

Chapter 2

Historical Development of Oral Cytology

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Introduction

The seminal work by Papanicolaou and Traut in studying the cells from precancerous and cancerous lesions of the cervical mucosa paved the way for oral cytology. Their work proved an effective tool for screening gynecologic malignant disease [1]. Initially, the use of oral cytology was limited to comparative studies of oral and cervical cytology, describing cytomorphological changes depending on the menstrual cycle. Interestingly, buccal smears also have been reported to show marked cyclic fluctuation in keratinization during the normal menstrual cycle [2–4].

In the second half of the nineteenth century, the morphology of malignant cells in sputum was first described from an oropharyngeal carcinoma [5]. This was followed by work of Morrison et al. in which they used Papanicolaou staining to diagnose nasopharyngeal carcinomas [6]. But it was Montgomery and von Haam who were the first to examine the usefulness of cytology in the oral cavity [7]. Some further studies showed the application of oral cytology by animal experiments [8, 9]. Oral cytology has been used effectively as a research tool for the evaluation of experimentally induced carcinomas in hamster cheek pouches and in healing of gingivectomy wounds [10]. Much later, Sandler, through his series of reports on Veterans Administration studies of oral cytology, focused attention on the potential of the technique as an aid to early detection of oral cancer [11]. These landmarks in the development of oral exfoliative cytology are listed in Table 2.1.

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Table 2.1 Landmarks in the development of oral exfoliative cytology

Year of publication	Author	Subject of publication
1860	Beale [5]	Cytological examination of sputum in a case of pharyngeal carcinoma
1940	Weinmann [12]	Cytological examination of oral cellular keratinisation
1941	Ziskin et al. [13]	Effects of the menstrual cycle on oral cellular morphology
1942	Papanicolaou [14]	Introduction of a staining procedure for cytological smears
1943	Papanicolaou and Traut [3]	Cytological diagnosis of uterine cancer
1949	Morrison et al. [6]	Cytological diagnosis of nasopharyngeal malignancies

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Cytology Techniques and Their Modifications

Over a period of time, as the field of oral cytology started to grow, many investigators including Montgomery and von Haam experienced the limitations of oral cytology and therefore felt the need for improvements. They devised numerous modifications which were intended to procure larger amount of cells, to sample a large cellular area and also to improve the quality of cell staining. However these modifications were not put to widespread use. Special stains have been advocated, either to define the best area for cell collection in a diffuse lesion or to enhance the definition of malignant cells for the examining pathologist. Some of these have been listed in Table 2.2. In order to reach the cells of the basal and parabasal layers, the atypical keratotic cell layers need to be removed. It is for this very purpose, the use of a metal spatula or a sharp spoon was recommended by some authors [20, 21].

Besides these techniques, numerous supportive analytical methods for light microscopy were used. Possible utilization of fluorescence microscopy and phase contrast microscopy was investigated. Fluorescent DNA-specific dyes like Acridine Orange were used to measure the cellular DNA content [22]. Analysis of nucleolar size and diameter, as additional parameters for malignancy, was carried out with the help of image cytometry [23]. Besides the classical applications of the oral cytological studies, detection of Epstein–Barr virus in oral lesions of hairy leukoplakia has also been done, thereby widening its possibilities [24].

Oral Cytology in Cancer Detection

Oral cytology appeared to be a promising diagnostic tool as it was thought to have potential for early detection of malignant lesions. The issue of whether oral cytology could be applicable for mass population screening is somewhat unsettled,

Table 2.2 Methodical modifications of oral exfoliative cytology

Year of publication	Author	Modification in material and methods
1951	Gladstone [15]	Improved quantities of obtained cells by use of a “sponge biopsy”
1952	Schneider [16]	Modifications of staining
1960	Cawson [17]	Modifications of staining
1963	King [18]	Use of frosted glass slides
1963	Staats and Goldsby [19]	Comparison of wooden and metal spatula. Recommendation of the metal spatula
1964	Sandler [20]	Removal of keratotic layers with a sharp curette
1981	Dumbach et al. [21]	Smear curettage’. Inclusion of deeper cell layers by use of a curette

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although the majority opinion seems to be that it was not practical at that time. Thus, investigators came up with numerous modifications in the techniques, some of which have been described above. These modifications did increase the sensitivity of cytology. However the main advantage of cytology was lost as compared with surgical biopsy as these kinds of scrapings were more invasive.

At the same time, oral cytology could not prove to be an effective tool for detecting early neoplastic lesion of the oral cavity in the way cervical cytology did. The reasons for this were manifold. First, oral topography and the size of the oral cavity made it virtually an impossible task to examine the complete mucosal surface. Thus, only the region which had visible lesions could be cytologically examined. It cannot be overemphasized that an adequate sample is essential if morphological evaluation of the collected cells is to yield representative findings. Even for histopathological examination, if a carcinoma covers a large area, it is important to carefully select the most appropriate site of the scalpel biopsy [25].

Secondly, a definite transformation zone as on the cervical mucosa, where malignant cells reach the epithelial surface in early tumor stages, does not exist in oral cancer [26]. Therefore, it was only possible to obtain malignant cells by conventional smears, if the carcinoma was fairly advanced or ulcerated [20]. Thus none of the existing minimally invasive techniques were able to sample deeper layers of the oral cavity—access to which was necessary to make oral cytology an effective preventive tool for oral cancers.

Conventional oral brush cytology has sensitivity ranging between 79% and 97% and specificity between 95.1% and 99.5%. Though there is an increase in the accuracy but this have not significantly increased compared with conventional exfoliative cytology. Thus, the need to improve analytics, which utilize technology like image analysis and automated machines, remains a challenge for the investigators.

Oral Brush

The introduction of the oral brush is a significant landmark in the history of oral cytology. Use of a brush for cervical cytology demonstrated better cell spreading on objective slides as well as an improvement in quality and validity of smears compared with smears obtained by using a wooden spatula [27]. But it is the ability of the oral brush to sample deeper mucosal layers, the site for squamous intra-epithelial lesions (SIL), with minimal invasion that is the principal basis for its success in oral cytology. [28]. It is a more convenient instrument, for the examiner, than the wooden tongue depressor, when dealing with oral lesions [26]. Moreover, this technique is a chair-side, easy to perform, painless test that can be used to evaluate any suspicious lesion, including common small white and red oral lesions, to rule out dysplasia.

A multicenter study emphasized the importance of brush biopsy with automated imaging in which clinically benign-appearing mucosal lesions were sampled using this technique and nearly 5% were later confirmed by using scalpel biopsy to represent dysplastic epithelial changes or invasive cancer [29]. Many other authors demonstrated that brush cytology could uncover similar lesions that were not clinically suspicious of carcinoma or pre-invasive disease [30]. Interestingly, a case report published a few years back emphasized the value of brush cytology in the follow-up of oral lesions which covered a large area [31]. In another report, Gupta et al. combined conventional oral brush cytology with the application of toluidine blue to localize the right site for brushing in suspected mucosal areas [32]. Mehrotra et al., in his study recently confirmed the utility of automated analysis in minimally suspicious (Class II) lesions [33].

Conclusions

Oral cytology has come a long way from its primitive Papanicolaou days. It has made major strides in its eventful development. The oral health professionals along with multidisciplinary scientific investigators have played a pivotal role in discovering optimal methods that would allow early diagnosis of oral cancer. Early detection should lead to less damage from cancer therapy and better prognosis. Surgical excision followed by a histopathological diagnosis, however, remains the widely accepted benchmark for the present. There are a number of recently introduced novel techniques that help in the diagnosis of oral malignancy. The future of these newer techniques appears extremely promising and should, hopefully, change the paradigm of oral cancer diagnostics.

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