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Stereomicroscope in Enhancing the Skill of Grossing Pathological Specimen

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Abstract: ***Introduction: Grossing refers to the examination of surgically excised specimen with bare eyes and dissecting the same in order to prepare the tissues for processing and embedding. It is an initial and the most important step in surgical pathology for reaching at an accurate diagnosis. Error in the step of grossing and later in embedding the proper tissue surface, especially of a small surgically excised tissue, may lead to diagnostic dilemma and ultimately delayed reporting. Stereomicroscope is an important tool for low magnification identification of any sample. Stereomicroscopic examination of a surgically excised specimen not only provides vital clues regarding the specimen, but also aid in proper orientation of the tissue for further examination. Objective: This study aimed to evaluate the efficacy of stereomicroscope in grossing and proper orientation of small surgically excised tissue samples. Material and Methods: Biopsy samples of size approximately 5x5 mm in greatest diameter were examined under stereomicroscope before and after grossing and processing, images were captured and oriented for histopathological examination. Result: The stereomicroscopic images of the biopsy samples aid to understand the nature and cut surfaces better than naked eye examination and helped in accurate orientation of the tissue surface. Conclusion: Regular use of stereomicroscope in routine laboratory procedure not only aids in analyzing the surface topography of the biopsy specimens better, but also acts as an effective way of embedding the tissues for further histopathological examination.***

Keywords: Stereomicroscope, Grossing, Histopathological diagnosis

1. Introduction

Grossing is an initial and most important step in surgical pathology. It refers to the examination of surgically excised specimen with bare eyes and dissecting the same in order to prepare the tissues for processing and embedding.[1]It reveals size, shape, surface and nature of the tissue specimen which act as a bridge between the clinical presentation of the lesion to the microscopic diagnosis by a pathologist. However, beside a pathologist, a resident, physician assistant, histotechnologist or a biomedical scientist can also perform the grossing.[2]

Error in the step of grossing and later embedding the proper tissue surface, especially of a small surgically excised tissue, may lead to diagnostic dilemma.[3] Accurate gross description and observation of the pathology specimen can give many clues to aid in the final diagnosis.[2] Selection of the tissue proper from the whole specimen, marking the areas on tissue, if needed, followed by embedding the proper tissue surface are the most crucial steps during grossing, as improper grossing will lead to lose of the vital clues forever. However, in case of a small surgically excised specimen this step is less significant,[4] hence many lead to improper orientation and inappropriate diagnosis.

With the advancement in technology, many adjuvant aids have come to facilitate the step of grossing, which include photographic facility, X-ray unit with view box, balances,[1] and digital photography.

Binocular stereoscopic microscopes have been used in dental technology for decades to improve visual acuity during mainly the fabrication processes like inspection of the impression, trimming and marking of the margin on the die, waxing of the margin, fitting of the initial restoration, final fitting of the restoration, including the addition of material and adjustments, final polishing of the marginal third of the restoration.[4]

Interestingly, the use of stereomicroscope during the grossing procedure, not only enhanced the ability to inspect the nature and surface of the tissue specimen better, but also acts as an adjuvant aid to guide the proper cut surface for embedding and further histopathological examination.With the help of stereomicroscopic images of the specimen, the nature of the tissue can be explored to an extent, that it will help to achieve the final diagnosis accurately.
Hence, this study aimed at evaluating the efficacy of stereomicroscope in grossing and proper orientation of surgically excised tissue samples.

2. Material and Methods

The biopsy samples of the patients reported in the Department of Oral Pathology and Microbiology, Government Dental college and Research Institute, Bengaluru, from July 2019 to December 2019 were examined before and after grossing under Magnus Trinocular Stereo Zoom Research Microscope Model-MSZTr, images were captured and the tissues were oriented for histopathological examination.The H and E stained sections were compared with the stereomicroscopic images.

3. Result

Case 1:
After an intraoral excision biopsy, we received a single bit of tissue measuring approximately 0.5x0.5cm in diameter with an irregular surface, whitish colour and firm consistency (fig 1a). Usually such small tissue bits are more prone for improper grossing followed by tangential orientation. Stereomicroscopic examination of the tissue revealed irregular whitish papillary projections with thin brownish extensions in the middle suggestive of papilloma (fig 1b). With the guidance of steromicroscopic appearance, the tissue was grossed and oriented for histopathological examination. The microscopic examination of the H & E stained section (fig 1c) showed numerous finger-like projection ofstratified squamous epithelium with hyperkeratosis and thin connective tissue core supporting the epithelium, confirming the diagnosis of papilloma.

On the other hand, another tissue bit with similar grossing details (fig 1d) was oriented for histopathological procedures without stereomicroscopic examination. On microscopic examination, numerous tangentially cut sections were seen (fig 1e), suggestive of improper grossing of the tissue bit followed by improper orientation.

Case 2:
A single bit of tissue measuring approximately 0.5x0.5cm in diameter with irregular surface, brownish white in colour and firm consistency (fig 2a). Though the tissue bit appeared typical macroscopically, stereomicroscopic examination showed multiple forceps indentation over the tissue (fig 2b). Interestingly, the same areas under research microscope revealed cyst like spaces surrounded by layers of epithelium giving a pseudocyst like appearance. However, comparing those areas with the stereomicroscopic images of forceps indentation, the diagnostic dilemma was avoided.

Case 3:
A single bit of tissue measuring approximately 1x1cm in diameter with a regular round surface, brown colour and firm consistency (fig 3a, 3b, 3c). The surface examination revealed a punctate like feature and with the help of stereomicroscope the cut was given so that the punctate comes in the cut surface(fig 3d, 3e, 3f). Following which the microscopic examination of the H & E stained section showed a hair follicle like structure withplenty of acute inflammatory cells around it. (fig 3g)

4. Discussion

"Grossing"- a term that refers to examination and dissection of surgical specimens, along with preparation of sections from those tissues requiring processing, is the initial step in surgical pathology dissection.[6] Gross tissue evaluation of a pathology specimen forms an indispensable, but often neglected, component for complete pathologic evaluation. It is in the time of grossing the pathologist gets vital clue about the specimen. Moreover, improper identification of the proper tissue, improper handling of the tissue or a surface orientation after grossingwill ultimately lead to a hard time during reporting. All this factors make the grossing a crucial step in surgical pathology.

Hence to improve the efficiency of grossing adjuvant aids like photographs, radiographs etc. are used in recent days.

Stereomicroscope which is an optical instrument for low magnification identification of any sample was introduced in the field of dentistry a decade ago. But the use of the instrument was limited in the fabrication procedures. Use of this instrument in histopathology, especially during the step of grossing, not only enhanced the nature of the tissue but also aided in understanding features like proper cut surface, possible origin and even an artefact to avoid misdiagnosis.

As in case 1, under stereomicroscope we could see the tiny projections of the tissue which was barely noted with naked eyes. In papilloma there are multiple projections which are not of same length. Such projections often mislead the gross resulting in improper orientation of the tissue. If the cut is given in cross section, high chances of getting multiple bits and also tangential cut. Whereas if the base is observed clearly and cut in the centre of the bigger projection longitudinally we can avoid these problems. With the help of stereomicroscope we were able to achieve a proper projections of epithelium and connective tissue on the histopathological section which confirms the diagnosis of papilloma.
Case 2 gave us a picture of forceps indentation on the tissue under stereomicroscope which was completely unnoticed with bare eyes.Those areas in the histopathological sections characteristically showed cyst like spaces in the epithelium. This finding could definitely lead us to dilemma without a knowledge of the artefacts over the tissue, rather without a stereomicroscopic picture of the same.

Case 3 showed that even before reaching to a research microscope a pathologist can gain an idea about the nature or origin of the tissue by stereomicroscopic examination. Without stereomicroscope there was a high chance of overlooking the punctate like feature in the specimen and grossing it in a wrong plane,leading to a dilemma while reporting.

5. Conclusion

Regular use of stereomicroscope in routine laboratory procedure not only aids in analyzing the surface topography of the biopsy specimens better, but also acts as an effective way in revealing many vital clues as well asproper embedding of the tissues for further histopathological

examination. Stereomicroscope as an adjuvant in grossing enables a confident histopathological diagnosis.

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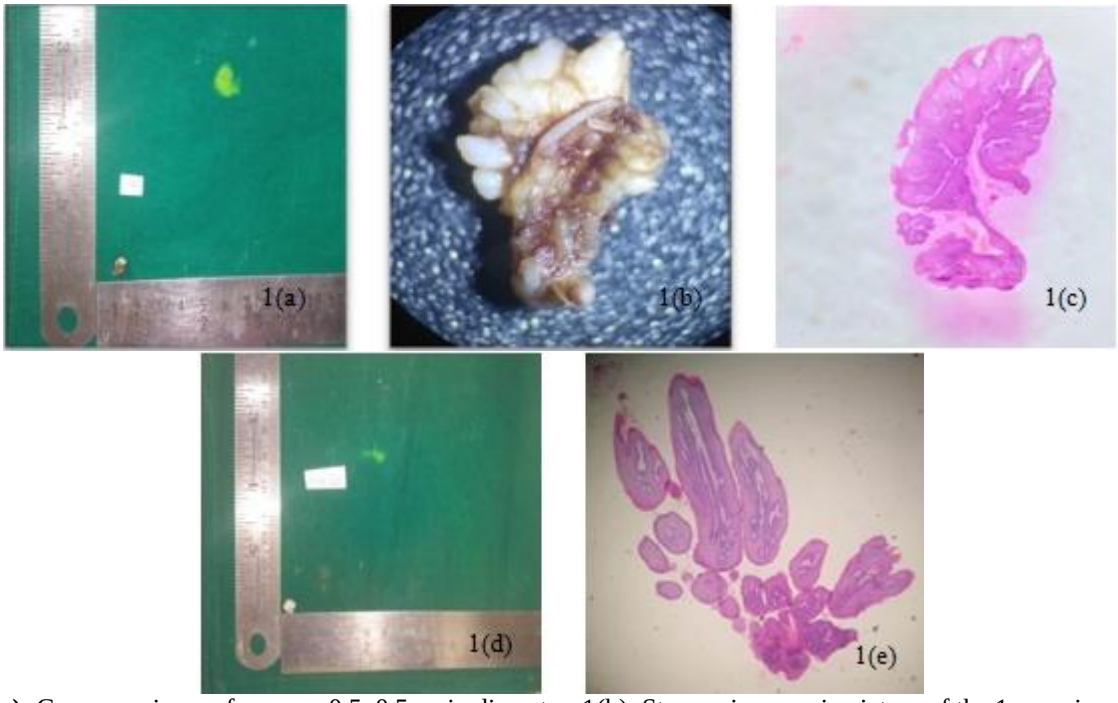

Figure 1(a): Gross specimen of approx. 0.5x0.5cm in diameter; 1(b): Stereomicroscopic picture of the 1a specimen showing whitish finger like projections; 1(c): Photomicrograph showing finger like projection of parakeratinized stratified squamous epithelium and thin connective tissue core; 1(d): Gross specimen of approx. 0.5x0.5cm in diameter; 1(e): Photomicrograph showing tangentially cut epithelium and connective tissue

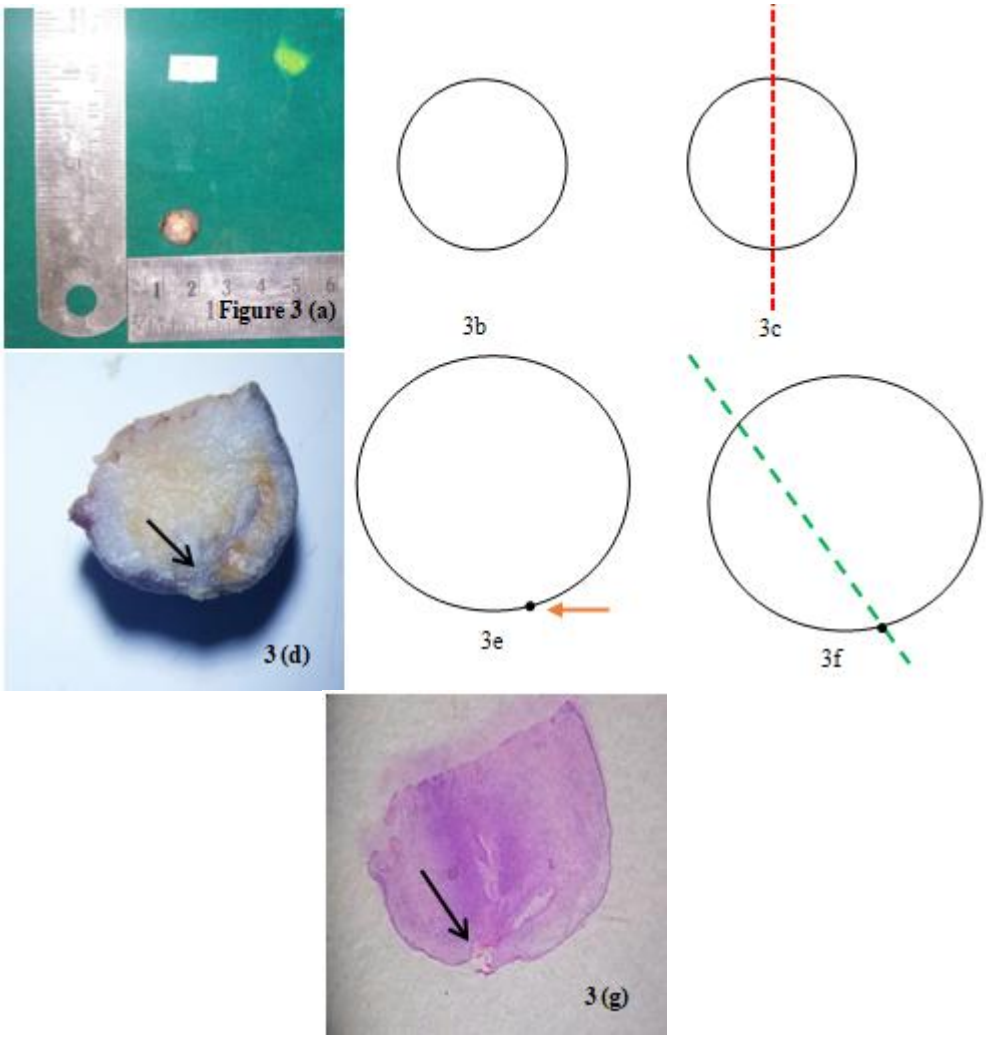

Figure 3 (a): Gross specimen of approx. 1x1cm in diameter; 3(b): Schematic representation of the specimen under naked eye; 3(c): Schematic representation of grossing of the specimen under naked eye overlooking the punctuate 3 (d): Stereomicroscopic image of the cut surface showing follicle like structure surrounded by brownish area (black arrow); 3(e). Schematic representation of the specimen showing a punctate like structure under the stereomicroscope (represented by the arrow); 3(f): Schematic representation of grossing of the specimen involving the punctate under stereomicroscope; 3(g): Photomicrograph showing hair follicle surrounded by inflammatory cells