

REVIEW ARTICLE



Grossing of Oral Pathologies—Revisited

¹Raju Shashidara, ²Shankargouda Patil, ³Roopa S Rao, ⁴Ebi Scindiya, ⁵Ann Tryphena

ABSTRACT

Grossing pathologies, otherwise known as macroscopic cutup for diagnostic information, is a vital laboratory step as it impacts the patient treatment and prognosis. However, it is a challenging skill acquired with keen observation, experience, and correlation between macroscopy and microscopy. Before we make an attempt to gross, it is imperative to have sound knowledge about its general principles and its applications as it differs among different lesions. Thereby, we have made an attempt to enhance the guidelines for gross description and also updated on the general principles. A practical insight has been provided with respect to grossing of oral mucosal biopsies, pathology of malignancies, odontogenic cysts, cystic odontogenic tumors, salivary gland pathologies, and lymph nodes along with the brief history of gross pathology.

Keywords: Grossing, Inking, Oral pathologies, Safety measures.

How to cite this article: Shashidara R, Patil S, Rao RS, Scindiya E, Tryphena A. Grossing of Oral Pathologies—Revisited. *J Contemp Dent Pract* 2017;18(12):1213-1222.

Source of support: Nil

Conflict of interest: None

INTRODUCTION

Surgical pathology has today become the cornerstone in patient care. Not only are pathological specimens important in establishing the diagnosis of a patient's disease, they have also become vital in dictating patient treatment and prognosis. It is indeed ironic that what was described as "Stuckchen-Pathologie," i.e., the pathology of small

fragments or piecemeal pathology, by the then famous European pathology professors who gave a preeminent status to autopsy pathology, is today an integral part of the diagnostic algorithm.¹

"Grossing"—the term refers to the detailed examination and the systematic dissection of surgical specimens in order to obtain tissue sections, which facilitate microscopic examination. It is the first step in the dissection procedure of a surgical pathology specimen.² It is generally performed by any trained personnel like a pathologist, resident, physician assistant, histo-technologist, or a biomedical scientist. In an oral pathology lab, this would inevitably fall on the junior-most trainee. However, descriptions of this important step are rather rare³ in oral pathology literature, and grossing procedures have often been handed down from senior pathologists to a novice in what has been described as the "Oral Tradition" by Waldemar Schmidt.⁴ These often times is random and on a specific case basis making generalizations irrelevant. This along with the fact that the specimens received are varied and with different degrees of complexity has led to most of us visualizing gross pathology as an insurmountable odd, leading further to its neglect. This article is an attempt to provide an algorithm to be followed in the oral pathology gross room.

Study the past if you would define the future.

—Confucius

A BRIEF HISTORY OF GROSS PATHOLOGY

The first book to describe gross pathology was probably written by Giovanni Battista Morgagni (1682–1771), titled "De sedibus et causis morborum per anatomen indagatis libri quinque" (The Sites and Causes of Disease Investigated by Anatomy). Xavier Bichat (1771–1802), the so-called "father of histology integrated anatomy, physiology and pathology," drew attention to the components of the organs, i.e., the tissues. Further contributions to this field came from Paris, which in the nineteenth century abounded in truly great physicians who practiced hospital medicine—Philippe Pinel (1745–1826), Pierre Bayle

^{1,4,5}Department of Oral and Maxillofacial Pathology, Coorg Institute of Dental Sciences, Virajpet, Karnataka, India

²Division of Oral Pathology, Department of Diagnostic Sciences College of Dentistry, Jazan University, Jazan, Kingdom of Saudi Arabia

³Department of Oral Pathology, Faculty of Dental Sciences M. S. Ramaiah University of Applied Sciences, Bengaluru Karnataka, India

Corresponding Author: Raju Shashidara, Department of Oral and Maxillofacial Pathology, Coorg Institute of Dental Sciences Virajpet, Karnataka, India, e-mail: shashidara_r@yahoo.com

(1647–1706), Jean-Nicolas Corvisart (1755–1821), and Laennec (1781–1826), who developed the stethoscope, thus contributing to the growth of macroscopic pathology by their voluminous writings brought about with large amounts of experiences in particular diseases.⁵

Thomas Hodgkin (1798–1866) devoted most of his time as a lecturer of morbid anatomy at Guy's hospital in London and brought out a two-volume monograph on autopsy pathology. Three contemporaries who shaped pathology during this period and later became known as "the three great men of Guy's" were Hodgkin, Richard Bright, and Thomas Addison. They methodically correlated the clinical findings of patients with the postmortem findings. This is probably the reason why all the three had diseases named after them.⁶ Dr. Johann Wagner (1800–1832) and Carl Rokitansky (1804–1878) at the Allgemeines Krankenhaus (General Hospital) in Vienna performed 30,000 autopsies and reviewed another 90,000. Their three-volume Handbook of Pathological Anatomy (Handbuch der Pathologischen Anatomie) immortalized this knowledge and probably represents the peak of macroscopic pathology.^{7,8} Despite the advent of microscopy in the seventeenth century through the work of Marcello Malpighi (1628–1694), Antoni van Leeuwenhoek (1632–1723), Robert Hooke (1635–1703), and several others, surprisingly the microscope seems to have been seldom used to study human tissues until Rudolf Virchow (1821–1902) in his seminal book, *Die Cellularpathologie in ihrer Begründung auf physiologische und pathologische Gewebelehre* (Cellular Pathology Based on Physiological and Pathological Histology) transformed medicine with his elucidation of the cell theory—*omnis cellula a cellula* (all cells come from another cell) that firmly established the foundations of cellular pathology, which is based on the modern diagnostic pathology. His work also greatly supplemented and expanded Rokitansky's contribution.⁸ Ironically, this also led to a decrease in interest in gross pathology. Virchow's later book on tumor pathology (*Die krankhaften Geschwulste—The Morbid Growths*), however, gave prominence to the combined macroscopic and microscopic examinations of tissues.

The twentieth century has seen an explosion of newer diagnostic techniques, such as immunopathology, molecular pathology, better imaging modalities, etc., which have contributed to the declining use of the autopsy thus, leading to ignorance of the gross pathology.

I had Five good friends what, why, how, when and where; they taught me all I need to know

—Author regrettably unknown

WHY, WHERE AND WHEN?

Grossing is the first step in biopsy reporting that ensures the best handling of the specimen and, hence, has been

equated to a "Biopsy triage".³ Special emphases on the type of specimen, urgency of reporting, and ensuring a representative sampling and specimen fixation are pivotal to this process. The gross room today acts as a bridge between the operating surgeon and the diagnostic pathologist and, hence, the final diagnosis and treatment plan depend upon the procedures carried out in this area. The ever-increasing level of advanced diagnostic and ancillary techniques makes adequate gross sampling today more important than ever.

The gross room should be sufficiently large depending upon the quantity of specimen expected to be received. Natural lighting would be an added asset to any gross room. Installation of antiskid floors with areas of floor drain would be ideal. The room should be well-illuminated and ventilated. Temperature control units and bio-safety hoods should ideally be present. Separate areas for photography, storage of liquid and solid waste, shelves for specimen containers, ready access to formalin, large tables for dissection of specimen, and sink with provision for hot and cold water, etc. are indispensable.⁹

As to whether it is advisable to gross tissues fresh or after fixation, there are differing points of view. A few believe that it is ideal to gross a tissue when fresh.¹⁰ What the histo-technologist receives is usually a specimen already in some form of fixative. The fresh tissue does have certain benefits, such as identification, inking, determination of tumor margins, harvesting and processing of lymph node dissections,¹¹ and splitting into specimens of uniform thickness to ensure uniform fixation, etc., to name a few. They are also requisite for a lot of the ancillary techniques; however, they are hazardous. It is the experience of the authors that both types yield similar results and we prefer the fresh tissue for excisional biopsies and fixed for incisional biopsies.

SPECIMEN ACCESSION AND IDENTIFICATION

Every Oral Pathology Department will surely have its own specimen identification system in place invariably comprising a combination of numbers and alphabets. This unique number/identification has to be carried from the specimen container until the report. This section will focus more on the requisite information needed before a specimen can be accessioned.

It is wrong always, everywhere and for anyone to believe anything on insufficient evidence.

—William Kingdon Clifford

SPECIMEN CONTAINERS

As a rule, specimen containers employed for patients should have a firmly attached label with relevant data of patient's name, age, medical record number, date,

and time of procedure performed followed by surgical request/requisition form.¹² Thereby, surgical specimen without a written request form or on an oral instruction should not be processed unless under exigent circumstances. Various types of specimen containers are available commercially, which may be utilized. It is imperative that the container have sufficient space for accommodating the fixative.¹³

REQUISITION FORM

The requisition form is probably the most important bit of document in the preanalytical phase of surgical pathology reporting, and there are serious risks of errors being made in this document.¹⁴ It should carry the patient's details, information on the anatomic site, nature of specimen, surgical procedure performed, and what the clinical suspicion is. All of this information should be succinctly conveyed. Information about the operating person and about the liaison person in the department in which the procedure was performed would be an added advantage in case they need to be contacted for any further queries. While the onus of submitting a specimen with a completed requisition form lies with the operating department,⁴ the pathology department equally holds the onus not to gross the specimen with incomplete/missing requisition forms. Performing the gross procedure on a telephonic request should also be completely avoided. Most centers today use bar codes¹⁵ to store all relevant data. Where such a facility exists, both the requisition form and specimen bottle should also carry the relevant bar code. While the emphasis on the aforementioned aspects might seem an overexaggeration, it would be more prudent to note that among all errors mentioned in diagnostic pathology, the specimen labeling errors are the most common with a range from 0.09 to 0.25 percentile.¹⁴ Such errors, when they occur, may have serious implications in patient care and make the pathologist liable for legal action.

The camera is an instrument that teaches
people how to see without a camera.

—Dorothea Lange

SPECIMEN PHOTOGRAPHY

An often overlooked part of the oral pathology gross room procedure is the quality of photographs of the gross specimen. Photographs are an important component of the documentation. They can be used in teaching, conference presentations, and publishing. It is indeed unfortunate that specimen photographs are not taken and, if taken, they suffer from a number of errors that could be easily avoided. Considering the pace at which digital image-capturing technology has been progressing, what

probably is required is a bit of commonsense to capture that optimal gross photograph.

Choosing a digital camera for gross photography is not all that difficult considering the wide array of choices available. However, a camera with a sufficient resolution (10 MP or more) and a good-quality sensor would be ideal. The camera should also have a good-quality macro setup and an image-stabilizing technology.

The availability of a good camera does not necessarily mean good-quality pictures. The use of appropriate backgrounds and proper lighting would be an absolute necessity for obtaining good pictures. While commercially available lighting tables, such as the aristo-4 would indeed be desirable, the use of custom-made tables or the use of simple-colored gels or fabrics is more economical and practical. For most specimens, the colors red, yellow, and green are best avoided. Probably, the most universally suited background would be a black velvet background.⁴

The specimen should be photographed fresh or after a brief fixation period. The brief fixation has the advantage of reducing glare associated with the fresh specimen.¹⁶ If tissue has been in the fixative for a longer duration, some reversal can be obtained by immersion in 70% alcohol for 10 to 15 minutes.^{17,18} Prior to photography, the tissue must be prepared and trimmed by washing to remove blood, blood clots, fat, and opening the ducts and vessels, and removing other unnecessary tissues around the lesion.

The use of scales and labels, though considered unnecessary by a few,⁴ would help the viewer better orient the specimen and be provided with a perception about the specimen size. All other distracters should be avoided. Photographs of lesional surface, cut-section view, and views of the cavity in case of cystic specimen will be beneficial in the final diagnosis. The photographs taken should also be stored along with the patient records with the appropriate accession number to aid in easy retrieval.

SPECIMEN RADIOGRAPHY

Though specimen radiography is more useful in orthopedic and breast pathology, its importance in oral pathology is no lesser. Radiographic examination of appropriate specimens, such as an odontogenic tumor or an excision specimen of an oral malignancy could provide important information about the extent of the lesion, the nature of spread of the lesion through bone, and also give insights about where the tissue has to be accessed for margins. Some pathologists have found specimen radiographs useful for locating lymph nodes in radical resection specimens.^{18,19} Commercially available units like Faxitron have been used to perform a microradiographic analysis of bone.²⁰ The normal intra oral periapical radiograph unit, in the experience of the authors, is usually sufficient

to provide good-quality images of specimen, albeit with use of lower exposure settings. "The devil is in the detail."

GROSS DESCRIPTION

The gross description is an important integral part of macroscopic surgical pathology; however, more often, the details are lost in extremely verbose descriptions, which are irrelevant, or suffer from insufficient information. Thus, it is better to have a gross description that is structured, succinct, and meticulous. The gross description is also an important document as it allows for a relative visualization of the histopathology slide in relation to the other surrounding structures thus, permitting an imaginative reconstruction of the specimen. The document also gives details about how the tissue has been distributed for various diagnostic modalities and serves as a slide index. Also, it provides the histopathologist with the precise location of the tissue in the glass slide in relation to the rest of the specimen.⁴

The first usually deals with the identification of the patient and the specimen. The reader should be able to correlate the patient, the specimen, and the structures present with this description. It would be imperative that the gross description encompasses both the normal and abnormal components of the tissue provided with the description, moving from a general picture to focusing on the specifics of the lesion in question. After a generic description of the specimen, characteristics, such as size, number, color, shape, consistency, and weight of the specimen should be recorded correctly. A description of various colors and consistencies and their interpretation is provided (Tables 1 and 2).

Paramount among these is the size of the lesion, especially, in tumor resections, as this is an important aspect

Table 1: Colors normally observed in oral biopsies and their interpretation²¹

| Color | Interpretation |
|-------------|-------------------------------------|
| White | Calcification/bone |
| Gray white | Fibrosis, invasion lymph node |
| Yellow | Lipoma, fat, necrosis |
| Brown/black | Hemosiderin, melanin |
| Red | Hemorrhage, blood vessel congestion |

Table 2: Different consistencies and their probable interpretations²¹

| Consistency | Interpretation |
|-------------|--|
| Soft | Necrosis, fluid |
| Firm | Fibrosis |
| Hard | Bone, calcification |
| Rubbery | Lymph nodes |
| Cheesy | Keratin (KCOT, sebaceous cyst), caseous necrosis |

KCOT: Keratinizing cystic odontogenic tumor

of the tumor staging. The clearance of the margins, which usually refers to the distance from the tumor boundary to the excision boundary, helps determine the adequacy of excision and either dictates or may preclude the need for adjuvant therapy. The use of general terms, such as walnut-sized e.g. should be avoided, and the size be mentioned in terms of centimeters. All dimensions, such as length, width, height, etc. ought to be included.

The second important aspect that ought to be described and done so in conjunction with size is the number. As oral pathologists, it is very common to receive curettage/excision specimen as multiple tissue bits. In this situation, it would be imperative to count the number of pieces of tissue received and describe each separately. When the number is few, a gross description of the size, color, consistency of each would be desirable. When the numbers of tissue bits are large, it would be advisable to have detailed descriptions of the larger tissue bits individually and the smaller bits as a group, e.g., giving a range for the size instead of describing each size individually.

Two things often erred in a gross description is an overdescription of the normal structures, which are irrelevant to the pathology in question, and a description of the process of grossing, which should be avoided.

The final part of the description would be to serve as means of slide cataloging. This is a very important piece of information, which communicates to the diagnosing pathologist each slide in perspective to its actual location in the gross specimen. It is common to use a combination of numbers and letters to designate each/part of a specimen. Since tissues are routinely converted into tissue blocks, this is also termed a block index (Fig. 2). The information catalog should ideally list the number of tissue pieces allocated to each block and their relative sizes, if possible. The slide/tissue block catalog will also be an important piece of document while communicating with other centers. It is again worth reiterating that the numbering system followed should be simple and reproducible.¹⁴

Another important piece of information that ought to be present in the final part of the description is the allocation of tissues for ancillary techniques, when applicable. Photographic techniques should be generously used during the grossing description to act as a visual guide used when referencing is necessary. In today's world of digital photography, where there are no restrictions on the number of photographs that can be captured, authors must deem it necessary that this simple inexpensive tool be utilized to its maximum.

APPROACH TO COMMON ORAL MUCOSAL BIOPSIES

A good number of mucosal biopsies received in oral pathology practice will be small incisional biopsies, which

require special care during processing. A variety of techniques and gadgets are available to deal with such specimens. Probably, the simplest and most commonly used is the tissue paper/lens paper employed during processing; however, it is advisable to use a wet paper rather than a dry one and have an area of marking to locate where the tissue has been placed postprocessing. The use of nylon cloth, fenestrated mesh processing cassettes, and cellular polyester urethane foam pads are various tools used to facilitate processing of these small tissues, albeit with their own disadvantages. Alternatively, the use of techniques, such as filtration directly into the cassette or the HistoGel tissue processing medium can be attempted.²²

The judicious use of inks can assist in notifying the proper embedding method and also to mark areas of special interest; however, care should be taken to prevent ink from spreading to cover the entire specimen rendering the entire inking process redundant.

INKS AT THE GROSSING TABLE

The use of various dyes in a liquid form (inks) or colored powder can be used to indicate while grossing the tissue to demonstrate areas of interest in the final histopathological slide. This procedure is called as inking.^{22,23} Ink can be applied using a variety of applicators ranging from wooden picks to Stratagene's Strata Tips (Stratagene Inc, La Jolla, CA).²⁴

Inking is most commonly used to indicate or mark the margins of an excision. Alternatively, it can also be used for indication of areas of interest and also to aid in orientation of the specimen during embedding. Usually, the unstained area is placed down in an embedding mold.²²

There are a variety of colored inks available for inking of histopathology specimen, such as the Davidson marking system²⁵ to the ubiquitous India ink. India ink has the benefit of being protein-based and, hence, being more firmly fixed to tissues by cross-linking fixatives. This property is, however, disadvantageous in rapid processing schedules, as the dye might wash off due to insufficient fixation time.²²

India ink has for long been the dye of choice in inking resection margins. However, the availability of different dyes and the necessity to code each margin in a different color so as to identify it separately during each step of processing to the microscopic slide has now led to the practice of use of multiple dyes for inking. However, the easy availability of these multitudes of commercial dyes in different parts of the world is a concern. In this situation, a variety of natural-colored compounds and subsets of the Fevicryl hobby colors© have been found to be viable alternatives giving good results under microscopic examination.²³

Irrespective of the dye used or the reason for use, the ink should be applied judiciously. Excessive use of the ink may lead to leakage of the ink into tissue spaces leading to misrepresentation of margins or surfaces, thus making the entire process superfluous and leading to false negative margins and false positive margins. Almost all commercial inks come with indications of a drying period that is necessary for complete fixation of the ink. This can be avoided by using a color enhancer, such as acetic acid 5% in 3 to 50% white vinegar, which accelerates the drying process.^{22,26}

The following steps should be followed while inking a gross specimen:

- Preferably apply the dye to unfixed specimen
- Pat the fresh tissue dry with paper towels before application of dye
- Use appropriate applicator
- Provide adequate time for drying or use fixer
- Apply dye before cutting the specimen
- Do not cut on wet ink.

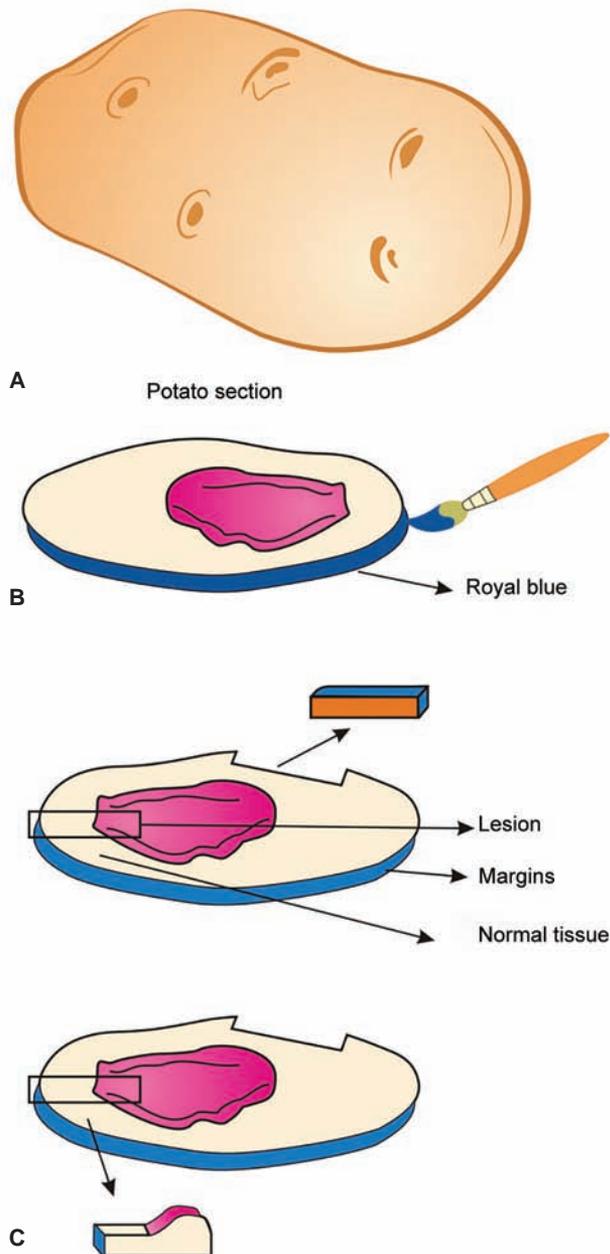
GROSSING OF MALIGNANCIES

While the "Stuckchen-Pathologie" is often purely diagnostic in nature, the grossing of malignancies assumes a different proposition, in that not only are they diagnostic, but also prognostic, and aid in staging. They have direct therapeutic implications as in the need for additional surgery or chemo/radiotherapy.

A surgical margin refers to that area of apparently normal tissue removed by the surgeon during excision of the tumor to ensure complete removal of the lesion. As mentioned earlier, the status of these resection margins is a very important part of postresection tumor staging, and has therapeutic implications and final prognosis. Hence, the margins probably represent the area of prime importance in excisions.

In addition, orientation of the specimen is of paramount importance, especially in head and neck resections, which may have many complex surfaces representing the resection margins; hence, it always makes sense to get the orientation of the specimen correctly before proceeding with inking the margins to prevent misinterpretations. Always establish the superior, inferior, anterior, posterior margins along with the medial and lateral margins before proceeding. When in doubt, confer with the surgical consultant before proceeding.²⁶

There exist various ways in which margins can be obtained. The commonly used ones are the En Face (parallel, horizontal) as shown in Figure 1. These margins will consist of the entire cut surface of the tumor margin and can be used when the tumor is not placed close to the margin, especially in small lesions. Such a margin will give information about clearance of margins. However,



Figs 1A to C: Methods of obtaining horizontal and vertical margins demonstrated using a potato. (A) The inked cut surface of the specimen indicates the resected margin; (B) sample to include lesional, normal, and marginal areas. Horizontal margin allows examination of greater surface area; and (C) assessment of margins. Vertical margin gives distance of clearance

the amount or distance of clearance from the tumor cannot be ascertained. The benefit of this method is in the ability to examine larger areas of tissue for marginal clearance.²⁷

In contrast, the “On edge” (vertical, perpendicular) margin will give the relationship of tumor to margin albeit with the disadvantage of having to make multiple samples and subject those to multiple serial sections²⁸ as demonstrated in Figure 1.

The third method or the Mohs method requires cutting of the margins at a slant between vertical and horizontal sections.²⁹ While smaller specimens can be grossed and mounted *in toto* onto a processing capsule

as demonstrated in Figure 2, larger specimens require a little more attention to obtain adequate margins.

The most common method employed is the bread-loaf method amidst varied ones. This method yields the following types of vertical margins.

- Transverse through tumor as shown in Figure 3;
- Longitudinal through tumor; and
- The third type of margin that can be used in combination with the bread-loaf method is the peripheral or perimeter sections.³⁰

Perimeter sections are obtained when no tumor tissue is expected near the margins; however, when obtaining perimeter margins, it is also mandatory to obtain sections with the most peripheral tumor extension as supplemental data.

While checking 100% of tumor margins, it is possible to do so using horizontal sections. Achieving this is, however, rather difficult for vertical sections and will necessitate multiple serial sections. Hence, a combination of both methods may be used to gather the most relevant diagnostic information. Thus, we can obtain a combination of transverse and longitudinal vertical sections as shown in Figure 4 or a combination of transverse and peripheral sections or if need be, a combination of all three methods as seen in Figure 5. By judicious methods of palpation, the areas with tumor closer to the margins can be identified and a vertical section may be obtained whereas the other areas can be sampled using a horizontal section.

Irrespective of the type of margin obtained, the fact remains that complete eradication in multifocal tumors is difficult to predict. However, to paraphrase Batsakis,³¹ “fact remains that despite the seemingly random quality of the status of surgical margins they still provide therapeutic and prognostic guidelines.” Probably, more standardizations in the way margins are obtained lead to a greater impact in treatment.

GROSSING LYMPH NODES

The second most important aspect in grossing of tumor excision is the harvesting of lymph nodes. Though identification of tumor metastasis to lymph nodes is of paramount importance in staging tumors, identification of the lymph nodes in the specimen is often difficult, especially in fixed tissue in which tissue hardens making palpation, which is the best method for lymph node identification, impossible. This is especially true of the smaller lymph nodes, which are covered by fibroadipose tissue, making identification difficult. Like all skills, identification of lymph nodes is developed with time and practice; however, the following caveats need to be observed⁴:

- Gross tissues fresh
- Orient the specimen first and identify the levels of lymph nodes

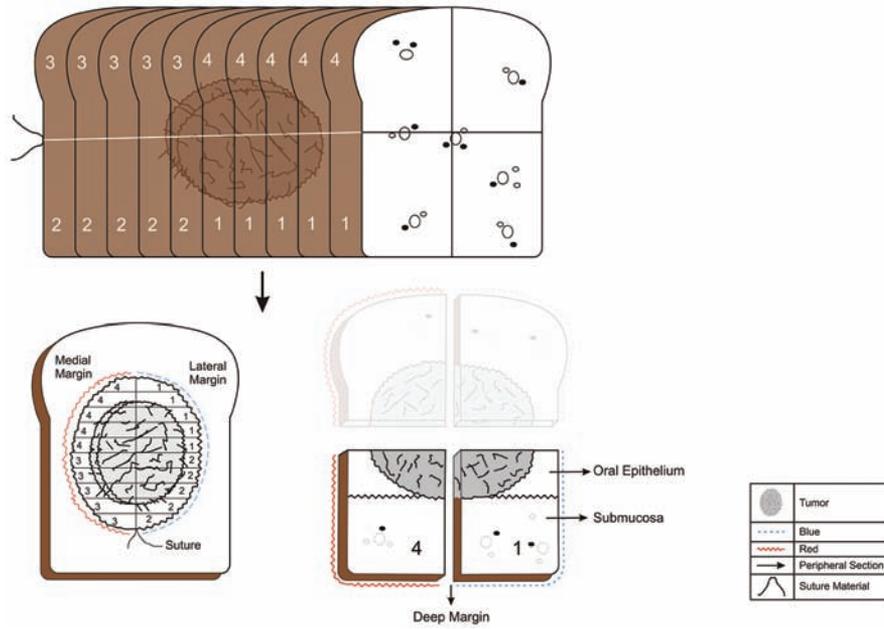


Fig. 2: Bread-loaf methods with transverse vertical sections for larger specimen (1–4) are representative numbers for the block index

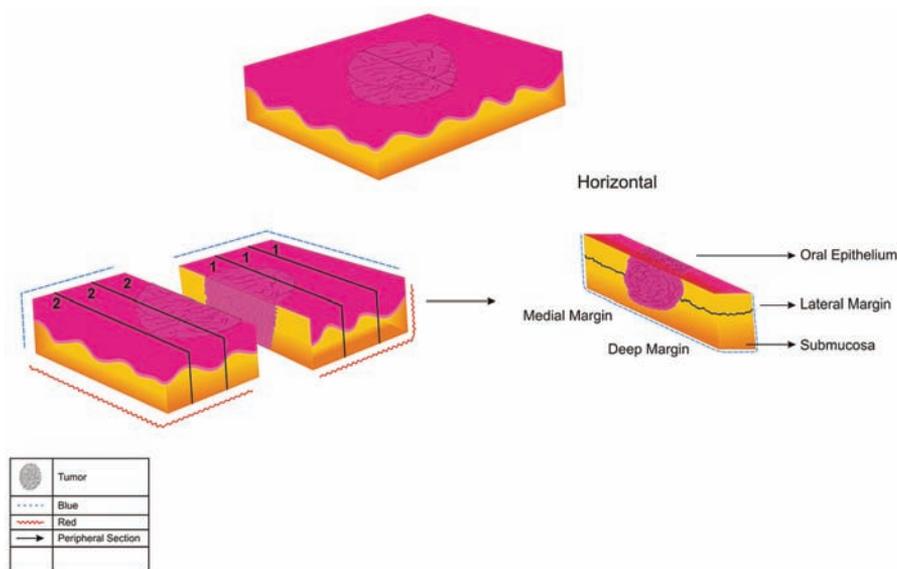


Fig. 3: Grossing a smaller specimen demonstrated using a cake

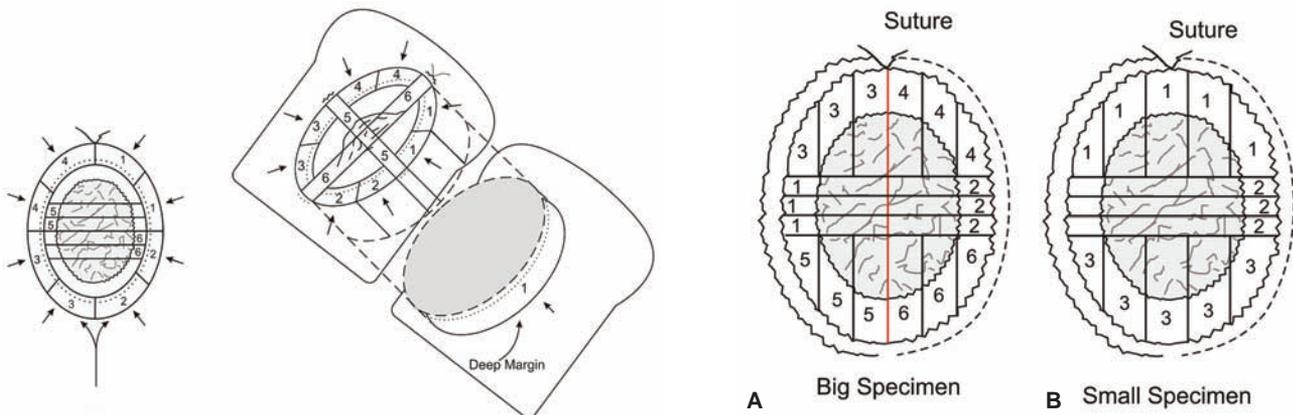


Fig. 4: Bread-loaf with a combination of transverse and longitudinal vertical sections

Figs 5A and B: Combination methods to obtain most relevant sections: (A) Combination of transverse and peripheral vertical sections; and (B) combination of bread-loaf transverse, longitudinal, and peripheral sections

- Gross the margins and other soft tissue before attempting to harvest lymph nodes
- All harvested lymph nodes should be submitted for histopathological examination.

Harvesting lymph nodes many a times requires substantial manipulation and induction of alterations. Hence, orientation of the specimen may not be possible after lymph node harvesting. Any node greater than 5 mm is generally considered a positive unless proved otherwise by histopathology. Hence, when larger nodes are harvested, they have to be cut into smaller pieces of 3 mm to aid in fixation and better representation of tissue. The cutting of lymph nodes into smaller slices can be performed along the long axis or the short axis, and this has been done on individual preferences with both methods having their share of proponents; however, recent mathematical simulations have favored a long-axis approach³² as in Figure 6. The long-axis approach also has the added benefit of maximization of lymphatic channels supplying the node and minimizing the number of sections.^{33,34}

In addition to the aforementioned guidelines, the following recommendations during grossing and reporting from the Association of Directors of Anatomic and Surgical Pathology can also be followed¹¹:

- No clearing of adipose tissue is necessary, although it may represent an institutional or individual preference.
- Submit the entire nodes cut unless they contain grossly visible tumor, in which case fewer slices are required, or if they are grossly largely replaced by adipose tissue, in which case processing is optional.
- Lymph node levels in a dissection specimen should be specified and submitted separately when

clinically appropriate (e.g., neck dissections, colectomy specimens).

- The summary of sections in the surgical pathology report should include how many sections of how many nodes are submitted in each cassette. Different colored inks may be used to distinguish different nodes submitted in a single cassette.
- One hematoxylin and eosin slide per cassette is recommended.
- Immunohistochemical analysis and other specialized techniques may be used as part of a research study or for differential diagnosis, but are not now considered mandatory.

REPORTING

- The number of lymph nodes positive for metastatic disease and the total number of lymph nodes examined microscopically should be reported, with specific levels mentioned when appropriate.
- The size of the largest metastasis (measured on the slide) should be reported, if clinically indicated.
- The presence of extracapsular extension may be reported, depending on the primary site and institutional preference (particularly for head and neck carcinomas).
- If the only tumor seen is in extranodal vessels, this should be stated.
- Deposits of tumor not associated with any structure recognizable as a lymph node should be designated separately.
- In rare situations, the grading of nodal metastases may be important.

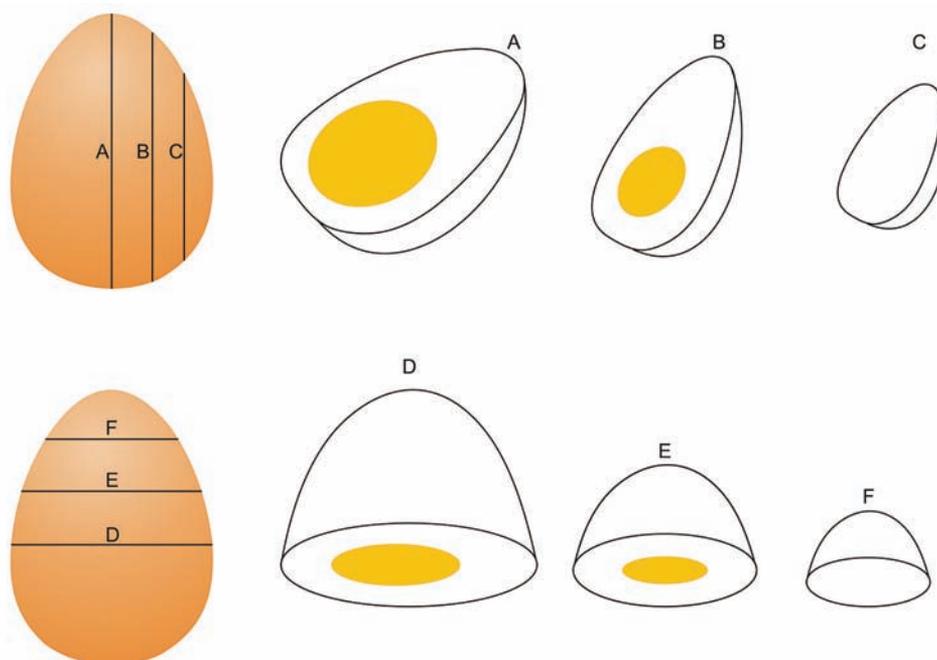


Fig. 6: Approaches to grossing lymph nodes demonstrated using an egg³⁵

- After preoperative chemotherapy and/or radiotherapy, notation of necrotic *vs* nonnecrotic tumor is recommended.

An important, yet often overlooked practice in grossing lymph nodes is the obtaining of imprint smears. These contain valuable information and can be obtained by application of the cut surface on the lymph node onto a glass slide and use of minimal pressure.

Occasionally, isolated lymph node biopsies of the head and neck are done for diagnosis of an infectious process or as a sentinel node biopsy. The protocols mentioned above work for such cases as well. However, in infections, such as tuberculosis, it is necessary to submit a small portion of the node for microbiology.³⁵

GROSSING OF SALIVARY GLANDS

While all the techniques mentioned until now are unique for a variety of reasons, the parotid contains numerous lymph nodes within it and special care should be taken to harvest these. Impressions of demarcation of tumor from surrounding tissue, presence of macrocapsular invasion, areas of cartilaginous tissue, etc., if described, may assist in the diagnostic process. While grossing salivary gland excisions for tumors, do not treat the entire tissue as one and bread-loaf it; rather section it in a way to show relationship of tumor to inked margins, to surrounding uninvolved glandular tissue, and relationship to nerves.

In case of cystic lesions of salivary glands, the lesions can be bisected and submitted *in toto*.^{4,35}

ODONTOGENIC CYSTS

The process of grossing odontogenic cysts and cystic odontogenic tumors requires attention to a few peculiarities that needs to be evaluated. Always examine the cyst wall closely for any elevations or irregularities. If it has a complex appearance, take separate slices of these areas and put them separately in a cassette as these could indicate areas of neoplastic transformation.

Another area frequently ignored while grossing odontogenic cysts and tumors is the bone, considering that close to 99% of odontogenic cysts and tumors are intrabony and have capabilities to infiltrate bone, the ignoring of bone is quite perplexing and probably occurs because of the preeminence given to soft tissue diagnosis.

Three factors are important in grossing bone:

1. Anatomic orientation
2. Immobilization
3. Proper instrument.

Vis-à-vis oral and maxillofacial specimens, good immobilization may be obtained either using a vacuum vise or a table vise, a mini hack saw/hand saw, or a Stryker saw may be used to obtain slices of adequate

thickness. Obtaining transverse sections is more desirable than longitudinal sections, since these provide better diagnostic information;³⁶ this has to be followed by a proper decalcification procedure using any one of the various decalcifying agents.

SAFETY AND PRECAUTIONS IN THE GROSS ROOM

Faculty who work regularly in the gross room are exposed to many possible risks including infections, chemicals which may be flammable, toxic, allergenic, or carcinogenic, and electrical, and physical hazards as well as cuts and the rather common needle stick injuries.^{37,38} Fragments of bone during cutting of bone, fine particles of bone etc. can get disseminated into the air during the grossing procedure and are potentially biohazardous.³⁶

Judicious handling of these chemicals goes a long way in preventing accidental contact. A proper gross room design and use of biosafety precautions would also be essential in preventing accidental injury. All tissues are to be considered potentially hazardous and universal precautions must be taken as per occupational safety and health administration regulations. Proper barrier techniques to prevent spread of infection must be undertaken, such as disposable gowns, gloves,³⁶ facemasks, and eye gear.

CONCLUSION

Like all skills, grossing is a skill that will develop in individuals over a period of time; however, skills cannot be gained in the absence of knowledge and efforts. Simple steps, such as observation of as many grossing procedures as possible, regularly attending autopsies, and bringing about a correlation between macroscopy and microscopy will help accelerate the learning curve. In the era where microscopic pathology has become important, books on gross pathology are rather rare to find. Nevertheless, there exist some good books,^{4,16,31} which can be accessed. In the era of e-learning, there do exist few online resources. Of which special mention needs to be made of grossing-technology.com, which is dedicated to gross pathology readings. This website will surely enrich your knowledge of grossing.

REFERENCES

1. Lattes, R. Pathology and therapeutics. In: Rosai J, editor. Guiding the surgeon's hand: the history of American surgical pathology. 1st ed. Washington (DC): American Registry of Pathology; 1997. pp. 41-60.
2. Romaguera R, Nassiri M, Morales AR. Tools to facilitate and standardize grossing. *Histologic* 2003 May;36(1):17-21.
3. Dimenstein IB. Grossing biopsies: an introduction to general principles and techniques. *Ann Diagn Pathol* 2016 Jan;13(2):106-113.

4. Westra, WH.; Hruban, RH.; Phelps, TH.; Isacson, C. Surgical pathology dissection. New York: Springer; 2003. pp. 88-92.
5. King LS, Meehan MC. A history of the autopsy. A review. *Am J Pathol* 1973 Nov;73(2):514-544.
6. Stone MJ. Thomas Hodgkin: medical immortal and uncompromising idealist. *Proc (Bayl Univ Med Cent)* 2005 Oct;18(4):368-375.
7. Ferraz de Campos FP. The dawn of modern pathology. *Autops Case Rep* 2016 Jan-Mar;6(1):1-5.
8. Day, CE. Histopathology – methods and protocols. New York: Springer; 2014.
9. Fisher, RE. A faculty guide to gross lab design. 2011. p. 1-7. Available from: http://www.anatomy.org/uploads/4/6/5/1/46517773/faculty_guide_gross_lab_design.pdf.
10. Bostwick, DG.; Cheng, L. Urologic surgical pathology. 3rd ed. Philadelphia (PA): Elsevier Health Sciences; 2014. p. 738. Available from: <https://books.google.co.in/books?id=wrHQAQAAQBAJ>.
11. Lawrence WD, Association of Directors of Anatomic and Surgical Pathology. ADASP recommendations for processing and reporting of lymph node specimens submitted for evaluation of metastatic disease. *Virchows Arch* 2001 Nov;439(5):601-603.
12. Bell WC, Young ES, Billings PE, Grizzle WE. The efficient operation of the surgical pathology gross room. *Biotech Histochem* 2008 Apr;83(2):71-82.
13. Dimenstein. fixation fixation fixation. Available from: <http://grossing-technology.com/newsite/home/grossing-in-dermatopathology-manual/skin-excision/>.
14. Layfield LJ, Anderson GM. Specimen labeling errors in surgical pathology: an 18-month experience. *Am J Clin Pathol* 2010 Sep;134(3):466-470.
15. Hanna MG, Pantanowitz L. Bar coding and tracking in pathology. *Surg Pathol Clin* 2015 Jun;8(2):123-135.
16. Edwards WD. Photography of medical specimens: experiences from teaching cardiovascular pathology. *Mayo Clin Proc* 1988 Jan;63(1):42-57.
17. Burgess CA. Gross specimen photography: a survey of lighting and background techniques. *Med Biol Illus* 1975 Aug;25(3):159-166.
18. Rao RS, Premalatha BR. Grossing in oral pathology: general principles and guide lines. *World J Dent* 2010 Apr-Jun;1(1):35-41.
19. Anderson J, Jensen J. Lymph node identification. Specimen radiography of tissue predominated by fat. *Am J Clin Pathol* 1977 Oct;68(4):511-512.
20. Dunn EJ, Beows DW, Rothert SW, Greer RB. .Microradiographic of bone, a new use for the versatile Faxitron (letter). *Arch Pathol* 1975;99:62.
21. William, GI. A primer of gross pathology. Springfield (IL): Charles C Thomas; 1972. p. 73.
22. Dimenstein IB. Grossing biopsies: an introduction to general principles and techniques. *Ann Diagn Pathol* 2009 Apr;13(2):106-113.
23. Tampi C. In search of the rainbow: colored inks in surgical pathology. *Indian J Pathol Microbiol* 2012 Apr-Jun;55(2):154-157.
24. Dimenstein, IB. Grossing technology in surgical pathology. Available from: www.grossing-technology.com.
25. Parkinson AV, Cannon CR, Hayne ST. Color coding surgical margins with the Davidson marking system. *J Histotechnol* 1990 Dec;13(4):293-295.
26. Peters, SR. A practical guide to frozen section technique. 1st ed. New York (NY): Springer; 2010. pp. 13-23.
27. Breastcancer.org. Surgical margins. 2016. p. 1-5. Available from: <http://www.breastcancer.org/symptoms/diagnosis/margins>.
28. Dimenstein, IB. Grossing technology in surgical pathology. 2014. p. 1-13. Available from: <http://grossing-technology.com/home/grossing-in-dermatology-manual/skin-excision>.
29. Ranjan R, Singh L, Arava SK, Singh MK. Margins in skin excision biopsies: principles and guidelines. *Indian J Dermatol* 2014 Nov-Dec;59(6):567-570.
30. Rapini RP. Comparison of methods for checking surgical margins. *J Am Acad Dermatol* 1990 Aug;23(2):288-294.
31. Batsakis JG. Pathology consultation surgical margins in squamous cell carcinomas. *Ann Otol Rhinol Laryngol* 1988 Mar;97(2):213-214.
32. Mahe E. Mathematical modelling of lymph node grossing techniques. *Can J Pathol* 2011 Jan;3(1):180-186.
33. Weaver DL. Pathology evaluation of sentinel lymph nodes in breast cancer: protocol recommendations and rationale. *Mod Pathol* 2010 May;23(Suppl 2):S26-S32.
34. van la Parra RF, Peer PG, Ernst MF, Bosscha K. Meta-analysis of predictive factors for non-sentinel lymph node metastases in breast cancer patients with a positive SLN. *Eur J Surg Oncol* 2011 Apr;37(4):290-299.
35. Allen, DC.; Cameron, RI.; editors. Histopathology specimens: clinical pathology and laboratory aspects. London: Springer London; 2004. p. 19-20.
36. Dimenstein IB. Bone grossing techniques: helpful hints and procedures. *Ann Diagn Pathol* 2008 Jun;12(3):191-198.
37. Bancroft, JD. Theory and practice of histological techniques. 6th ed. Edinburgh: Elsevier; 2008. p. 75-82.
38. Thirumala S, Pinkhasov D, Medalie N. A simple yet effective technique to improve laboratory safety for the grossing of large surgical specimens. *Ann Diagn Pathol* 2000 Feb;4(1):44-45.