

CYTO-HISTOPATHOLOGICAL CORRELATION OF PREMALIGNANT AND MALIGNANT LESIONS OF THE ORAL CAVITY IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Objectives: Globally, oral cancer seems to be the sixth-utmost common cancer. Oral cancer is the chief among entire malignant tumors of men in the Indian subcontinent and squamous cell carcinoma is the commonest among all oral malignancies. Hence, in the present study, the utility of aspiration and scrape cytology in the screening and initial diagnosis of oral lesions, to confirm by histopathology, were studied.

Methods: The enrolled patients underwent complete clinical history, examination, fine-needle aspiration cytology (FNAC), scrape cytological assessment, and histopathological observations at VIMSAR, Odisha. Cytological smears were stained with Diff-Quik and Papanicolaou stains. Cytological observations were interrelated with concluding histopathological identification.

Results: There were 32 premalignant and 60 malignant lesions of the oral cavity. Overall 69.56% diagnostic accuracy of scrape cytology for numerous intraoral premalignant and malignant lesions was found. The most common site for premalignant lesions was in the buccal mucosa, and malignancy was the tongue. Among the malignant lesion, squamous cell carcinoma was major, constituting 86.66% of cases, and most of the patients were in the fifth decade. Risk factor, mostly tobacco exposure, was associated with 66.66% of cases of a malignant lesion.

Conclusion: FNAC and scrape cytology were observed as precise convenient, simple, and economical in the initial diagnosis of pre-cancerous and cancerous lesions of the oral cavity.

Keywords: Fine-needle aspiration cytology, Oral cavity, Scrape cytology, Squamous cell carcinoma.

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INTRODUCTION

In India, oral cancers are the most common cancers, with an age-unvarying incidence of 12.6/100,000 population. Oral cancers are one of the foremost cancers in Indian males accounting for nearly 30% of cancer encumbrance. It is estimated that almost one million new cases are being identified yearly in the Indian subcontinent. A pre-cancerous lesion is a morphologically transformed tissue, in which oral cancer is further likely to happen than in its normal counterpart, for example, leukoplakia and erythroplakia. As per the World Health Organization, lesions and circumstances of the oral mucosa, which may experience malignant alteration, are well-defined as potentially malignant disorder. Leukoplakia is the most common potentially malignant lesion within the oral epithelium. The worldwide incidence of leukoplakias is at 2% [1]. The average age of people with oral leukoplakias is 60 years old and is close to the average age of patients with oral cancer. A sub-type of verrucous leukoplakias is proliferative leukoplakia, which is categorized by multi-focal lesions, resistance to therapy and frequent, i.e., 70% conversion into oral squamous cell carcinoma [2]. Erythroplakia is not as common as leukoplakia and has an incidence documented among 0.02% and 0.83% [3]. These lesions are referred to as hazardous oral mucosa as it characteristically presents as carcinoma in situ, severe epithelial dysplasia, or superficially invasive carcinoma when observed through the microscope. Oral submucous fibrosis (OSMF) is a long-lasting illness categorized by fibrosis of the lining mucosa of the oral cavity.

Aspects comprising areca nut chewing, ingestion of chilies, genetic and immunologic procedures, nutritional insufficiencies, and other factors and influences have been assimilated in connection with OSMF. In India, who approaching the health centers, 0.2–1.2% of the urban population were found to be affected with this [4]. There occurs an optimistic link between the frequency of leukoplakia and oral cancer with OSMF.

Squamous cell carcinoma in its early stage frequently occurs as a white patch, i.e., leukoplakia, inflamed patch, erythroplakia or an assorted inflamed and white lesion, i.e., erythroleukoplakia. By this period, superficial ulceration of the mucosal surface will progress. When this lesion propagates, it develops an exophytic growth with a fungating and/or papillary surface. Further tumors have an endophytic progression that is categorized by a depressed ulcerated surface with an elevated, rolled boundary [5]. Verrucous carcinoma is low-grade variant of oral squamous cell carcinoma and encompasses around 3% of total primary invasive carcinomas of the oral mucosa [6]. Verrucous carcinoma presents as diffused, solidified plaque or mass with a warty and/or papillary surface and is typically white. Verrucous carcinoma is slowly developing, exophytic as well as well differentiated: it is allied with a better prediction than conventional squamous cell carcinoma of the mouth [7].

Scrape cytology and aspiration cytology are valued techniques for the preliminary assessment of intra-oral lesions as they can simply discriminate inflammatory from neoplastic lesions and benign from malignant neoplasm. These tests are modest, harmless, prompt, consistent, painless, economical, comparatively non-invasive, and stress-free techniques, which can be done as an outpatient department process deprived of any superior kit or preparation. Scrape cytology has increased acceptance as a prompt, economic, comparatively effortless, specific, and operative concern to diagnose the intra-oral lesion. Hence, in the present study, an attempt was made to assess the effectiveness, and dependability of scrape cytology and fine needle aspiration cytology (FNAC) in the analysis of oral lesions.

METHODS

The proposed research was initiated at VSS Institute of Medical Science and Research, Burla, Odisha, India, The study was continued for a

duration of two consistent years from September 2018 to August 2020. Institutional ethical committee approval has been obtained proceeding to the experiment initiation. Entire cases reporting with problems associated with the oral cavity, like whitish or reddish patches, ulcerative lesions, or a growth, were included in this study.

Inclusion criteria

This study included patients of any clinically suspected cases of premalignant and malignant lesions of the oral cavity, any case with the lesion of the oral cavity having a history of risk factors for oral cavity malignancy, like addiction to smoking, alcohol or betel nut chewing and any new case, cytologically diagnosed with premalignant and malignant lesion of the oral cavity.

Exclusion criteria

Patients with previously diagnosed/treated cases of malignancy of the oral cavity and any case with the history of exposure to radiation for any head-and-neck tumor.

All patients were required to provide written informed consent for this study. After detailing all study procedures to the patients, written informed consent was collected from every participant. Proceeding to the procedure, detailed clinical history, including history of addiction, was collected, followed by a proper examination of the oral cavity was done in each case. A light source and tongue depressor are also used to observe lesions when required. Patients were asked to wash their oral cavity scrupulously with water before proceeding to sample collection. All the FNAC and scrape cytology were performed by cytopathologist. Supine/sitting postures with head support, all the samples were collected. Scrape smears were done by wooden Ayer's spatula from pathological sites, frequently from mucosal lesions and from other representative sites. For fine-needle aspiration, after localizing the lesion. A 23–24G needle attached to 10 ml disposable plastic syringe was used. Aspirated material was spread on the clean numbered slides. Wet smears were fixed in 95% ethanol for Papanicolaou Stains, although air-dried smears were stained with Romanowsky (Diff-Quik) stain. All the malignant or dysplastic, or clinically suspicious lesions were recommended to histopathological study. Biopsy tissues from lesions were subjected to histotechnical process; slides were stained with hematoxylin and eosin for microscopic examination. Cytological outcomes were then interrelated with observed histopathological diagnosis.

Statistical analysis

Statistical analysis was executed by SPSS software, and the variables have been denoted in mean value with standard deviation, and categorical data were denoted in percentage.

RESULTS

One hundred and twenty-eight cases presenting with different lesions of the oral cavity have been exposed for fine-needle aspiration cytology, scrape cytology, and histopathological study. Ten cases were withdrawn from the study due to insignificant correlation or unavailability of data, leaving 118 cases for cyto-histopathological correlation. The study population was segregated into 78 (66.10%) males and 40 (33.89%) females. The male outnumbers the female by the ratio being 1.9:1. The enrolled patients were largely from the group of 41–50 years with 44 (37.28%) cases, followed by 51–60 years with 28 (23.72%) cases (Table 1).

All 118 patients were categorized into three foremost categories, benign lesions 26 (22.03%) cases, premalignant lesions 32 (27.12%) patients, and malignant lesions 60 (50.85%) cases depending on concluding histopathological interpretation.

Samples taken from different sites by aspiration or scraping of oral cavity lesions showed the maximum number of cases 48 (40.67%) were from the tongue, followed by 42 (35.59%) cases from buccal mucosa, 12 (10.16%) cases from the gingiva, 06 (5.08%) cases from the palate

and 10 (8.40%) cases from others site (Fig. 1a-c). Buccal mucosa was the furthestmost predominant location for pre-malignant, and the tongue seems to be the common location in malignant lesions.

A total of 32 patients with pre-malignant lesions were comprised in this report, with the most common type being dysplasia 12 (37.5%) cases, followed by SMF 10 (31.25%) cases. (Table 2) The highest number of premalignant lesions were established in the buccal mucosa followed by the tongue. Out of 32 cases of premalignant lesions, 26 cases were correctly diagnosed by cytology with a diagnostic accuracy of 81.25%. Four patients' dysplasia was diagnosed as squamous cell carcinoma and 2 patients' dysplasia was diagnosed as the inflammatory lesion, with a false-negative rate of 18.75 by cytology. The cytological identification was precise in 8 out of 8 cases of leukoplakia, 10 out of 10 cases of SMF and 02 out of 02 cases of erythroplakia with a diagnostic accuracy of 100%. Statistical evaluation of cytology study regarding premalignant lesion as per Table 3 stands a sensitivity of 81.25%, a specificity of 67.44%, a false negativity of 18.75%, a false positive of 32.55%, a positive predictive value of 48.14%, a negative predictive value of 90.62%.

Among 60 patients of malignant lesions, squamous cell carcinoma (Figs. 2a and b, 3b) forms the common malignancy in 52 (86.66%) patients, followed by verrucous carcinoma (Fig. 3a) 08 (13.33%) cases. (Table 2) The tongue was the furthestmost common location for malignant lesions followed by buccal mucosa. From the total 60 cases of malignant lesions, 38 cases were diagnosed correctly by cytology with a diagnostic accuracy of 63.33%. Eight patients of squamous cell carcinoma were diagnosed with dysplasia, 6 patients of squamous cell carcinoma and 8 patients of verrucous carcinoma was diagnosed as inflammatory lesion on cytology, with false-negative rate being 36.66%.

Table 1: Distribution of age and sex among the patients

Age (years)	Male (%)	Female (%)	Total (%)
21–30	12 (10.16)	06 (05.08)	18 (15.25)
31–40	14 (11.86)	08 (06.77)	22 (18.64)
41–50	30 (25.42)	14 (11.86)	44 (37.28)
51–60	18 (15.25)	10 (8.47)	28 (23.72)
>60	04 (3.38)	02 (1.69)	06 (05.08)
Total	78 (66.11)	40 (33.89)	118 (100)

Table 2: Distribution of premalignant and malignant diseases (n=92)

Group	Diseases	Number (%)
Premalignant (n=32)	Leukoplakia	08 (25)
	Erythroplakia	02 (6.25)
	SMF	10 (31.25)
	Dysplasia	12 (37.50)
	Total	32 (100)
Malignant (n=60)	Squamous cell carcinoma	52 (86.66)
	Verrucous carcinoma	08 (13.33)
	Total	60 (100)

SMF: Submucous fibrosis

Table 3: Evaluation of cytological diagnosis regarding pre-malignant and malignant lesions

Group	Cytology	Histology (Final diagnosis)	
		Positive	Negative
Premalignant lesion	Positive	54	28
	Negative	64	58
	Total	118	86
Malignant lesion	Positive	44	06
	Negative	74	52
	Total	118	58



Fig. 1: (a) 43 years male with leukoplakia over buccal mucosa and lower lip. (b) 48 years male with ulceroproliferative growth over the right lateral border tongue. (c) 57 years female with ulcerative growth over the soft palate

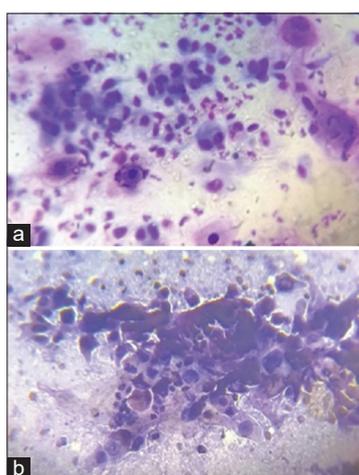


Fig. 2: (a) Scrape cytology reveals malignant squamous epithelial cells (Diff-Quik Stain: $\times 400$). (b) Scrape cytology reveals malignant squamous epithelial cells (Papanicolaou Stain: $\times 400$)

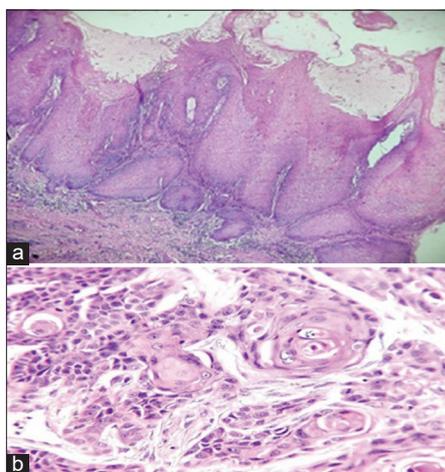


Fig. 3: (a) Histopathological section showing feature of verrucous carcinoma. (H and E Stain: $\times 400$). (b) Histopathological section showing features of squamous cell carcinoma. (H and E stain: $\times 400$)

Statistical evaluation of cytology study regarding malignant lesion as per Table 3 stands a sensitivity of 63.33%, a specificity of 89.65%, a false

negativity of 36.66%, a false positive of 10.34%, a positive predictive value of 86.36%, a negative predictive value 70.27%.

Therefore, among 128 patients of oral lesions, cytological and histopathological correlation was obtainable in 118 patients. Out of total of 92 patients with premalignant and malignant lesions, concordant results were observed in 64 patients with an overall analytical accuracy of 69.56%. Among 92 significant cases, i.e., premalignant and malignant lesions, 54 numbers of cases having a history of tobacco exposure. Out of the total of 32 cases of pre-malignant lesions, 14 patients had a history of tobacco exposure (43.75%). 40 (66.66%) patients had tobacco exposure 60 cases in the malignant category. The history of tobacco exposure was more in cases of malignant lesions as compared to pre-malignant lesion.

DISCUSSION

The current study was assumed with the interpretation of the assessment of premalignant and malignant lesions in the oral cavity. Scrape cytology and aspiration cytology of these lesions were correlated with histopathology to get the efficacy of this method. Although, biopsy seems to be the gold standard technique in identifying oral cavity lesions, it is not possible to conduct in each case as particularly with asymptomatic lesions; some cases may be medically compromised, and few may not give their consent for biopsy.

The patient parameters, including sex-based incidence, age, and tobacco addiction history, were studied in patients with oral cavity lesions, and the respective findings were compared with published results. In our study maximum number of patients premalignant and malignant lesions of the oral cavity as in 5th and 6th eras of life, which was associated with Bhandari and Gadkari observation, whereas Agrawal and Chouhan they established peak occurrence was in the fourth decade [8,9]. In the present study, the higher prevalence was in men than women, with a ratio of 1.9:1, similar to another study [10]. Buccal mucosa was the common location for premalignant lesion, whereas malignancy was most commonly found in the tongue. Babshet *et al.* found buccal mucosa was as the predominant location of the contribution of malignancy [11].

Among these 92 patients with oral cavity lesion. 32 cases were premalignant lesion. Leukoplakia accounted for 25% of lesion. It has been reported as the most common potential malignant disease in another study [12]. The global prevalence of leukoplakia is 2.6% [13]. Leukoplakia was further predominant in men than women in the existing study. We have two cases of erythroplakia. The report observed that 90% of erythroplakia lesion is histopathologically either severe dysplasia or superficial invasive squamous cell carcinoma [14]. Rendering to numerous reports, OSMF occurrence is 0.03–3.2% in India, with a malignant transformation rate of 7.6% [15]. In our study prevalence of OSMF was 31.25%, which was much higher than the study of Panda *et al.* [16]. Dysplasia was observed in our study was 37.50%. Severe dysplasia or carcinoma *in situ* is pre-cancerous lesion, although other believe that the lesion is an actual carcinoma revealed before invasion [17].

Out of 92 patients, 60 patients presented malignancy. More malignant lesions were found as most of the cases with initial cancerous lesions, which were usually unproblematic, and approached the hospital when they noticed the ulcer or growth. Many patients from the rural part of this area were referred for diagnosis. Carcinoma of the oral cavity is a common malignancy of head and neck areas, amid which squamous cell carcinoma is most common one [18]. In malignant lesions, males are affected more due to the possibility of addiction to tobacco, smoking, and alcohol. Most malignant patients were in the fifth and sixth eras of lifespan correlated with study of Modi *et al.* [19]. Verrucous carcinoma seems to be a warty squamous cell carcinoma characterized by exophytic growth of keratinizing epithelium [20]. Scrape cytology or superficial biopsy may show a false negative which

shows the typical pattern of verrucous carcinoma on an adequate and deeper biopsy [21].

Surgical biopsy is the old technique for assessing numerous intraoral lesions. Over the years, scrape cytology and aspiration cytology has been found to be beneficial, simple, economical, and precise in evaluating and identifying numerous neoplastic as well as non-neoplastic lesions of various human locations as well as oral cavity lesions. Scrape cytology shows a significant part in analyzing the intraoral lesions presenting as an ulcer and/or proliferative growth. In this study, all 118 patients were generally categorized into three chief groupings, benign lesions 26 patients, pre-malignant or potentially malignant lesions 32 patients and malignant lesions, 60 patients considering the ultimate histopathological results. Cyto-histopathological correlation was available in 32 patients with pre-malignant lesions. A concordant result was occurred in 26 patients giving analytical accurateness of 81.25%, which was higher than as documented by Seetharam and Ramachandran [22]. The false-positive rate of pre-malignant lesion by cytology was 32.55% and false-negative rate of 18.75%.

Malignant lesions cyto-histopathological correlaton seems accessible among 60 cases. Concordant observation was found in 38 patients reporting diagnostic accurateness of 63.33%, which was comparatively lower than the values of 76.49% as reported by Domanski and Akerman [23]. In this study, we found 22 false negative cases regarding malignant lesions. Eight patients of squamous cell carcinoma were diagnosed as moderate-to-severe dysplasia and six patients of squamous cell carcinoma and eight patients of verrucous carcinoma were diagnosed as inflammatory lesion on cytology with false-negative rate being 36.66%. Out of the total of 44 cases of malignant lesions diagnosed by cytology, 6 false-positive patients of squamous cell carcinoma turned out as 2 patients of carcinoma *in situ*, 2 patients of moderate dysplasia and 2 patients of inflammatory lesion on histopathology with false-positive rate of cytology being 10.34%. Therefore, the present results show the rates of false negatives and false positives both are slightly higher than in other study [24,25]. The possible cause behind this may be due to the smaller exposed area, difficulty in opening the mouth for trismus, which causes the inability to collect atypical cells for cyto-diagnosis. In addition, with well-differentiated squamous cell carcinoma and verrucous carcinoma lacking strong atypia causes false negatives. Overall 69.56% diagnostic accuracy of scrape cytology and aspiration cytology for various intraoral premalignant and malignant lesions were obtained in this current observation, which was comparatively lower than the value by other studies [10,26].

CONCLUSION

This study greatly endorses scrape cytology as an appreciated and useful technique for the preliminary screening of all intraoral lesions as it is an objectively delicate and precise technique, a simple procedure, economical and comfortable to all patients, and in addition, this technique offers a prompt and accurate diagnosis. However, to detect invasion in squamous cell carcinoma, for dysplastic premalignant lesions and clinically suspicious lesions, histopathological examination is required for definitive diagnosis. This offers effective treatment plans together in preventing further complications.

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AUTHORSHIP CONTRIBUTIONS

Dr. Ghanashyam Meher and Dr. Pabitra Hembram— Design and Data collection or processing, editing the manuscript. Dr. Kshetra Mohan Tudu and Dr. Binod Kumar Sahu - analysis or interpretation, literature search, manuscript writing, and submission.

CONFLICT OF INTEREST

Nil.

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