

CHAPTER 1

INTRODUCTION TO SALIVARY GLAND LESIONS CYTOLOGY

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1.1 INTRODUCTION

The salivary glands are part of the exocrine secretory apparatus that are traditionally considered part of the upper gastrointestinal tract. They are a very small organ with an average total weight of 50 g in adults compared with other systems. They are composed of two major groups: the major and minor salivary glands. The major glands are composed of three paired relatively larger glands: the parotid, submandibular, and sublingual. The minor group is numerous and widely distributed in the upper aerodigestive tract (Figure 1.1).

1.2 BASIC HISTOLOGY AND PHYSIOLOGY

Salivary glands secrete digestive enzymes from their main functional unit “the acinus.” The major histological components of salivary glands are as follows (Figure 1.2 illustrates these components and their cytological correlates):

1. **Acinus:** The main functional unit that is composed of wedge-shaped cells, each with abundant cytoplasm pushing a small round-to-oval nucleus to its periphery. They can be serous where they mainly secrete amylase, and their cytoplasm appears basophilic and densely granular with zymogen granules. These granules are periodic acid Schiff

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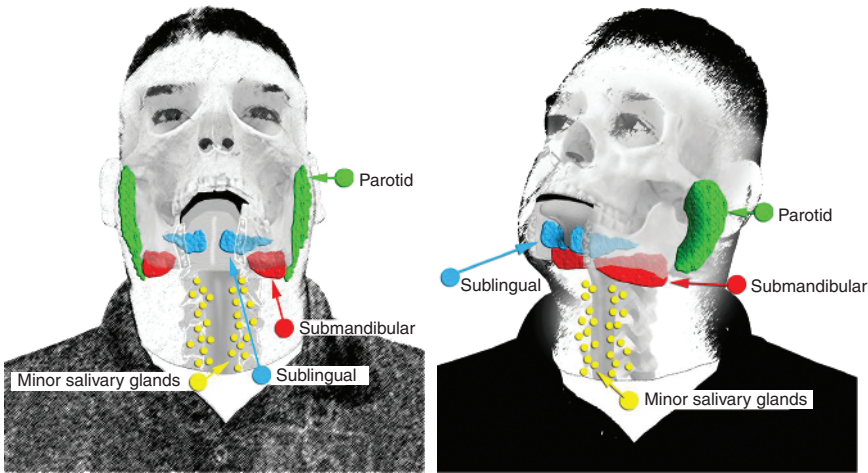


FIGURE 1.1. A diagram showing the general anatomy of salivary glands. The major salivary glands are composed of three paired, relatively larger glands: the parotid, submandibular, and sublingual. The minor salivary gland groups are numerous and widely distributed in and around the upper aerodigestive tract and are predominantly in the submucosal areas.

positive and diastase resistant. The mucinous acini secrete sialomucin, and their cytoplasm appears clear with vacuoles. The parotid gland is almost purely serous, whereas both the submandibular and the sublingual are mixed. The submandibular is more serous, and the sublingual is more mucinous.

2. **Ducts:** They start as small, intercalated ductules between acinar cells that are lined by single, small cuboidal cells with relatively large, centrally located nuclei. These are difficult to see on histological sections. These ductules will then join and form larger, striated ducts lined by taller columnar cells with much more abundant and eosinophilic cytoplasm rich in mitochondria. These in turn will join larger interlobar excretory ducts lined by pseudo-stratified columnar epithelium with similar features.
3. **Myoepithelial cells:** These stellate-shaped cells are contractile and are located outside the basement membrane of the acinar cells. They contain smooth muscle actin, myosin, and intermediate filaments such as keratin. They are difficult to see histologically.

1.3 DISEASES THAT AFFECT SALIVARY GLANDS

Many diseases can affect salivary glands. The common entities range from inflammatory/infectious non-neoplastic lesions, benign neoplasms, and

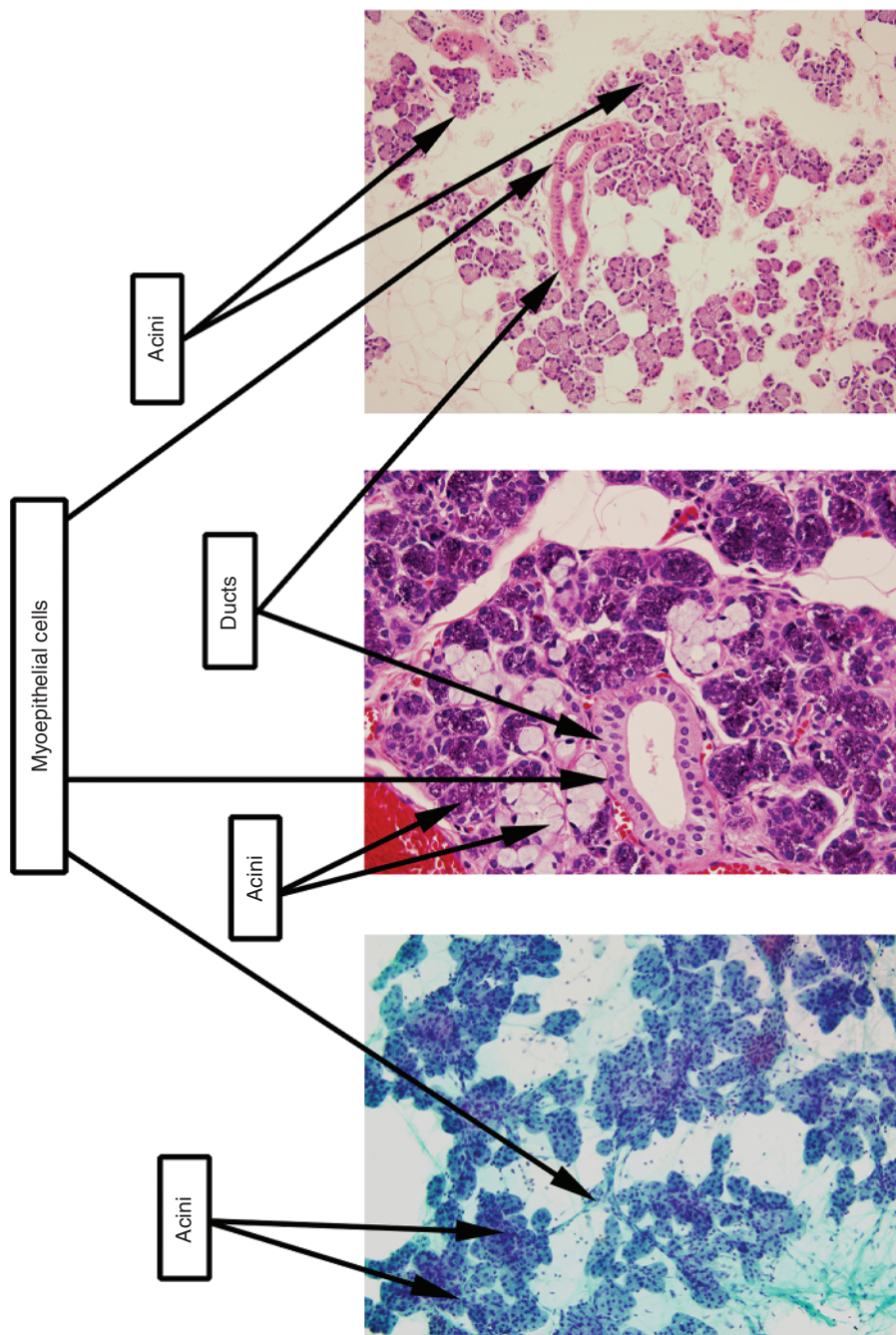


FIGURE 1.2. The three images were combined to show the three major cellular components of salivary glands. The left image is from an aspirate smear (Papanicolaou stain, 200 $\times$); the middle image is the corresponding histological section (hematoxylin and eosin, 400 $\times$); and the right image is from a cell block (hematoxylin and eosin, 200 $\times$).

TABLE 1.1. Questions to be answered when evaluating aspirates from salivary gland masses

1. Is the lesion a salivary gland lesion?
2. Is the lesion neoplastic?
3. If neoplastic, is it benign or malignant?
4. If malignant, is it high grade or low grade?
5. Can the diagnosis be specific?

malignant tumors. With an active otolaryngology service, pathologists are frequently asked to perform or interpret fine needle aspirates from salivary gland masses. Most mass lesions suspected to develop from salivary glands pose diagnostic challenges and are aspirated to determine the underlying disease process. Masses of the parotid gland are the most frequent. In these circumstances, the major questions that face pathologists are summarized in Table 1.1. Chapter 2 was written by an oncologic otolaryngology surgeon (Dr. Tulunay-Ugur) and clearly illustrates the preoperative approach of these tumors and lesions and what the surgeon would like to see in the fine needle aspiration report.

1.4 EPIDEMIOLOGY OF SALIVARY GLAND TUMORS

Despite its small size, tumors of the salivary glands are numerous and they characteristically exhibit a relatively significant degree of overlap on both morphologic and cytologic grounds. The most recent World Health Organization (WHO) list of primary tumors included 10 benign epithelial tumors, 24 malignant epithelial tumors, 1 soft tissue benign tumor (hemangioma), and lymphomas (Table 1.2). Secondary and metastatic tumors can occur, but they are less frequent and most are secondary to other head and neck neoplasms. Benign tumors are much more common than malignant ones, and the parotid gland is the most frequently involved. In addition, it is well known that the relative frequency of malignancy is inversely proportional to gland size. Therefore, malignant tumors approximately comprise 25% of parotid gland tumors, 45% of submandibular gland tumors, 80% of sublingual tumors, and 50% of minor salivary gland tumors. Therefore, extra attention has to be paid to the salient features of malignancy on cytological grounds when dealing with sublingual and minor salivary gland masses. Most tumors that occur in the floor of the mouth, the tongue, and the retro molar areas are essentially malignant. There are well-known geographic variations and gender disparities. However, these will be tackled in the following chapters. In the United States, malignancies of salivary glands comprise approximately 6% of all head and neck cancers and less than 1% of all malignancies.

TABLE 1.2. Salivary gland tumors^a**Benign epithelial tumors:**

Pleomorphic adenoma
 Warthin's tumor
 Myoepithelioma
 Basal cell adenoma
 Sebaceous adenoma
 Lymphadenoma (Sebaceous and nonsebaceous)
 Canalicular adenoma
 Oncocytoma
 Cystadenoma
 Ductal papilloma (intraductal papilloma, inverted ductal papilloma, sialadenoma papilleferum)

Malignant epithelial tumors:

Mucoepidermoid carcinoma
 Acinic cell carcinoma
 Adenoid cystic carcinoma
 Carcinoma ex pleomorphic adenoma
 Polymorphous low-grade adenocarcinoma
 Epithelial-myoepithelial carcinoma
 Basal cell adenocarcinoma
 Salivary duct carcinoma
 Oncocytic carcinoma
 Myoepithelial carcinoma
 Clear cell carcinoma, not otherwise specified
 Metastasizing pleomorphic adenoma
 Small cell carcinoma
 Squamous cell carcinoma
 Lymphoepithelial carcinoma
 Sialoblastoma
 Large cell carcinoma
 Cystadenocarcinoma
 Low-grade cystadenocarcinoma
 Mucinous adenocarcinoma
 Sebaceous carcinoma
 Sebaceous lymphadenocarcinoma
 Carcinosarcoma
 Adenocarcinoma, not otherwise specified

Soft tissue tumors: *hemangioma*

Hematolymphoid tumors: *Hodgkin's lymphoma, diffuse large cell lymphoma, extranodal marginal zone lymphoma*

Metastatic tumors

^a Adapted from the most recent WHO classification. Lyon: IARC Press; 2005.

Overall, the most common tumor type is pleomorphic adenoma followed by Warthin's tumor. Mucoepidermoid carcinoma is the most frequent carcinoma followed by acinic cell carcinoma and adenoid cystic carcinoma. In general, tumors can occur at any age with a wide age variation; detailed descriptions of each entity will be demonstrated in the following chapters. It is important to mention that mucoepidermoid and acinic cell carcinoma are two malignancies that not uncommonly may occur in children. Figure 1.3 demonstrates major salivary gland tumors and their similarity of different cell components.

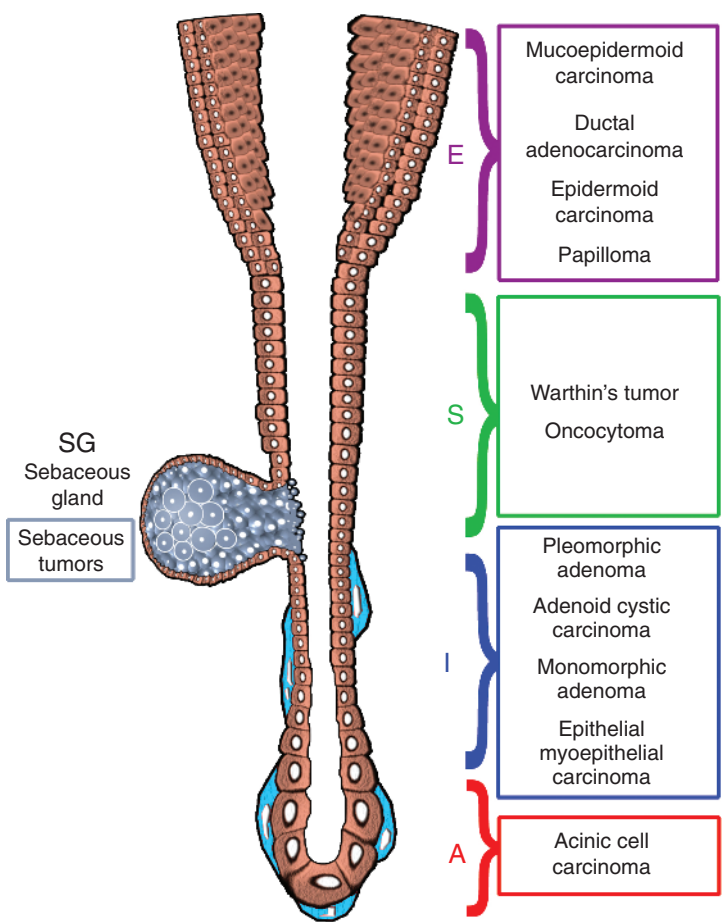


FIGURE 1.3. An illustration that shows some of the common tumors and potential similarity and proposed cell of origin. (Modified with permission from Histology for Pathologists, 2nd ed., Edited by Stephen S. Sternberg, Lippincott Williams & Wilkins, page 423.)

1.5 INDICATIONS FOR SALIVARY GLAND FINE NEEDLE ASPIRATION AND PRACTICAL CONSIDERATIONS

There is still some resistance against the concept of pretreatment fine needle aspiration (FNA) of unexplained salivary gland masses. On the one hand, the opponents of FNA argue that significant diagnostic overlap occurs among different salivary gland lesions and, ultimately, that surgical removal will be needed. On the other hand, the proponents believe that specific accurate FNA diagnosis can be reached in most cases and that surgery can be avoided in many circumstances. Scenarios where surgery can be avoided include inflammatory diseases, lymphomas, metastases, or benign neoplasms in otherwise elderly patients with multiple comorbidities. In addition, it is safe to say that even if there is no specific diagnosis, the preliminary cytological impression in most cases help guide the surgeon to determine what type of salivary gland surgery the patient will have (radical or simple excision).

Complications from salivary gland FNA are rare, and when they occur, they are not serious. Bleeding, infection, and pain from facial nerve trauma are among the most frequent. FNA-induced infarction has been reported and most commonly affects Warthin's tumor and oncocytic neoplasms. Tumor seeding is an extremely rare occurrence. No salivary gland FNA adequacy criteria have been established yet.

1.6 ACCURACY OF SALIVARY GLAND FINE NEEDLE ASPIRATION

The accuracy of FNA of salivary gland lesions is variable and depends on multiple variable factors. According to older data, the rate of correctly establishing a malignant or benign neoplasm can be achieved in more than 80% of the cases, whereas reaching a specific diagnosis ranges between 60% and 75% of the cases. However, in more recent data, the accuracy is higher where both sensitivity and specificity approaches more than 90%. The accuracy depends on multiple factors that include aspirator experience, availability of clinical and radiological data about the lesion, and using different types of stains of the aspirated smears. Reaching a specific diagnosis may not be possible in all cases; however, a major, categorical, nonspecific diagnosis would be extremely helpful for the treating clinicians. A diagnosis of "negative for neoplasm," "benign neoplasm," "low-grade carcinoma," or "high-grade malignancy/carcinoma" are extremely informative to surgeons. Although the accuracy of fine needle aspiration is variable and in some reports was not high enough to be acceptable, we believe that it is the best initial diagnostic approach. False-negative diagnosis can occur and results from sampling issues or interpretation. False-positive diagnosis also can occur but less frequently than false-negatives and mostly from overcalling atypia. The

TABLE 1.3. Recommendations that increase accuracy of salivary gland aspiration

1. The clinical and radiological data should be known before the procedure
2. Multiple passes should be performed in different directions (Figure 1.4)
3. Multiple stains should be used (Diff-Quik, Papanicolaou, and hematoxylin and eosin)
4. Conventional smears are preferred
5. Reaspiration of cystic lesions should be performed before its collapse (Figure 1.5)
6. Mild atypia can be seen in pleomorphic adenoma (the most common tumor of salivary glands)
7. Ancillary studies should be performed when needed at the time of aspiration (such as culture and flow cytometry)

value of a frozen section for salivary gland lesions is controversial, and in general, it is well accepted that a preoperative FNA is superior. We strongly believe that the sensitivity and specificity may be enhanced if certain precautions and steps are followed. These recommendations are as follows (Table 1.3):

1. It is advisable to have the radiological images conducted before the aspiration. The radiological information are very helpful for the aspirator; they confirm the exact location of the lesion and determine whether the lesion is in the salivary gland and shows the relation of the lesion with the surrounding structures. Chapter 3 includes more details. Furthermore, the availability of clinical data especially when the pathologists themselves perform the procedure and communicate with the patient adds an important dimension.
2. In most circumstances, one pass may not be adequate. Therefore, multiple passes in different directions are highly recommended to help sample as much as possible from the lesion. From our experience, two to four passes is usually adequate (Figure 1.4).
3. Utilization of multiple different stains is extremely critical. As many salivary gland tumors contain stroma, the presence of air-dried type smears such as Diff-Quik stain is very important. In our practice, we use Diff-Quik stain on the initial air-dried smears, Papanicolaou stain for the rest of smears, and always try to prepare the cell block. The final product that will be evaluated will include smears stained with Diff-Quik stain, Papanicolaou stain, and hematoxylin and eosin stain. Combining all these stains into a single case is valuable in interpreting salivary gland lesions.
4. Despite the shortage of data regarding using liquid-based smears in salivary gland aspiration, we strongly believe that direct conventional smears are preferred and superior.

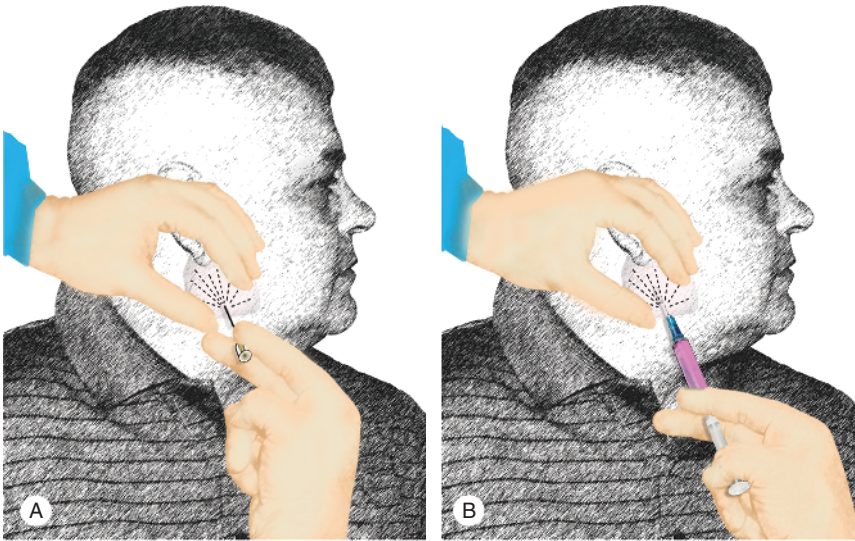


FIGURE 1.4. Diagram demonstrates the techniques we use to perform fine needle aspiration. (a) We prefer to start with aspiration using a needle without syringe or suction (also known as the French technique or Zajdela technique). The advantage of this technique is providing a nice, thin smear with less crush artifacts enabling the interpreter of optimum cytological morphology to proceed with appropriate triage. (b) The following passes can be used by employing a syringe with suction using negative pressure to increase cellularity. The aspiration can be performed with or without commercially available “guns” depending on the aspirator preference.

5. Reaspiration of cystic lesions while keeping the needle of the first pass inside the mass is a very helpful trick that helps sample the wall of the lesion and is believed to increase sensitivity (Figure 1.5).
6. Pleomorphic adenoma aspirates may show a mild degree of atypia.
7. A proper medium and tubes may be needed for ancillary studies, such as culture and immunophenotyping for lymphoid lesions.

Despite the aforementioned discussion, some lesions always pose diagnostic challenges and are problematic. The list includes basaloid tumors, lymphomas, low-grade mucoepidermoid carcinoma, acinic cell carcinoma, carcinoma ex pleomorphic adenoma, and myoepithelial cell tumors. Additionally, although not absolutely required by surgeons, establishing a specific diagnosis when dealing with high-grade carcinoma smears is sometimes impossible. These issues will be discussed in details in the following chapters.

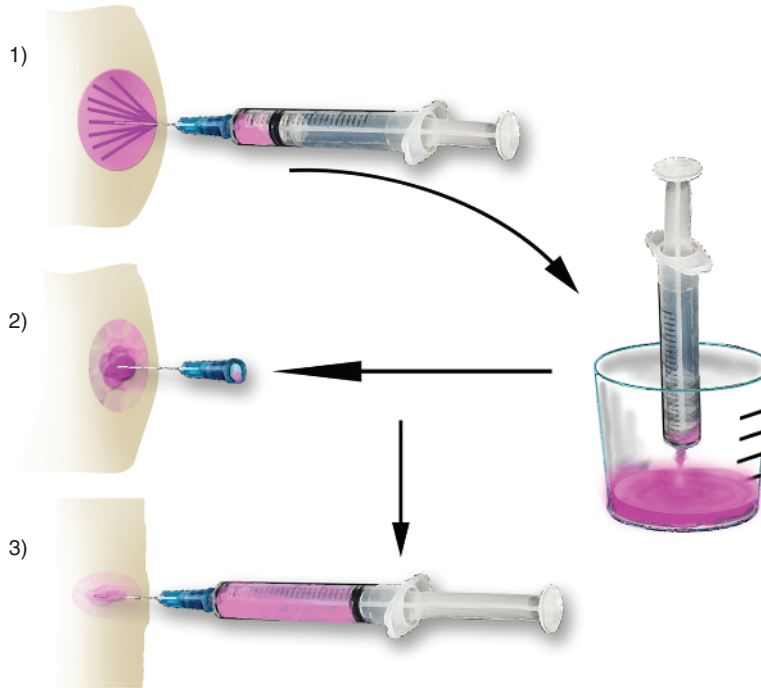


FIGURE 1.5. A diagram that shows the steps that are used when aspirating a cystic mass. Reaspiration of cystic lesions while keeping the needle of the first pass inside the mass is a very helpful trick that assist in sampling the wall of the lesion and is believed to increase sensitivity.

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