven and one case proved to be seven moderate dysplasia on histopathological examination (87.5% concordance). Lesions were seen on the face in all the cases of BCC in our study, similar to the findings by Allen and Malberger et al.

[19,20] Our cytological findings correlated with the observations of Layfield and Glasgow, [17] Malberger et al. [20] as well as Arya et al. [21] and Dey et al. [22] Correlation between cytology and histopathology was possible in three of four cases (75% concordance). The average age incidence of malignant melanoma was 52.75 years. It correlated with the study by Hadju and Savino [23] who reported maximum number of cases in age group of 41-60 years. However, this was in contradiction with the study of Perry et al. [24] which reported higher incidence of tumor in young adults. Scrape smears were made from the ulcerated lesions in two cases but were hemorrhagic. The presence of melanin pigment, intracellularly, in a large number of cells was the single most valuable morphological feature that favored the diagnosis of malignant melanoma. Histopathological examination showed features of malignant melanoma though correlation between cytology and histopathology was available in only one case. Intranuclear pseudo inclusions were not seen by us, though their presence was taken as supportive evidence in cases of hypomelanotic melanoma by Perry et al. [24] Most cases of MCC reported in literature are in the head and neck region.

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[25] We studied a single MCC case with nodules over the head and neck region. Cytological examination of the nodules in our case was hemorrhagic. Diagnosis
of MCC was made on histopathology. Findings on histology were similar to those described by Anderson et al. [26] Immnohistochemistry was positive for neurofilament and chromogranin markers. A rare case of SGC over the back was diagnosed in a 40-year-old female. Most of the cases of SGC reported in the literature are of the eyelid. The cytological findings observed in the smear were similar to those described by Gao et al. [27] and Malhotra et al. [28] Diagnosis on cytology was confirmed by histopathology. The sensitivity of cytology (FNAC) in diagnosing various neoplastic lesions came out to be 88.9%. The study was conducted to correlate the clinical diagnosis with cytology and histopathology for the diagnosis of various non-neoplastic and neoplastic lesions. A total of 85 cases of clinically diagnosed skin lesions were studied.

Cytohistological correlation was available in 60 of 85 cases. In 55 cases (91.7%) an accurate diagnosis was made by cytology whereas in five cases (8.3%) discordant results were seen thus a considerably high degree of correlation was achieved among cytological and histological modalities of diagnosis, especially in vesicobullous lesions. The sensitivity of cytology in diagnosing vesicobullous lesions, granulomatous lesions and neoplastic lesions was 96, 88.2 and 88.9% respectively. It was seen that biopsy provided complete tissue details for accurate diagnosis of skin lesions; however, diagnosis takes a longer time as compared to the early diagnosis provided by cytology. Yet it may not be readily available, as is evident from this study, where 25 cases (29.4%) of skin lesion did not comply with a request for biopsy.

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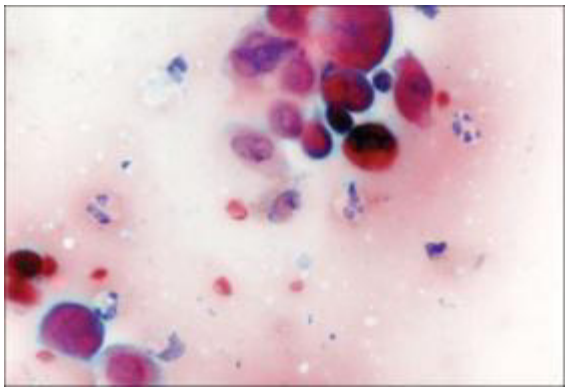

Figure 1 : Herpes Simplex (Tzanck smear): Mono and multi nucleate giant cells with bland inclusion containing nuclei. Molding is absent in the smear (Pap, X400)

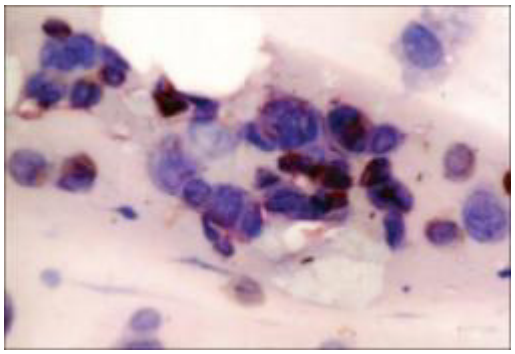

Figure 2 : Herpes Zoster (Tzanck smear): Multi nucleated giant cells with bland inclusion containing nuclei against each other with variable amount of cytoplasm (Pap, X400)

Figure 3 : Pemphigus Vulgaris – Ruptured bullae with erosion and crusting

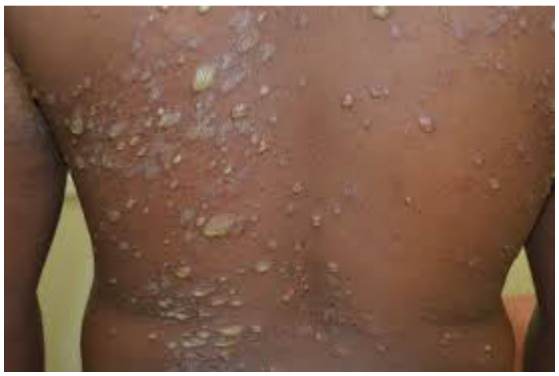

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Figure 4 :Pemphigus (Tzanck smear): Clusters of acantholytic cells with round hyperchromatic nucleus and darker staining peripheral cytoplasm (Pap, X400)

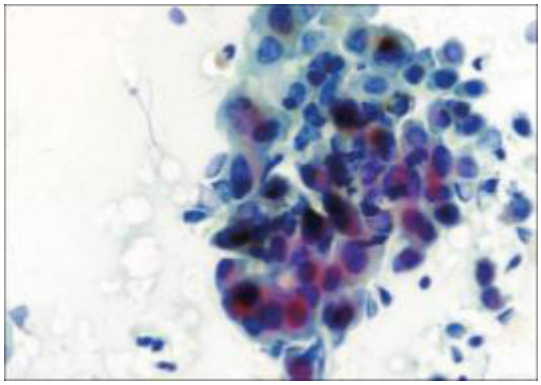
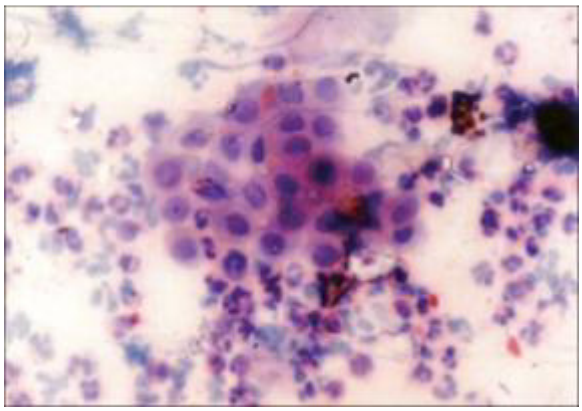


Figure 5 :Bullous Impetigo (Tzanck smear): Acantholytic cells with round hyperchromatic nucleus and plenty of neutrophils (Pap, X400)



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