The term biopsy indicates removal of tissue from a living body for microscopic diagnostic examination. Biopsy is the most precise and accurate of all diagnostic tissue procedures and should be performed whenever a definitive diagnosis cannot be obtained using less invasive procedures.

Health History

Frequently a close interrelationship exists between the medical and dental health of patients and that oral lesions can reflect, or contribute to, systemic health problems. Therefore, documentation of a detailed health history coupled with a thorough clinical evaluation, is essential for two basic reasons:

1. A preexisting medical problem may affect or be affected by the dentist's treatment of the patient.
2. The lesion under investigation may be the oral manifestation of a significant systemic disease → Surface ulcerations in a chronic smoker should alert the dentist to the possibility of oral or pharyngeal cancer.

History of the Specific Lesion

"If you listen to the patient long enough, he or she will generally lead you to the diagnosis."

Many systemic diseases (up to 85% to 90%) can be diagnosed by gathering a detailed medical history. Questioning of the patient who has a pathologic condition should include the following:

1. How long has the lesion been present? (duration - e.g. Several years lesion →might be congenital). Duration must be taken in context with other elements of the history because it is possible that the lesion was present before the patient became aware of it.
2. Has the lesion changed in size? (radiographic or clinical size or both). As an example: An aggressive, enlarging lesion → likely malignant.
3. Has the lesion changed in character or features (e.g., a lump becoming an ulcer or an ulcer starting as a vesicle)? e.g. Ulcer began as a vesicle → could be vesiculobullous or viral disease.
4. What symptoms are associated with the lesion (e.g., altered function anesthesia or paresthesia, abnormal taste, or odors)? Cancers, erroneously thought by many to be painful, are often painless unless secondarily infected.
   - Sensory nerve changes such as numbness or tingling →often malignant or inflammatory process.
   - Dysphagia → changes in the floor of the mouth or parapharyngeal tissues.
   - Swelling → expansile process from: inflammation, infection, cysts, or tumor
   - Painful lymph nodes → inflammatory, infectious or, malignancy.
5. What anatomic locations are involved? Certain lesions have a predilection for certain anatomic areas or tissues.

6. Are there any associated systemic symptoms (e.g., fever, nausea, or malaise)? E.g. Many systemic viral conditions like mumps can cause oral manifestations.
7. Is there any historical event associated with the onset of the lesions (e.g., trauma, parafunctional habits, hard or hot foods, application of medications, visits to foreign countries)?

Clinical Examination

An examination is classically described as a process that includes inspection, palpation, percussion, and auscultation. In the head and neck region, inspection and palpation are more commonly used as diagnostic modalities, with inspection always preceding palpation.

- Early inspection facilitates description of the lesion before it is handled because some lesions are so fragile that manipulation would compromise subsequent examinations.
- Percussion is reserved for examination of the dentition.
- Auscultation is infrequently used but is important when examining suspected vascular lesions.

The following are some important additional points to be considered during the inspection of a lesion.

1. Anatomic location of the lesion. → which tissues are contributing to the lesion based on the anatomic location. For example, if a mass appears on the dorsum of the tongue → epithelial, connective tissue, lymphatic, vascular, glandular, neural, or muscular origin.
2. Overall physical characteristics of the lesion. Appropriate medical terminology should always be used to describe clinical findings.

- **Bulla** (pl. Bullae): a blister; an elevated, circumscribed, fluid-containing lesion of skin or mucosa.
- **Crusts** (crusted): dried or clotted serum on the surface of the skin or mucosa.
- **Dysplasia** (dysplastic): abnormal cellular size, shape, or organization
- **Erosion**: a shallow, superficial ulceration
- **Hyperkeratosis**: an overgrowth of the cornified layer of epithelium
- **Hyperplasia** (hyperplastic): an increased number of normal cells
- **Hypertrophy** (hypertrophic): size increase due to increase in cells size.
- **Keratosis** (keratotic): An overgrowth and thickening of cornified epithelium.
- **Leukoplakia**: Slowly developing, firmly attached thickened white mucosal patches
- **Macule**: a circumscribed nonelevated area of color change
- **Malignant**: anaplastic; a cancer that is potentially invasive and metastatic
- **Nodule**: large, elevated, circumscribed, and solid palpable mass
- **Papule**: small, elevated, circumscribed, and solid palpable mass of the
- **Plaque**: any flat, slightly elevated superficial lesion
- **Pustule**: a small, cloudy, elevated, and circumscribed pus-containing vesicle
- **Scale**: a thin, compressed, superficial flake of keratinized epithelium

- **Stomatitis**: any generalized inflammatory condition of the oral mucosa
- **Ulcer**: a crater-like surface lesion resulting from necrosis of the epithelium
- **Vesicle**: a small blister, circumscribed elevation containing serous fluid

3. Single versus multiple lesions. → multiple or bilateral neoplasms in the mouth is unusual, whereas vesiculobullous, bacterial, and viral diseases commonly present such a pattern.
4. Size, shape, and growth presentation of the lesion. A small metric ruler made of a material that can be disinfected is useful to have on hand. The growth presentation should also be noted: whether the lesion is flat or slightly elevated, endophytic (growing inward) or exophytic (growing outward from the epithelial surface), and sessile (broad based) or pedunculated (on a stalk).
5. Surface appearance of the lesion. Smooth, lobulated (verruciform), or irregular. If ulceration present: characteristics of its base and margins should be recorded:
   - Margins → flat, rolled, raised, or everted.
   - Base → smooth, granulated, or covered with fibrin membrane or slough or hemorrhagic crust (scab) or fungating (some malignancies).
6. Lesion coloration.
   - Dark bluish swelling that blanches on pressure → vascular lesion
   - Lighter-colored, bluish lesion that does not blanch → mucus-retaining cyst.
   - Pigmented lesion within mucosa → tattoo of restorative material or melanotic tumor.
   - Keratinized white lesions → reaction to repetitive local trauma or premalignant changes.
   - Erythematous (or mixed red and white) →more ominous prognosis for dysplastic changes than a white lesion.
   - Inflammation can be superimposed on areas of mechanical trauma or ulceration→varied presentation from one examination to the next.
7. Sharpness of lesion borders and mobility: fixed to adjacent bone, arising from bone, and extending into adjacent soft tissues, or only infiltrating the soft tissue.
8. Consistency to palpation: soft or compressible (e.g., a lipoma or abscess), firm or indurated (e.g., a fibroma or neoplasm), or hard (e.g., torus or exostosis). Fluctuant: lesion with nonrigid walls that contains fluid.
9. Presence of pulsation → vascular (significant when dealing with Intrabony lesions). The pulsation can be accompanied by a palpable vibration, called a thrill →auscultation of the area with a stethoscope may reveal a bruit, or audible murmur. Invasive procedures on lesions with thrills, bruits, or both should be avoided, → referral to specialists (life-endangering hemorrhage if biopsied).
10. Examination of regional lymph nodes:
    - Should be accomplished before any biopsy (Sometimes lymphadenitis develops following biopsy→diagnostic dilemma).

- In adults, normal lymph nodes are not palpable unless they are enlarged by inflammation or neoplasia, but cervical nodes of up to 1 cm in diameter can be palpated in children up to the age of 12 years.
- In recording lymph node, the following five characteristics documented: (1) location, (2) size (preferably recording the diameters in centimeters), (3) presence of pain or tenderness, (4) degree of fixation (fixed, matted, or movable), and (5) texture (soft, firm, or hardened).
- When multiple nodes are slightly enlarged but barely palpable, they can feel like bird shot and are described as "shotty nodes."
- The lymph node examination should include the following groups:

(1) occipital,
(2) preauricular and postauricular
(3) mandibular, submandibular, and submental
(4) deep anterior cervical chain
(5) superficial cervical nodes
(6) deep posterior cervical chain
(7) supraclavicular nodes.

*Buccal lymph nodes may or may not be palpable.

Presumptive Clinical Differential Diagnosis

After completing the initial dental, medical, clinical, radiographic, and laboratory examinations (as indicated), the dentist next should compile a presumptive list of differential diagnoses. These diagnoses convey the clinician's impression to the pathologist of what the lesion most likely is based on the total assessment. These diagnoses are important because the pathologist rules out entities that may have similar clinical and pathologic presentations.

General Principles of Biopsy

A biopsy is more likely to rule out malignancy than to diagnose cancer because most oral and odontogenic lesions are benign. The four major types of biopsies performed in and around the oral cavity: (1) cytologic biopsy, (2) incisional biopsy, (3) excisional biopsy, and (4) aspiration biopsy.

Incisional Biopsy

Removes small portion of a lesion. If the lesion is

- large (>1 cm in diameter)
- demonstrates differing characteristics in different locations → more than one area of the lesion may require sampling.

1- Excised as a wedge to include normal- and abnormal-appearing tissues
2- Central areas of a large lesion often necrotic and offer little diagnostic value
3- Active growth is at the perimeter; therefore, its inclusion with normal-appearing tissue demonstrates cellular changes.

4- Include an adequate depth of tissue so that cellular features from the base of the lesion are included. Generally, it is better to take a narrow, deep specimen than a broad, shallow one.
5- Care should be taken not to compromise adjacent anatomic structures unless they seem to have a relationship with the origins or pathology

Excisional Biopsy
Removal of a lesion in its entirety, to include a 2 to 3 mm perimeter of normal tissue around the lesion, ***indicated for lesion sized 1 cm or less and it is clinically benign***. The width of the perimeter of normal tissue may vary, depending on the presumptive diagnosis. An additional 2 to 3 mm in tissues may be required for specimens suspected of malignancy. Complete excision often constitutes **definitive treatment** of the lesion biopsied.

Aspiration Biopsy
Performed with a needle and syringe by penetrating a suspicious lesion and aspirating its contents. Two main types of aspiration biopsy in clinical practice are:

1) Biopsy to explore whether a lesion contains a fluid. Routine aspiration of intraosseous radiolucent lesions is also performed before entering into the bony defect to rule out the potential of the lesion being vascular in origin and to define whether it is cystic or solid.
2) Biopsy to aspirate cells for pathologic diagnosis (fine-needle aspiration) and is often performed by pathologists and used when
   - Soft tissue mass is detected beneath the skin or mucosal surface and the patient wishes to avoid a scar or adjacent anatomic structures pose a risk.
   - Diagnosis of neck masses from which it can be difficult to obtain a biopsy.

Aspiration is performed on any fluid-filled lesion except a mucocele. A 16- to 18-gauge needle connected to an aspirating syringe is used. The needle tip may have to be repositioned repeatedly to locate a suitable fluid-containing cavitation.

Indications for Biopsy:
1. Any persistent pathologic condition that cannot be clinically diagnosed
2. Any lesion that is felt to have malignant or premalignant characteristics.
3. Confirmation of clinical diagnostic suspicions
4. Any lesion that doesn't respond to routine clinical management (i.e., removal of local irritant) over a 10- to 14-day period.
5. Any lesion that is the basis of extreme concern to the patient (cancer phobia)

Characteristics of Lesions that Raise Suspicion of Malignancy
- Bleeding: lesion bleeds on gentle manipulation
- Duration: lesion has persisted more than 2 weeks
- Erythroplasia: totally red lesion or has a speckled red and white appearance.

- Fixation: lesion feels attached to adjacent structures
- Growth rate: lesion exhibits rapid growth.
- Induration: lesion and surrounding tissue is firm to the touch
- Ulceration: lesion is ulcerated or presents as an ulcer

Prebiopsy Monitoring

Any undiagnosed or suspicious change in oral tissues that cannot be explained by localized trauma (and the source corrected) or other factors should be followed up in 7 to 14 days, with or without local treatment. If the lesion enlarges or expands, develops an altered appearance, or does not respond as expected to local therapy, then biopsy is usually indicated.

Soft Tissue Biopsy Techniques and Surgical Principles:

Anesthesia

- Block local anesthesia techniques are preferred over infiltration.
- Anesthetic solution should not inadvertently be incorporated in the surgical specimen → distortion of the cellular architecture of the specimen and make pathologic diagnosis more difficult.
- Peripheral infiltration of local anesthetic with a vasoconstrictor is often helpful (injected at least 1 cm away from the lesion perimeter)
- The vasoconstrictor will decrease hemorrhaging in the wound and improve the surgeon's visibility during surgery.

Tissue Stabilization

Oral soft tissue biopsies involve mobile structures; incisions placed with ease when the involved tissues are first stabilized, accomplished by

- Assistant can grasp the lips on both sides of the biopsy site with fingers, reducing bleeding by compressing area blood vessels.
- Retractors can perform the same function.
- Towel clips, Adson (fine tip) forceps, chalazion forceps.
- Heavy retraction suture: placed deeply into the tissues, away from the planned biopsy site (avoid damaging the tissues).

Hemostasis

The use of a suction device should be minimized (increase bleeding + accidental aspiration of sample)

- The assistant can use gauze sponges to blot the site, or
- Place a gauze pad over the end of the suction.

Incisions

- Sharp scalpel, usually No. 15 blade

- Two football-shaped surface incisions angled to converge at the base:
  1- Yield an optimal specimen. 2- Wound is easy to close.
- The use of laser and electrosurgical equipment for biopsies incisions is not desirable (distort histologic architecture)
- Variations in the size of the ellipse and degree of convergence toward the base of the lesion depend on the depth of the lesion into normal tissues.
- Palpation offers clues regarding depth of submucosal portions of the lesion.
- Ensure a perimeter of normal tissue **beneath** the lesion.
- Thin, deep specimens are preferable to wide, shallow ones.
- To minimize secondary injuries and for esthetic reasons; incisions made to parallel the normal course of, nerves, blood vessels, and lines of muscular tension (i.e., smile lines and facial creases)
- About 2- to 3-mm band of normal tissue included around the specimen
- Additional 2 to 3 mm of normal-appearing peripheral tissue excised if the lesion appears:
  a- Malignant
  b- Pigmented
  c- Vascular or
  d- Has diffuse borders.
- In larger lesions with variable surface characteristics; incisional biopsy taken from different areas.

Wound Closure

- Primary closure of the wound is desirable.
- If the wound is deep, incorporating different tissue layers, **deep closure** should be carried out for each layer, using a resorbable suture material (e.g., polyglycolic acid or chromic gut).

Closure of deeper tissues:

1- Mucosa is undermined using a spreading action of the tips of small scissors (e.g., Iris or Metzenbaum scissors) to separate the mucosal from the submucosal tissues (Submucosal layer loose connective tissue easily dissected without sharp incision). This permits closure of the mucosa as a separate layer without regard to closure in the deeper layers (permits tension-free approximation).

2- The extent to which this undermining is carried out is determined by the size of the wound and the anatomic location. In the lips, cheek, floor of mouth, and soft palate, margins undermined in all directions by a distance that is at least the width of the defect.

- Suture materials of choice; black silk or a nonreactive, slowly resorbable material (polyglycolic acid “Dexon”) or polyglactin 910 (Vicryl) sutures.
- Wounds on attached mucosal surfaces generally not closed (heal by secondary intention). Protective periodontal dressings or acrylic splints lined with a tissue-conditioning liner used to
  A- Protect this area(s)

B- Enhance patient comfort.
C- Promote healing.

*Postsurgical splints may be secured to adjacent teeth with circumdental wires or heavy suture material
*Postsurgical splints left in place for 7 to 10 days.

- Biopsy wounds on the dorsum or lateral border of the tongue require deeply placed sutures at close intervals to counteract muscle movements.
- Resorbable sutures may be used, but gut sutures are not recommended because they
  a- Have poor knot security (resulting in lost sutures)
  b- Undergo rapid enzymatic degradation.

Handling of Tissues; Specimen Care

Tissue specimen must be maintained for preserving the histologic architecture. Avoid surgical instrument damage to the specimen during manipulation.

1- Tissue sample should **not:**
   a- Be wrapped in gauze (wet or dry) (risk of getting thrown out accidentally) along with the gauze.
   b- Be allowed to dry out.
2- Specimen immediately placed in a container containing 10% formalin solution (4% formaldehyde) 20 times the volume of specimen.
3- The specimen totally immersed in the preservative solution at all times.
4- Suture Tagging of Specimens; Margin Identification
   A- If dysplasia or malignancy is suspected, "tags" a margin of a specimen with a loose suture to orient its anatomic alignment allowing precise pathologic report on areas require wider or deeper excision.
   B- Also used to identify multiple specimens from one lesion; The 1st specimen receives one tagging suture, the 2nd receives two, and so on (Each submitted in its own container).

INTRAOSSEOUS (HARD TISSUE) BIOPSY

- Often the cause of jaw lesion is odontogenic, and the lesion will resolve once the dental problem is addressed.
- If unrelated to the dentition or does not respond to treatment; should be removed for diagnosis.
- Most common are:
  a- Periapical granulomas
  b- Odontogenic cysts.
  Treatment of these two lesions

a) Surgical removal of the lesion by way of excisional biopsy (enucleation).
b) If large, perforating into soft tissue or where a suspicion of malignancy; incisional biopsy.

TECHNIQUES AND PRINCIPLES

I. Palpate the area of the jaw and compare it with the contralateral side.
A- Firm and smooth bone suggests lesion has not expanded or eroded cortical plate.
B- Spongy; indicates erosion or thinning of the cortical plate; aggressive neoplastic lesion.

II. **Mucoperiosteal Flaps**

The flap should

- Follow basic principles of flap design like removing a tooth.
- Avoid major neurovascular structures.
- Rest entirely on sound bone for closure, 4 to 5 mm beyond the surgical margins of bony defects.
- Be approached in an area away from the lesion margins over sound bone when elevated in eroded cortical bone.
- Be full thickness (through mucosa, submucosa, and periosteum).

III. Precautionary Aspiration

To determine whether lesion contains fluid:

1- After achieving local anesthesia
2- A 16- or 18-gauge needle connected to a 5- or 10-ml syringe.
3- Penetrate cortical plate.
   a- By pressing the needle firmly through the mucoperiosteum with a twisting movement
   b- If not possible to penetrate, a flap is reflected and a large round burr, under constant irrigation, is used to penetrate the cortical plate.
4- The needle tip may be repositioned if an area of fluid initially not located.
   a- Inability to aspirate fluid suggests solid tumor.
   b- If straw-colored fluid is aspirated, suggest cyst (enucleate)
   c- If pus is aspirated, an inflammatory or infectious process.
   d- Aspiration of air without any fluid is suggestive of a traumatic cavity.
   e- If blood is aspirated, several diagnoses:
      i- Pulsatile vascular lesion (e.g., hemangioma or arteriovenous malformation); life-threatening
      ii- Aneurysmal bone cyst
      iii- Central giant cell lesions

5- Lesion aspirant may be submitted for chemical analysis, microbiologic culturing, and even microscopic evaluation.
6- If no aspirant is found→incisional biopsy on the soft tissue mass within the bone

IV. Osseous Window

Creation of a cortical window for access.

1- If the cortical plate is intact, a round surgical burr under constant fluid irrigation can be used over the lesion site
2- If erosion of cortical plate (osseous defect) is noted when flap is elevated; defect enlarged with rongeurs or a round surgical burr
3- The size of the window depends on
a- Size of the lesion
b- Proximity of significant anatomic structures
4- Enlarged window with a rongeur, as necessary, for access.
5- The removed bone should be submitted along with the primary specimen.

V. Specimen Management

1. Small lesions that have a connective tissue capsule (e.g., cysts) are **enucleated** in their entirety:
   A. A dental curette is used to peel the specimen away from bone and dentition.
   B. Instrument kept constantly in contact with the osseous surface.
   C. When lesion completely freed; placed immediately into the formalin.
   D. If lesion does not separate easily; exact location(s) of adherence noted on the submission form.
   E. The resulting bone cavity irrigated, suctioned, and examined for residual soft tissue. If any noted; curetted out.
   F. Mucoperiosteal flap is repositioned and sutured.
2. Solid soft tissue lesion of small size that **separates readily**:
   A. Curetted and enucleated in the same manner as the cystic lesion.
   B. If resistance to curettage noted→ remove a millimeter of adjacent osseous tissue after the bulk of the lesion is removed.
   C. Tooth root surfaces within the bone defect thoroughly curetted.
   D. If incisional biopsy is indicated, a section of tissue is removed, and the remaining lesion is left undisturbed.
3. Diagnostic radiographs included if possible.

Post biopsy Follow-Up.

If the lesion is thought to be benign, then routine follow-up is carried out, with periodic radiographs to monitor osseous healing. If an incisional biopsy was performed, once the microscopic diagnosis becomes available, plans should be

formulated for any required definitive treatment, referral for additional treatment, or both.

Notes

- A negative biopsy report should never be taken at face value but interpreted with clinical and historical findings in mind. If doubt exists, a **second** biopsy might be indicated. At the very least, plans for continued close observation at appropriate intervals (within 1 month and then at 3, 6, and 12 months during the first year. Thereafter, if clinical and radiographic findings are unchanged, the interval between follow-up visits can be increased to 6 and then 12 months)
- Patients should always be counseled to contact the dentist immediately if any changes are noted between visits.