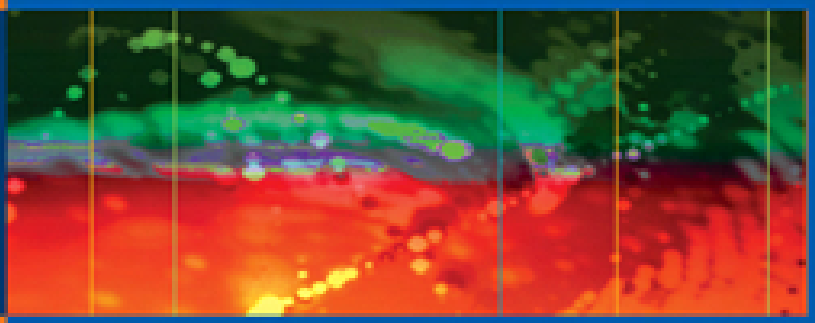
William C. Faquin
Celeste N. Powers



Salivary Gland Cytopathology

Essentials in Cytopathology
Series Editor
Dorothy L. Rosenthal

Springer

Faquin • Powers
Salivary Gland Cytopathology

Salivary Gland Cytopathology

ESSENTIALS IN CYTOPATHOLOGY SERIES

Dorothy L. Rosenthal, MD, FIAC, Series Editor

Editorial Board

Syed Z. Ali, MD
Douglas P. Clark, MD
Yener S. Erozan, MD

---

1. D.P. Clark and W.C. Faquin: Thyroid Cytopathology. 2005
ISBN 0-387-23304-0

2. D. L. Rosenthal and S.S. Raab: Cytologic Detection of Urothelial Lesions. 2005
ISBN 0-387-23945-6

3. D.C. Chhieng and E.B. Stelow: Pancreatic Cytopathology. 2007
ISBN 978-0-387-68946-3

4. S.Z. Ali and A.V. Parwani: Breast Cytopathology. 2007
ISBN 978-0-387-71594-0

5. W.C. Faquin and C.N. Powers: Salivary Gland Cytopathology. 2008
ISBN 978-0-387-76622-5

William C. Faquin, MD, PhD

Department of Pathology, Massachusetts General Hospital,
Harvard Medical School, Boston, Massachusetts

Celeste N. Powers, MD, PhD

Department of Pathology, Medical College of Virginia Hospitals,
Virginia Commonwealth University, Richmond, Virginia

Salivary Gland Cytopathology

Foreword by Mary K. Sidaway, MD

Springer

William C. Faquin, MD, PhD
Associate Professor of Pathology
Department of Pathology
Massachusetts General Hospital
Harvard Medical School
Boston, Massachusetts 02114
USA

Celeste N. Powers, MD, PhD
Professor of Pathology
and Otolaryngology
Department of Pathology
Medical College
of Virginia Hospitals
Virginia Commonwealth
University
Richmond, Virginia 23298
USA

Series Editor
Dorothy L. Rosenthal, MD, FIAC
Professor of Pathology, Oncology, and Gynecology/Obstetrics
The Johns Hopkins School of Medicine
Baltimore, MD 21287
USA

ISBN: 978-0-387-76622-5 e-ISBN: 978-0-387-76623-2
DOI: 10.1007/978-0-387-76623-2

Library of Congress Control Number: 2007938596

Printed on acid-free paper

9 8 7 6 5 4 3 2 1

springer.com

Foreword

Salivary gland neoplasms display a striking range of morphologic diversity that can confound cytologic as well as histologic interpretation. The overlapping cytologic features, the lack of markers to accurately distinguish various tumors, and the overemphasized FNA-induced histologic changes have fostered uncertainty in the role of FNA in the management of salivary gland lesions.

Despite its limitations in the classification of salivary gland neoplasms, FNA is superior to core biopsy, and its accuracy is at least comparable to frozen sections. In the hands of cytopathologists equipped with awareness of the diagnostic challenges, FNA of salivary glands is a reliable technique that can be used as a diagnostic tool or as a screening tool to triage patients into the most appropriate management scheme.

Classifying salivary gland neoplasms may be difficult, as the majority of the tumors arise from epithelial and myoepithelial cells that also have the ability to undergo a variety of metaplastic changes (squamous, mucinous, sebaceous, oncocytic, and chondroid). When a specific diagnosis cannot be made with a high level of certainty, the lesion is best reported by generating a differential diagnosis. Some experts advocate classifying salivary gland lesions into diagnostic categories based on the cell type and extracellular material.

This book, written by two experienced authors, will fulfill the need of those who wish to familiarize themselves with the FNA of salivary glands. Recognizing that the interpretation of FNA of salivary gland lesions can be fraught with challenges and pitfalls, the authors of *Salivary Gland Cytopathology* present a simple and practical algorithm to classify these tumors based on the overall cytomorphologic pattern of the aspirate (cellular, inflammatory,

and cystic). The “cellular” category is further divided into seven subcategories, each of which is discussed in a separate chapter. The book also provides in-depth discussions and exquisite images of the more unusual appearances and less frequently encountered neoplasms. The information is presented in a concise format, yet knowledge is imparted without sparing any detail. This book will not only serve as a basic comprehensive review but as a valuable reference to be consulted whenever diagnostic difficulties confront the reader. The authors are to be congratulated for producing a valuable addition on this important and complex topic. They have indeed done an excellent job of covering and clearly illustrating the critical aspects of every conceivable differential diagnostic entity and pitfall.

Mary K. Sidaway, M.D.

Series Preface

The subspecialty of cytopathology is 60 years old and has become established as a solid and reliable discipline in medicine. As expected, cytopathology literature has expanded in a remarkably short period of time, from a few textbooks prior to the 1980's to a current and substantial library of texts and journals devoted exclusively to cytomorphology. *Essentials in Cytopathology* does not presume to replace any of the distinguished textbooks in cytopathology. Instead, the series will publish generously illustrated and user-friendly guides for both pathologists and clinicians.

Building on the amazing success of *The Bethesda System for Reporting Cervical Cytology,* now in its second edition, the *Series* will utilize a similar format, including minimal text, tabular criteria, and superb illustrations based on real-life specimens. *Essentials in Cytopathology* will, at times, deviate from the classic organization of pathology texts. The logic of decision trees, elimination of unlikely choices, and narrowing of differential diagnosis via a pragmatic approach based on morphologic criteria will be some of the strategies used to illustrate principles and practice in cytopathology.

Most of the authors for *Essentials in Cytopathology* are faculty members in The Johns Hopkins University School of Medicine, Department of Pathology, Division of Cytopathology. They bring to each volume the legacy of John K. Frost and the collective experience of a preeminent cytopathology service. The archives at Hopkins are meticulously catalogued and form the framework for text and illustrations. Authors from other institutions have been selected on the basis of their national reputations, experience. and enthusiasm for cytopathology. They bring to the series complementary

viewpoints and enlarge the scope of materials contained in the photographs.

The editor and authors are indebted to our students, past and future, who challenge and motivate us to become the best that we possibly can be. We share that experience with you through these pages, and hope that you will learn from them as we have from those who have come before us. We would be remiss if we did not pay tribute to our professional colleagues, the cytotechnologists and preparatory technicians who lovingly care for the specimens that our clinical colleagues send to us.

And finally, we cannot emphasize enough throughout these volumes the importance of collaboration with the patient care team. Every specimen comes to us as a question begging an answer. Without input from the clinicians, complete patient history, results of imaging studies and other ancillary tests, we cannot perform optimally. It is our responsibility to educate our clinicians about their role in our interpretation, and for us to integrate as much information as we can gather into our final diagnosis, even if the answer at first seems obvious.

We hope you will find this series useful and welcome your feedback as you place these handbooks by your microscopes, and into your book bags.

Dorothy L. Rosenthal, M.D., FIAC
Baltimore, Maryland
drosenthal@jhmi.edu
July 15, 2004

Contents

1
Introduction to FNA and Salivary Gland Neoplasia

Salivary gland fine-needle aspiration (FNA) represents one of the most challenging areas of cytopathology. This is due in part to the wide range of lesions, both reactive and neoplastic, that can be encountered in the more than 500 salivary glands present in the human body. In fact, histologically (and cytologically), salivary gland tumors have been described as one of the most heterogeneous groups of human tumors; over 40 subtypes of neoplasms have been reported. An additional challenge for salivary gland FNA is the significant cytomorphologic diversity and overlap between many benign and malignant salivary gland tumors. In general, ancillary marker studies are of limited value in solving this problem because many salivary gland tumors have a similar composition of epithelial and myoepithelial cells. With all of these hurdles to overcome, it is quite impressive that salivary gland FNA emerges as an accurate and effective tool for diagnosing this complex group of lesions. Currently, FNA has gained wide acceptance as a first-line procedure in the evaluation of a salivary gland mass. In this book, we use an algorithmic approach (see Chapter 3) to guide you through the principles of salivary gland cytology, pointing out pitfalls, limitations, and differential diagnoses along the way, to allow you to use salivary gland FNA accurately and effectively.

FNA was first described in 1847 by Kun, and later, in the 1930's, it was reintroduced and promoted by Martin and Ellis. While FNA remained popular in Europe and particularly in Scandinavian countries, it is only in the past 30 years that it has seen a resurgence in popularity in the United States. Currently, FNA is used extensively throughout the country for the assessment of both palpable and

deep-seated lesions in most anatomic sites. FNA is performed manually by cytopathologists, clinicians, and surgeons for the assessment of palpable masses, and it is performed under ultrasound or CT-guidance for nonpalpable deep-seated or cystic lesions. It is considered standard of care to use FNA as an initial step in the evaluation of any thyroid nodule, and more and more it is being used to provide a preoperative diagnosis and thereby guide the clinical management of patients presenting with a salivary gland mass.

Indications and Contraindications for Salivary Gland FNA

At the majority of medical institutions throughout the country, FNA is used in conjunction with both clinical and radiologic findings in the initial evaluation of most swellings in the major as well as minor salivary glands. Patients presenting for FNA often complain of a mass and/or pain in the face, upper neck, or mouth, or in some cases, partial paralysis or paresthesia of the face. Contraindications to FNA are uncommon but include a bleeding diathesis, the presence of a pre-existing skin infection overlying the area, and an extremely uncooperative or agitated patient. In addition, clinicians will occasionally send patients for an FNA who do not have a palpable (or radiologically detectable) mass, and we would discourage performing an FNA in these cases since it has the potential to lead to a false negative diagnosis due to sampling issues. FNA is a simple and inexpensive procedure that yields valuable clinical information and, in our opinion, should be performed routinely in the initial evaluation of most salivary gland masses.

In conjunction with clinical and radiologic findings, FNA should be performed routinely during the initial evaluation of a salivary gland mass.

Contraindications to Salivary Gland FNA

- Bleeding diathesis
- Skin infection at FNA site
- Extremely uncooperative or agitated patient
- Absence of a detectable mass

Benefits of Salivary Gland FNA

There are many benefits to using FNA in the evaluation of a salivary gland mass. Salivary gland FNA is easily performed, minimally invasive, safe, cost effective and accurate, provides a rapid diagnostic interpretation (usually within 15–30 minutes), and can easily be used to obtain material for special ancillary studies. Depending upon the type of lesion present, salivary gland FNA provides important diagnostic information about the mass that will impact the subsequent clinical management of the patient. For example, in some cases, the lesion may be non-neoplastic, possibly obviating the need for surgical intervention, or in other cases it may be a benign, low-grade or high-grade neoplasm each of which falls into a different category of clinical management (see Chapter 2). In the case of lymphoid lesions (see Chapter 5), FNA can provide essential material for ancillary immunophenotyping and/or molecular analysis which is critical for the proper evaluation, subtyping, and treatment of lymphomas. When there is a clinical suspicion that the mass is due to infection, salivary gland FNA can provide material for microbiologic cultures and Gram stains. Overall, FNA is a very safe, minimally invasive procedure that rarely has complications. Even without the use of a local anesthetic, FNA is usually associated with only minimal discomfort to the patient. Relative to larger core biopsies, FNA avoids the possible risk of nerve damage, and it averts the risk of tumor spread due to seeding of the biopsy tract. With regard to cost effectiveness, salivary gland FNA is inexpensive to perform. Compared to open surgical biopsy, it has been estimated that substantial savings can be achieved using FNA, in part by sparing up to one-third of patients unnecessary surgery.

Benefits of Salivary Gland FNA

- Safe
- Cost effective
- Accurate
- Rapid assessment
- Provides information for preoperative strategy
- Avoids unnecessary surgery in some patients
- Provides material for ancillary studies (e.g. flow cytometry)
- Provides material for microbiologic cultures
- Complications are rare

Potential Complications Associated with Salivary Gland FNA

Despite its widespread use and the many benefits of salivary gland FNA as outlined above, it still does not have universal acceptance. In part this is due to the belief by some that FNA of a salivary gland neoplasm will lead to seeding by tumor along the needle track. This concern has largely been disproven in various reports, including a study with an extensive literature search that found only 2 cases of needle track seeding, and both were from Trucut biopsies. Others contend that FNA of pleomorphic adenomas will lead to an increased rate of recurrence. This too has been shown to be false in a large series by Engzell and colleagues. Overall, complications from FNA are infrequent. Minor complications secondary to the FNA procedure itself include a risk of hemorrhage resulting in a localized bruise, but this can be avoided in most cases by careful application of firm pressure to the area after each FNA pass. An occasional patient will experience a syncopal episode in response to the FNA procedure, and the FNA clinician should be prepared for this possible scenario by quickly adjusting the patient to the Trendelenburg position at the first sign of syncope. Infection is rarely encountered as a complication of salivary gland FNA, but just as with venipuncture, the possibility exists. Following standard sterile practices, including preparation of the FNA site with alcohol, reduces the risk of infection. Occasionally short-term facial nerve pain has been reported; however, FNA does not carry the risk of permanent facial nerve damage as can occur using large-bore core biopsies.

Finally, potential histologic changes have been reported secondary to FNA of some salivary gland tumors. While these are rare, the pathologist should be aware of them to avoid potential diagnostic errors such as misinterpreting FNA-induced pseudocapsular invasion as true invasion, or extensive squamous metaplasia as carcinoma. Most histologic changes are only focal, and include hemorrhage, inflammation with multinucleated giant cell reaction, squamous metaplasia, subepithelial stromal hyalinization, and formation of granulation tissue. Probably the

most frequently discussed histologic change is the potential for complete tumor necrosis secondary to infarction, which would hinder the final pathologic interpretation. This is rare, but when it does occur, it is most common in oncocytic lesions such as oncocytoma and Warthin tumor, but it has also been described in pleomorphic adenomas.

Possible Complications of Salivary Gland FNA

- Local hemorrhage
- Infection
- Facial nerve pain
- Syncope
- FNA-induced histologic changes:
  - Tumor infarction
  - Granulation tissue formation
  - Squamous metaplastic changes
  - Inflammation with giant cell reaction
  - Subepithelial stromal hyalinization

Common Diagnostic Challenges in Salivary Gland Cytopathology

Salivary gland cytopathology is a diagnostically challenging area in part because of the wide variety of neoplasms arising in the salivary glands and the overlapping cytomorphologic features of so many of these tumors. As they say, “forewarned is forearmed”; by being aware of certain specific problem areas in salivary gland cytology, one can more readily avoid diagnostic errors. Within the context of the algorithm presented in Chapter 3 and applied within subsequent chapters of this book, we will address specific problem areas in detail. Below is a list of some of the most common problem areas of salivary gland FNA where the cytologist should be particularly cautious.

Diagnostically Challenging Areas of Salivary Gland Cytology

- **Lymphoid lesions**: lymphoepithelial sialadenitis (LESA) vs lymphoma
- **Matrix-containing lesions**: pleomorphic adenoma versus adenoid cystic carcinoma
- **Basaloid neoplasms**: basal cell adenoma and basal cell adenocarcinoma versus solid variant of adenoid cystic carcinoma
- **Oncocytic lesions**: Warthin tumor and oncocytoma versus acinic cell carcinoma
- **Mucinous cysts**: low-grade mucoepidermoid carcinoma versus mucocele
- **High-grade carcinomas**: high-grade mucoepidermoid carcinoma versus salivary duct carcinoma and metastasis
- **Spindle cell lesions**: schwannoma vs myoepithelioma
- **Clear cell tumors**: epithelial-myoepithelial carcinoma vs myoepithelioma

Accuracy of Salivary Gland FNA

Many salivary gland FNA series have been reported, and they all tend to agree that FNA is an accurate diagnostic test (Table 1.1). When one analyzes the data, however, what we find is a more complex picture: salivary gland FNA is highly accurate for distinguishing benign from malignant lesions, and non-neoplastic from neoplastic, but it is much less precise for making a specific diagnosis. In general, salivary gland FNA has an overall accuracy of 81%–98%. The range in accuracy reflects differences in the method of classifying lesions and analyzing data, as well as more practical

TABLE 1.1. Accuracy of salivary gland FNA.

Statistical Measurement	Percent (%)
Overall accuracy	81–98
Accuracy for benign vs malignant	81–100
Accuracy for specific diagnosis	48–94
False negative rate	1–15
False positive rate	5–8
Sensitivity	86–100
Specificity	90–100

variables such as the experience of the cytopathologist and the quality of the FNA specimen. For benign versus malignant salivary gland lesions, the accuracy of FNA ranges from 81%–100%. In contrast, the accuracy of salivary gland FNA when used to specifically subtype a neoplasm is only 48%–94%. As we will discuss and illustrate in subsequent chapters, FNA is remarkably precise at diagnosing neoplasms such as pleomorphic adenoma and Warthin tumor, but it is less precise for diagnosing certain tumors such as basal cell adenocarcinoma or epithelial-myoepithelial carcinoma, among others. **A key in the assessment of any salivary gland FNA is to be aware of which lesions can be specifically diagnosed by FNA, and which require a descriptive report** (Table 1.2).

The reported sensitivity of salivary gland FNA in most series ranges from 86% to 100%, and the specificity ranges from 90%–100%. False negative and false positive diagnoses are low. The range of false negative diagnoses is 1%–15%, and most cases were due to misdiagnosis of low-grade malignancies and hypocellular cysts such as low-grade mucoepidermoid carcinoma (Table 1.3).

TABLE 1.2. Ability of salivary gland FNA to make a specific versus descriptive diagnosis.

Usually Specific Diagnosis	Sometimes Specific Diagnosis	Usually Descriptive Diagnosis
Pleomorphic adenoma	Adenoid cystic carcinoma	Basal cell adenoma, tubulotrabecular and solid types
Warthin tumor	Low-grade mucoepidermoid carcinoma	High grade mucoepidermoid carcinoma
Acute and chronic sialadenitis	Carcinoma ex-pleomorphic adenoma	Salivary duct carcinoma
Basal cell adenoma, membranous type	Metastasis	Polymorphous low-grade adenocarcinoma
Reactive lymph node	Small cell carcinoma	Basal cell adenocarcinoma
Lymphoma	Mucocele	Epithelial-myoepithelial carcinoma
	Oncocytoma	
	LESA	
	Acinic cell carcinoma	

TABLE 1.3. Common causes of false positive and false negative salivary gland FNAs.*

False Positive	False Negative
Monomorphic adenoma	Lymphoma
Intraparotid lymph node	Acinic cell carcinoma
Oncocytoma	Low-grade mucoepidermoid carcinoma
Granulomatous sialadenitis	Adenoid cystic carcinoma

* American Pathologist Interlaboratory Comparison Program in Nongynecologic Cytology (Arch Pathol Lab Med 2005;129:26–31).

The false positive rate of salivary gland FNA ranges from 5%–8%, and is most often due to overcall of benign lesions with reactive and metaplastic changes particularly in the setting of inflammation. In a recent report from the College of American Pathologist Interlaboratory Comparison Program in Nongynecologic Cytology, false positive diagnoses were most commonly due to misdiagnosis of monomorphic adenoma, intraparotid lymph node, and oncocytoma. False negative diagnoses were most common for lymphoma, acinic cell carcinoma, low-grade mucoepidermoid carcinoma, and adenoid cystic carcinoma.

FNA has been compared to frozen section, which is also often used in the evaluation of salivary gland tumors. Relative to the frozen section diagnosis of salivary gland tumors, FNA has a similar accuracy, but it also has the important added advantage of providing a preoperative rather than intraoperative diagnosis. FNA tends to be more sensitive than frozen section, while the latter is often more specific and has the advantage of being able to assess margins. In one study by Seethaia and colleagues, FNA and frozen section, as expected, were complementary, with frozen section being most useful for assessing nondiagnostic FNA cases and for confirming malignancy in some cases.

Background to Salivary Gland Neoplasia

Salivary gland neoplasms are among the most heterogeneous group of tumors of any tissue in the body. This array of tumors includes those arising within the salivary gland parenchyma (sialomas), those derived from the supporting connective tissues of the salivary gland

(synsialomas), and those from the surrounding extrasalivary gland tissue (parasialomas). The sialomas represent the largest, most common, and most clinically important of the 3 groups. While definitive evidence is lacking, one of the most popular theories of salivary gland tumorigenesis suggests that epithelial tumors of the sialoma group arise from basal reserve cells of the intercalated or excretory ducts.

Relative to others, salivary gland neoplasms are rare, representing about 0.6% of all tumors. The frequency and types of salivary gland neoplasms vary markedly between the major (parotid, submandibular, and sublingual glands) and the minor salivary glands. The overall global incidence of salivary gland neoplasia has been reported to range from 0.4 to 13.5 cases per 100,000 per year, and in western countries the rate is estimated at 2.5 to 3.0 cases per 100,000 per year. Fifty-four percent to 79% of salivary gland tumors are benign, and the majority (65%–80%) occur in the parotid gland. Among the minor salivary glands, most tumors occur in the oral cavity, with the palate being the most common site.

The incidence of salivary gland malignancy in the United States is estimated at 1.2 per 100,000 per year or about 3,300 new salivary gland cancers annually. Salivary gland cancers represent approximately 6% of head and neck cancers and about 0.3%–0.5% of all malignancies. The rate of salivary gland cancer, including cases with advanced disease, is reported to be increasing, but the reasons for this are unknown. The cancer rate within the major and minor salivary glands can generally be equated to the size of the gland, with 15%–32% of parotid tumors, 35%–50% of

TABLE 1.4. Statistics of salivary gland neoplasia.*

Statistical Measure	Value
Western incidence of salivary gland neoplasia	2.5–3.0 cases per 100,000
U.S. incidence of salivary gland malignancy	1.2 cases per 100,000
Overall rate of malignancy among salivary gland tumors	21%–46%
Most common site of neoplasia	Parotid gland
Parotid cancer rate	15%–32%
Submandibular cancer rate	35%–50%
Sublingual cancer rate	70%–90%
Minor salivary gland cancer rate	>50%

*Statistics derived from several sources including the National Cancer Institute website for salivary gland cancer (see “Suggested Reading”).

submandibular tumors, and 50%–90% of sublingual and minor salivary gland tumors being malignant. With regard to absolute numbers of salivary gland cancers, the parotid gland is the most common anatomic site, followed by the submandibular gland, palate, cheek, and tongue.

Histologic Types of Salivary Gland Neoplasia

With so many types and subtypes of salivary gland tumors, the nomenclature can be challenging. A summary of the 2005 WHO nomenclature for benign and malignant salivary gland tumors is listed in Tables 1.5 and 1.6. Histologically, pleomorphic adenoma is by far the most common salivary gland tumor, comprising 50%–60% of all salivary gland neoplasms, and approximately 60%–80% of parotid gland neoplasms (Table 1.7). Warthin tumor (aka. Warthin's tumor) is usually cited as the second most common neoplasm and comprises 3.5%–14% of all salivary gland tumors. In children, salivary gland tumors are rare, representing less than 3% of all salivary gland tumors. The most common salivary gland lesions in infants are nonepithelial neoplasms, particularly hemangiomas. Among malignant salivary gland tumors, mucoepidermoid carcinoma is the most common in children and adults as well as in both major and minor salivary glands. Mucoepidermoid carcinoma accounts for approximately 10%–15% of all salivary gland tumors and about 35% of all malignant salivary gland neoplasms.

TABLE 1.5. WHO nomenclature for benign salivary gland tumors.

Benign Tumors
Pleomorphic adenoma
Myoepithelioma
Basal cell adenoma
Warthin tumor
Oncocytoma
Canalicular adenoma
Sebaceous adenoma
Lymphadenoma
Ductal papilloma
Cystadenoma
Hemangioma

TABLE 1.6. WHO nomenclature for malignant salivary gland tumors divided by tumor grade.*

Low Grade	Intermediate Grade	High Grade
Mucoepidermoid carcinoma	Mucoepidermoid carcinoma	Mucoepidermoid carcinoma
Acinic cell carcinoma	Adenoid cystic carcinoma	Salivary duct carcinoma
Polymorphous low-grade adenocarcinoma	Adenocarcinoma, NOS	Carcinoma ex-pleomorphic adenoma
Epithelial-myoepithelial carcinoma	Myoepithelial carcinoma	Carcinosarcoma
Basal cell adenocarcinoma	Sebaceous carcinoma	Squamous cell carcinoma
Sialoblastoma		Small cell carcinoma
MALT lymphoma		Lymphoepithelial carcinoma
Clear cell carcinoma, NOS		Diffuse large B-cell lymphoma
Cystadenocarcinoma		Adenocarcinoma, NOS
Adenocarcinma, NOS		Large cell carcinoma
		Oncocytic carcinoma

* Tumors are divided into 3 histologic grades, but this subcategorization is variable for some tumors.

TABLE 1.7. Approximate percentages of the most common salivary gland neoplasms.

Salivary Gland Tumor Type	Percentage (%)
Pleomorphic adenoma	50–60
Warthin tumor	5–15
Mucoepidermoid carcinoma	10–15
Adenocarcinoma, NOS*	9
Acinic cell carcinoma	6
Adenoid cystic carcinoma	4–10

* The percentage of adenocarcinoma, NOS is highly variable due to inconsistent reporting and variations in diagnosis of this entity.

Risk Factors for the Development of Salivary Gland Cancer

Risk factors for the development of salivary gland cancer are not well documented and many are controversial, but several potential risk factors have been identified. Probably one of the most

significant risks for the development of head and neck neoplasia is exposure to ionizing radiation, particularly among patients receiving radiation treatment involving the head and neck. Workers in certain industries, including rubber products manufacturing, plumbing, and woodworking, or with exposure to certain substances, including asbestos, nickel alloy, chromium, or silica dust, have also been reported to have an increased risk of developing salivary gland cancer. Smoking has been indicated as a risk factor for head and neck squamous cell carcinoma, possibly including primary cases in the salivary gland, and it has been strongly implicated in the development of Warthin tumors. However, neither smoking nor alcohol use have shown a definite link to other salivary gland cancers based upon a case/control study. Finally, except for an increased risk of salivary gland basal cell tumors among patients with Brooke-Spiegler syndrome, inherited genetic factors do not appear to play a significant role in the development of salivary gland tumors.

Possible Risk Factors for Salivary Gland Neoplasia

- Ionizing radiation
- Rubber manufacturing
- Woodworking
- Asbestos
- Nickel alloy
- Chromium
- Silica dust

Patient Population and Presentation of Salivary Gland Neoplasia

Salivary gland neoplasia occurs over a wide age range. Some of the most common types of salivary gland tumors (e.g., pleomorphic adenoma and mucoepidermoid carcinoma) occur most frequently in the third to fourth decade, but overall, most types of either benign or malignant tumors occur in the fifth to seventh decades of life, with an average occurence at 46 to 47 years. Approximately one-third of salivary gland cancers are reported to occur in people under the age of 55, which is younger than for most other human cancers. Salivary gland neoplasia is more common in women than in men, but epidemiologic variations occur, depending upon the

specific tumor type. For example, Warthin tumor and high-grade carcinomas are more common in men than in women.

Most patients with a salivary gland tumor present with a slowly enlarging, painless, virtually asymptomatic mass. The problem is that these features characterize both benign tumors as well as most of the malignant ones. In part, this probably reflects the fact that a majority of malignant salivary gland tumors are low-grade carcinomas that are slow growing and lack perineural invasion. It is estimated that only 30% of salivary gland cancers are detected based upon clinical features. Approximately 10%–15% of patients with salivary gland cancer do present with a painful mass, and about 10% present with facial nerve weakness or paralysis, the latter being an ominous sign suggestive of an aggressive tumor. Other clinical signs of a potentially malignant salivary gland lesion include rapid increase in size, fixation to surrounding structures, skin involvement, paresthesias, hoarseness, and cervical lymphadenopathy.

Clinical Features of Malignant Salivary Gland Tumors

- Most are asymptomatic
- Rapid increase in size
- Pain
- Nerve paralysis or weakness
- Paresthesia
- Fixation to surrounding structures
- Skin involvement
- Hoarseness
- Cervical lymphadenopathy

Salivary Gland Cancer and Prognosis

The overall 5-year survival rate for patients with salivary gland cancer is 68% (see separate specific chapters for details about individual cancer types) (Table 1.8). The rate of 5-year survival, however, ranges from 5%–95%, depending upon a variety of factors including the gland involved, histologic tumor type, tumor grade, clinical stage, and whether the tumor involves the facial nerve or surrounding structures. Low-grade tumors tend to have a good to excellent prognosis with low mortality (5%–20%) and a tendency for local recurrence. In contrast, high-grade cancers usually present with an advanced clinical stage of

TABLE 1.8. Prognosis for salivary gland cancer.

Statistical Measure	Percentage (%)
Overall 5-year survival	68
Low-grade cancer mortality	5–20
High-grade cancer mortality	55–95

disease, and mortality ranges from 55%–95%. The site of origin of the salivary gland cancer is also an important predictor. Cancers arising in the parotid gland have the most favorable outcome, followed by the submandibular gland; the least favorable outcomes are associated with cancers in the sublingual and minor salivary glands. Overall, it can be said that the most important prognostic predictor for salivary gland cancer is clinical stage, especially tumor size. This is reflected in the staging by TNM classification as outlined by the American Joint Committee on Cancer (AJCC).

The most important prognostic predictor for salivary gland cancer is clinical stage, especially tumor size.

For cancers of the major salivary glands, a clinical staging system using the TNM classificataion scheme has been designed, and is based upon tumor size, local extraparenchymal extension (clinical or macroscopic evidence of soft tissue or nerve invasion), cervical lymph node involvement, and the presence of distant metastases. Tumors involving the minor salivary glands are staged based upon the specific anatomic site of origin.

Summary of AJCC TNM Classification of Salivary Gland Carcinomas (see AJCC Cancer Staging Manual for Full Details)

Primary Tumor (T)

- TX: Primary tumor cannot be assessed
- T0: No evidence of primary tumor
- T1: Tumor ≤ 2cm in greatest dimension without extraparenchymal extension
- T2: Tumor > 2cm but ≤ 4cm without extraparenchymal extension

- T3: Tumor > 4cm and/or extraparenchymal extension
- T4a: Tumor invades skin, ear canal, mandible, and/or facial nerve
- T4b: Tumor invades skull base and/or pterygoid plates and/or encases carotid artery

Regional lymph nodes (N)

- NX: Regional lymph nodes cannot be assessed
- N0: No regional lymph node metastasis
- N1: Metastasis to a single ipsilateral lymph node ≤ 3cm in greatest dimension
- N2: Metastasis in a single ipsilateral lymph node, > 3cm but ≤ 6cm in greatest dimension, or in multiple ipsilateral lymph nodes, ≤ 6 cm in greatest dimension, or in bilateral or contralateral lymph nodes, ≤ 6cm in greatest dimension (See AJCC Cancer Staging Manual for N2a, b, c details)
- N3: Metastasis in a lymph node > 6cm in greatest dimension

Distant Metastasis (M)

- MX: Distant metastasis cannot be assessed
- M0: No distant metastasis
- M1: Distant metastasis

AJCC STAGE GROUPINGS:

Stage I		
• T1	N0	M0
Stage II		
• T2	N0	M0
Stage III		
• T3	N0	M0
• T1,2,3	N1	M0
Stage IV A		
• T1,2,3	N2	M0
• T4a	N0,1,2	M0
Stage IV B		
• T4b	Any N	M0
• Any T	N3	M0
Stage IV C		
• Any T	Any N	M1

Suggested Reading

Barnes L, Eveson JW, Reichart P, Sidransky D. World Health Organization Classification of Tumours: Pathology and Genetics of Head and Neck Tumours. Lyon: IARC Press, 2005.

Engzell V, Esposti DL, Rubio C, Sigurdson A, Zajicek J. Investigation of tumor spread in connection with aspiration biopsy. Acta Radiol Ther 1971;10:385–398.

Frable WJ. Thin-needle aspiration biopsy. Am J Clin Pathol 1976;65:168–181.

Li S, Baloch ZW, Tomaszewski JE, LiVolsi VA. Worrisome histologic alterations following fine-needle aspiration of benign parotid lesions. Arch Pathol Lab Med 2000;124:87–91.

Hughes JH, Volk EE, Wilbur DC. Pitfalls in salivary gland fine-needl aspiration cytology: Lessons from the College of American Pathologists Interlaboratory Comparison Program in Nongynecologic Cytology. Arch Pathol Lab Med 2005;129:26–31.

Kakagia D, Alexiadis G, Kiziridou A, Lambropoulou M. Brooke-Spiegler syndrome with parotid gland involvement. Eur J Dermatol 2004;14:139–141.

Major salivary glands (parotid, submandibular, and sublingual). In: American Joint Committee on Cancer: AJCC Cancer Staging Manual. 6th ed. New York: Springer, 2002; 53–58.

Mukunyadzi P. Review of fine-needle aspiration cytology of salivary gland neoplasms, with emphasis on differential diagnosis. Am J Clin Pathol 2002;118S:S100–115.

Muskat JE, Wynder EL. A case/control study of risk factors for major salivary gland cancer. Otolaryngol Head Neck Surg 1998;118:195–198.

National Cancer Institute website for salivary gland cancer: http://www.cancer.gov/cancertopics/pdq/treatment/salivarygland/healthprofessional

Powers CN. Complications of fine needle aspiration biopsy: the reality behind the Myths. In: Schmidt WA (ed), Cytopathology, Vol.1, ASCP Reviews in Pathology. Chicago:ASCP Press, 1996;69–96.

Rimm DL, Stastny JF, Rimm EB, Ayer S, Frable WJ. Comparison of the costs of fine-needle aspiration and open surgical biopsy as methods for obtaining a pathologic diagnosis. Cancer Cytopathol 1997;81:51–56.

Seethaia RR, LiVolsi VA, Baloch ZW. Relative accuracy of fine-needle aspiration and frozen section in the diagnosis of lesions of the parotid gland. Head & Neck 2005;27:217–223.

2
Salivary Gland FNA: Anatomic, Clinical, and Technical Considerations

Anatomic Considerations

The major salivary glands consist of the parotid, the submandibular, and the sublingual glands. Under the control of the parasympathetic nervous system, the major salivary glands are responsible for the principal portion of saliva produced, which can be as much as 1.5 liters per day. The minor salivary glands of the oral cavity, pharynx, and upper airways contribute only a small percentage of the overall volume of saliva, but they are particularly important for supplying the mucus layer that protects the tissues of the oral cavity and upper respiratory tract.

Embryologically, the major salivary glands develop during the 6th to 8th weeks of gestation. The parotid gland arises first from ingrowth of the oral ectoderm into the surrounding mesenchyme. By the 7th week of development, the parotid gland moves in a dorsal and lateral direction to reside in the preauricular region, and by the 10th week, the facial nerve divides the surrounding parotid gland into anatomic superficial and deep compartments. A majority of parotid gland tumors develop within the superficial lobe, making FNA and clinical management relatively easy. Tumors originating within the deep lobe of the parotid gland usually present as pharyngeal swellings due to expansion into the parapharyngeal space. The parotid gland is unique among the salivary glands in that it incorporates lymphoid tissue during development, sometimes with entrapment of salivary gland epithelial cells. The latter are believed to be the source of lesions such as Warthin tumor and lymphoepithelial cysts. The minor

salivary glands develop after the major glands and derive from the oral ectoderm and the nasopharyngeal endoderm.

The adult parotid gland, which weighs approximately 15 g, is enclosed in a fibroadipose tissue capsule, and has as its anatomic borders the masseter muscle anteriorly, the zygomatic arch superiorly, the external auditory canal posteriorly, and the styloid process, styloid muscle, and great vessels inferiorly (Fig. 2.1). The tail of the parotid extends over the mastoid tip and lies over the sternocleidomastoid muscle. The main excretory duct of the parotid gland is known as Stenson's duct, which courses along the masseter muscle and the buccal fat pad, and then passes through the buccinator muscle before opening into the oral cavity near the second maxillary molar. Accessory parotid gland tissue is found in approximately

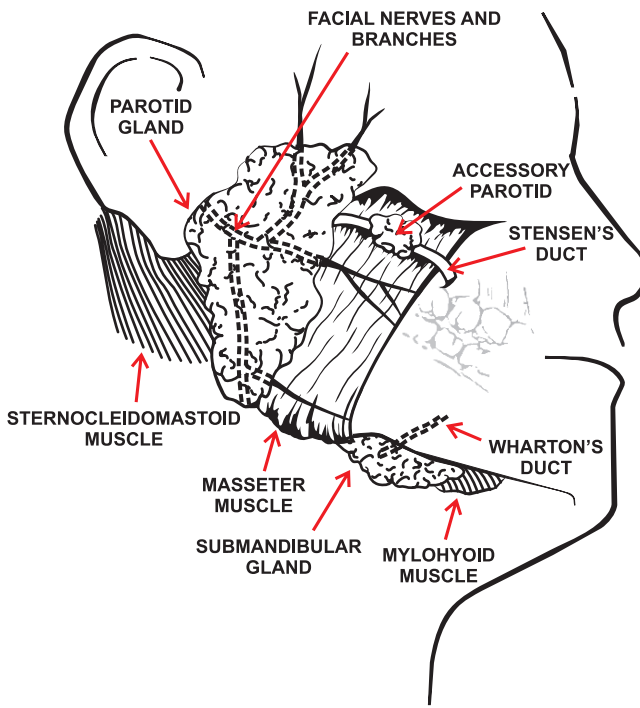


FIG. 2.1. Anatomic relationship of the major salivary glands.

20% of individuals and can occur along the anterior surface of the parotid gland as well as along the length of Stenson's duct. Blood supply to the parotid gland is from branches of the external carotid artery. Lymphatics to the paraparotid lymph nodes derive from the temporal region, scalp, auricle, eyelids and lacrimal glands, while the intraparotid lymph nodes drain the nasopharynx, soft palate, middle ear, and external auditory canal. Parotid gland lymphatics drain into the superficial and deep cervical lymph nodes.

The submandibular gland and the sublingual gland, unlike the parotid gland, are derived from endodermal tissues. The submandibular gland (aka submaxillary gland) weighs 7–8 g, and resides within the submandibular triangle formed by the anterior and posterior bellies of the digastric muscle and the inferior margin of the mandible (Fig. 2.1). The gland is divided into superficial and deep lobes by the mylohyoid muscle, with the deep lobe being the largest. The main excretory duct of the submandibular gland is Wharton's duct, which empties into the oral cavity on the anterior floor of mouth. The blood supply is from submental branches of the facial artery, and lymphatic drainage is to the deep cervical and jugular lymph nodes. The sublingual gland is the smallest of the major glands and weighs only 3 g. It rests in the sublingual fossa of the mandible bounded by the genioglossus and mylohyoid muscles (Fig. 2.1). There are multiple small excretory ducts from the sublingual gland that may connect directly to the oral cavity or join to form Bartholin's duct, which usually empties into Wharton's duct. The vascular supply comes from the sublingual and submental arteries, and lymphatic drainage goes to the submandibular lymph nodes.

Clinical Considerations

Diagnostic Workup and Physical Examination

For a patient presenting with a major salivary gland mass, the clinical workup includes:

- Complete history and physical examination
- Imaging studies (sialography, ultrasound, CT, or MRI)
- FNA
- Summary evaluation and possible presurgical planning

A complete history and focused physical examination is performed to assess the extent of salivary gland disease. Key points to cover include the approximate length of time that the lesion has been present, rate of change in the size of the mass (rapid or slow), associated fever or pain, drug exposures, history of malignancy and chronic illnesses such as rheumatologic disease or sicca syndrome (dry eyes, mouth). Physical examination of a salivary gland lesion is best accomplished with the patient comfortably seated with adequate back and head support. After carefully examining and palpating the mass and its relation to surrounding head and neck structures, particular emphasis is given to evaluating the patient for the presence of trismus, status of facial nerve function, presence of hypesthesia or anesthesia of the skin of the face or neck, and any otologic or oral findings. In addition, the entire neck should be palpated to detect any cervical lymphadenopathy.

Key Physical Exam Findings for a Major Salivary Gland Mass

- Size of mass
- Relationship to surrounding structures (e.g., fixation to skin)
- Trismus
- Facial nerve dysfunction
- Hypesthesia or anesthesia of skin
- Cervical lymphadenopathy

Imaging Studies

When imaging studies are used, they are most often obtained prior to the performance of an FNA since the latter has the potential to introduce traumatic changes in the tissue. A variety of imaging techniques are available for evaluating a major salivary gland mass, including sialography, ultrasonography, computed tomography (CT) scanning, and magnetic resonance imaging (MRI), with the latter two being the most popular. Sialography is applied to a limited extent for evaluation of ductal disease such as calculi, obstruction, and penetrating trauma. Ultrasonography can be used for superficial masses to distinguish extrinsic from intrinsic lesions, and it has also been used to direct FNA for difficult-to-palpate and complex cystic masses. It has the advantage of being inexpensive

and free of complications, but is limited to superficial masses and by its lack of anatomic detail.

High-resolution imaging such as CT or MRI is most commonly used in current practice and is essential for those salivary gland masses with clinical findings suggestive of malignancy, for deep-seated lesions, and for tumors of the submandibular or minor salivary glands. CT scanning is the most cost-effective imaging study for the evaluation of both intrinsic and extrinsic parotid masses; however, it is not very useful for the assessment of general parenchymal disease or ductal architecture. CT can be applied with or without simultaneous sialography and intravenous contrast enhancement (Fig. 2.2). In addition to its value in assessing salivary gland neoplasia, CT is excellent for the detection of salivary gland calculi, and it can also be applied to the evaluation of cystic

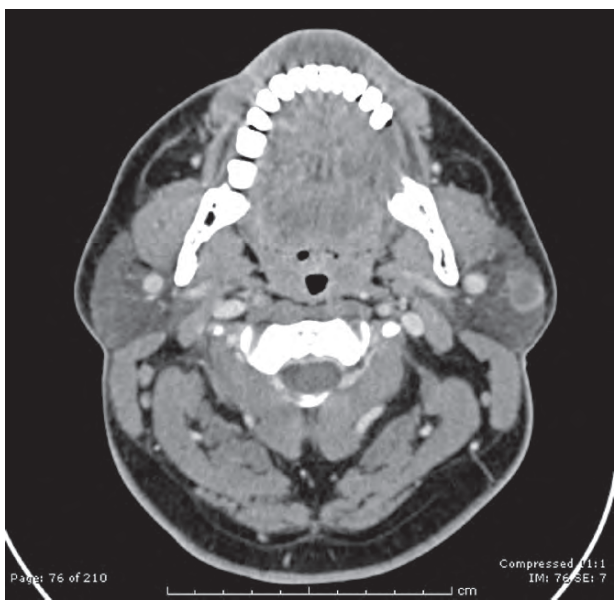


FIG. 2.2. Axial CT with intravenous contrast of a superficial left parotid gland tumor. The mass measures 1.2 cm, has sharp margins, and shows slight enhancement. Cytologic evaluation of the mass revealed a pleomorphic adenoma.

lesions. In addition, CT is often combined with FNA to sample tumors, particularly those that are deep-seated, and at our institutions this is done in conjunction with a cytopathologist who provides a "rapid interpretation" for assessing sample adequacy and guiding ancillary studies.

MRI is currently the method of choice for the evaluation of salivary gland lesions (Fig. 2.3), especially those involving soft tissues, and with gadolinium-enhancement, MRI is considered equal or superior to contrast-enhanced CT. Contraindications to the use of MRI include patients with pacemakers and

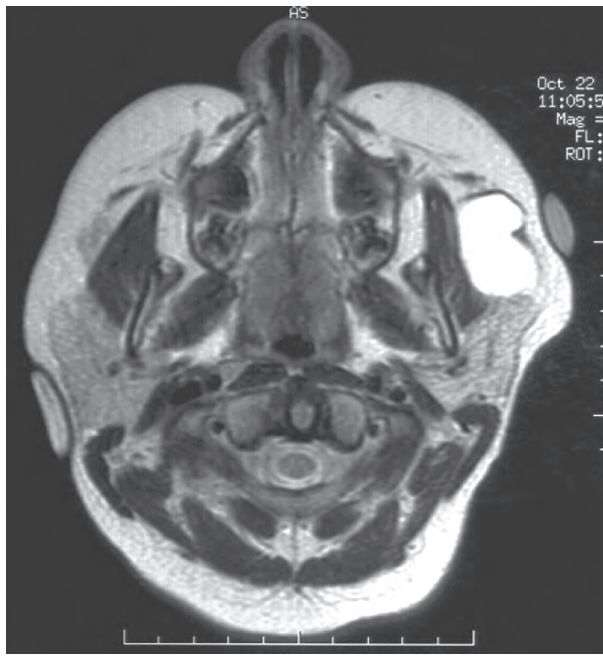


FIG. 2.3. MRI with contrast reveals a 2.8 cm well circumscribed, cystic mass superficial to the left masseter muscle and anterior to the left parotid gland along the tract of Stenson's duct. FNA revealed an acinic cell carcinoma that on surgical resection was present within accessory parotid gland tissue of the cheek.

those patients who are agitated or unable to be compliant during the imaging procedure. MRI is considered quite sensitive to the presence of masses within the salivary gland, but less sensitive to inflammatory disorders, and insensitive to calcifications. In addition, MRI is less sensitive to the detection of cystic lesions than is CT. For lesions within or near bone, CT and MRI are complementary.

FNA and the Clinical Management of Salivary Gland Lesions

The role of FNA as a diagnostic test for lesions of the salivary gland is strongly rooted in the clinical management algorithm for salivary gland tumors. While a detailed discussion of the management of salivary gland neoplasia is beyond the scope of this book, some general statements can be made regarding treatment options that have implications for FNA evaluation. For inflammatory and other non-neoplastic causes of salivary gland enlargement, nonsurgical management can often be used. Thus, when properly combined with clinicoradiologic findings and an adequate sample, FNA can, in a subset of cases, obviate the need for surgical intervention. When the mass is malignant, FNA can influence the clinical management by distinguishing between primary and metastatic disease. The most common clinical scenario, however, is a primary mass lesion for which surgery will almost certainly be performed, but the extent of the surgery will depend upon a number of factors, such as the grade and stage of the tumor. For benign and low-grade tumors, surgery alone is usually the treatment of choice. Particularly since a majority of parotid gland tumors arise in the superficial lobe, treatment will usually entail a simple superficial parotidectomy with negative margins. Postoperative radiation therapy is considered for those cases where resection margins are positive. In contrast, high-grade or high-stage tumors are managed by radical surgery that may include sacrifice of the facial nerve and lymph node dissection. Since FNA is highly accurate at distinguishing between low- and high-grade neoplasms, it is very useful for guiding the preoperative planning for such cases.

FNA and the Clinical Management of Salivary Gland Masses

- Primary versus metastatic disease (metastatic workup)
- Non-neoplastic salivary gland enlargement (nonsurgical treatment)
- Benign and low-grade tumors (limited surgery)
- High-grade tumors (radical surgery ± nerve sacrifice ± LN dissection)

Technical Aspects of Salivary Gland FNA

Fine-needle aspiration of the salivary gland is similar to FNA of palpable lesions in other anatomic sites. Probably the most important aspect of performing a good salivary gland FNA is adequate sampling and appropriate sample preparation. An adequate sample that includes both air-dried and alcohol-fixed preparations and good clinicoradiologic correlation are the keys to success in salivary gland FNA. In this context, many different "FNA styles" can be used to obtain a similar FNA result, and it will be up to each individual through experience to develop a style that is comfortable.

An adequate sample that includes both air-dried and alcohol-fixed preparations, and good clinicoradiologic correlation, are the keys to success in salivary gland FNA.

FNA Equipment

The standard equipment that we use in the performance of a salivary gland FNA includes 25-gauge sterile needles of either 1″ or 1-1/2″ length attached to a 10 cc syringe and a Cameco syringe pistol or other suitable syringe holder. Of note, some aspirators, including those at our institutions, prefer to use a needle and 10 cc syringe without a holder and without applying negative pressure in what we refer to as the "pencil or French technique." Other materials used during the FNA procedure include 95% ethanol in Coplin jars for slide fixation, normal saline, Hank's buffered saline, or RPMI in 1.5 ml tubes for rinsing the needle, glass "plus" slides with one end frosted for labeling, gauze, alcohol pads, and adhesive bandages. In

general, we do not use local anesthetic prior to peforming the FNA except for special circumstances such as very painful lesions.

Performing the FNA

After obtaining informed consent, and using universal precautions against blood contact, the aspirator cleans the skin overlying the nodule to be aspirated with an alcohol pad. Using the first finger and thumb or first and second fingers of the free hand, the nodule is held firmly in place while the other hand is used to smoothly and rapidly insert the needle into the nodule. Pressure on either side of the nodule will help to reduce any pain from the needle stick. The aspirator should then apply a vacuum to the syringe, followed by multiple short, quick back and forth movements with the needle for approximately 5–10 seconds without significantly changing the direction of the needle. In our opinion, a "fanning" movement of the needle during the aspiration could result in more tissue trauma and potential for bruising. The aspirated material (except for cystic lesions) should remain within the barrel of the needle. Once aspirated material or blood appears within the needle hub, the vacuum should be released and the needle withdrawn from the site; this prevents unnecessary dilution of the sample with blood. Upon withdrawal of the needle, a gauze pad is placed over the site and moderate pressure is applied to prevent development of a hematoma. A drop of the aspirated material is expressed onto a glass slide, and 2 smears are made – one for alcohol-fixation and one for air-drying. The needle is then rinsed into a tube containing saline (or other physiologic balanced buffered solution). This aspiration technique is generally repeated 3–5 times (in slightly different areas to maximize sampling) until an adequate sample (based upon rapid assessment) is obtained.

When possible, the salivary gland FNA should be performed by, or in collaboration with, a cytopathologist in order that a preliminary interpretation of the sample can be made before the procedure is completed. This allows for assessment of sample adequacy, but it also permits the triage of the sample for ancillary studies. For example, the needle rinsings from lymphoid lesions can be sent for flow cytometric evaluation for potential lymphoproliferative lesions, a cell block can be made for cases where histochemical and immunohistochemical studies will be needed, microbiologic cultures can be sent for lesions that appear to be inflammatory/

infectious, and a sample can be placed into glutaraldehyde fixative for ultrastructural studies on selected challenging cases.

Summary of Salivary Gland FNA Technique

- Obtain informed consent
- Sterilize skin with alcohol swab
- Immobilize nodule with pressure from 2 fingers of left hand
- Smoothly and rapidly insert needle
- Apply vacuum followed by multiple short, rapid needle strokes
- Release vacuum and withdraw needle
- Obtain an adequate sample by performing 3–5 separate needle sticks
- Prepare both air-dried and alcohol-fixed smears

FNA Specimen Processing

As mentioned previously, a combination of both alcohol-fixed and air-dried smears are essential to maximize the diagnostic evaluation of a salivary gland FNA sample. Because many salivary gland tumors contain a variable combination of cells and matrix material, Diff-Quik and Papanicoloau stains are complementary in the evaluation of salivary gland aspirates. Diff-Quik staining highlights the cytologic features and tinctorial properties of any matrix material that may be present. This is key in distinguishing certain common salivary gland tumors such as pleomorphic adenoma and adenoid cystic carcinoma (see Chapter 6), where the appearance of the matrix rather than the cells is the most important diagnostic feature (Fig. 2.4). Diff-Quik stained smears are also more useful than Papanicoloau-stained preparations for the evaluation of cytoplasmic vacuoles as in acinic cell carcinomas (see Chapter 8). In addition, for the rapid assessment of an FNA specimen, air-dried Diff-Quik preparations are much less time-consuming to prepare. Alcohol-fixation and Papanicoloau staining are useful for better visualizing the nuclear features of the cell, including chromatin pattern, nuclear membrane irregulaties, nucleoli, and inclusions. In our opinion, a detailed evaluation of nuclear atypia is best achieved using Papanicoloau staining.

As an adjunct to standard smears, needle rinsings are important since they can be used to produce a thin-layer preparation,

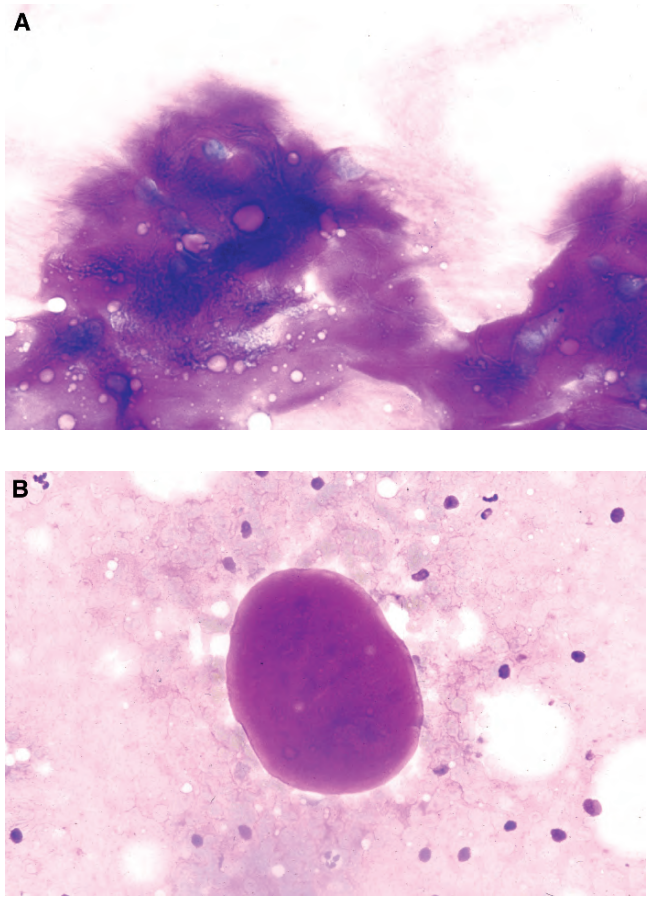


FIG. 2.4. Diff-Quik staining of air-dried smears provides key differential diagnostic information about matrix-containing salivary gland tumors by highlighting characteristic features of the matrix as demonstrated by aspirates of pleomorphic adenoma (A) and adenoid cystic carcinoma (B).

cytospin, and/or cellblock. Thin-layer (TP) or cytospin preparations in our opinion should not be the sole method for evaluating salivary gland lesions, especially those containing matrix material. TPs do have the advantage of concentrating the cells onto a single slide and of removing excess obscuring blood. For cystic lesions where the cellular components are diluted within

a large volume, TP is probably the best preparatory method to use. A cell block can be prepared from the needle rinsings for those cases where histochemical (e.g., mucicarmine, PTAH) or immunohistochemical (e.g., S-100, cytokeratin) stains would be useful in the diagnostic evaluation.

FNA Specimen Processing

- Air-dried, Diff-Quik staining (rapid processing, highlights matrix material and cytoplasmic vacuoles)
- Alcohol-fixed, Papanicoloau staining (highlights nuclear details and atypia)
- Thin-layer preparation (concentration of cells into monolayer, removal of obscuring blood)
- Cell block (histochemical and immunohistochemical stains)
- Needle rinsings (flow cytometric and microbiologic studies)

Suggested Reading

Allen SM, Boon AP, Brownridge DM, Chadwick CH, Buckley JG. Fine needle cytology of palpable head and neck lesions: a comparison of sampling methods with and without suction. Cytopathology 1999;10:97–106.

David O, Blaney S, Hearp M. Parotid gland fine-needle aspiration cytology: an approach to differential diagnosis. Diagn Cytopathol 2007;35:47–56.

Kontis TC, Johns ME. Anatomy and physiology of the salivary glands. In: Bailey BJ (ed): Head and neck surgery – Otolaryngology, 2nd ed. Philadelphia: Lippincott-Raven Publishers, 1998:531–539.

Martinez-Madrigal F, Micheau C. Major salivary glands. In: Sternberg SS (ed), Histology for pathologists. New York: Raven Press, 1992:457–478.

NCCLS. Fine needle aspiration biopsy (FNAB) techniques: approved guideline. NCCLS document GP20-A. NCCLS, 940 West Valley Road, Suite 1400, Wayne, PA, 1986 [www.clsi.org].

Powers CN and Frable WJ. Fine Needle Aspiration of the Head and Neck, Boston: Butterworth-Heinemann, 1996:1–21.

Stewart B, Kruskal B, Kelly D, Sosna J, Kane RA. Utility and safety of ultrasound-guided fine-needle aspiration of salivary gland masses including a cytologist's review. Ultrasound Med 2004;23:777–783.

Wong DS, Li GK. The role of fine-needle aspiration cytology in the management of parotid tumors: a critical clinical appraisal. Head Neck 2000;22:469–473.

3
Algorithmic Approach to Salivary Gland FNA: An Overview

This book uses a simplified and practical algorithmic approach to evaluate and diagnose a wide range of non-neoplastic, benign, and malignant entities encountered in salivary gland cytopathology (Fig. 3.1). After verifying that the FNA sample is indeed satisfactory for evaluation, the cytotechnologist or pathologist focuses upon the general types of cells, matrix, and inflammatory components that are present to fit the aspirate into 1 of 3 major arms of the algorithm based upon the overall cytomorphologic pattern. This will result in a focused differential diagnosis for the lesion. Detailed application of this algorithmic approach and distinction between different entities within the differential diagnoses are discussed and illustrated within specific subsequent chapters of this book. Because some salivary gland tumors can exhibit more than one general cytomorphologic pattern (e.g., mucoepidermoid carcinoma can be cystic, high-grade, or oncocytic), several entities will be presented and discussed in the differential diagnosis of more than one arm of the algorithm.

Assessing Sample Adequacy

The first step in the evaluation of any FNA sample, including aspirates of the salivary gland, is to determine whether the overall specimen is "satisfactory for evaluation." "Unsatisfactory" salivary gland FNA samples are most often caused by acellular or hypocellular specimens, but can also be due to a number of other factors, including

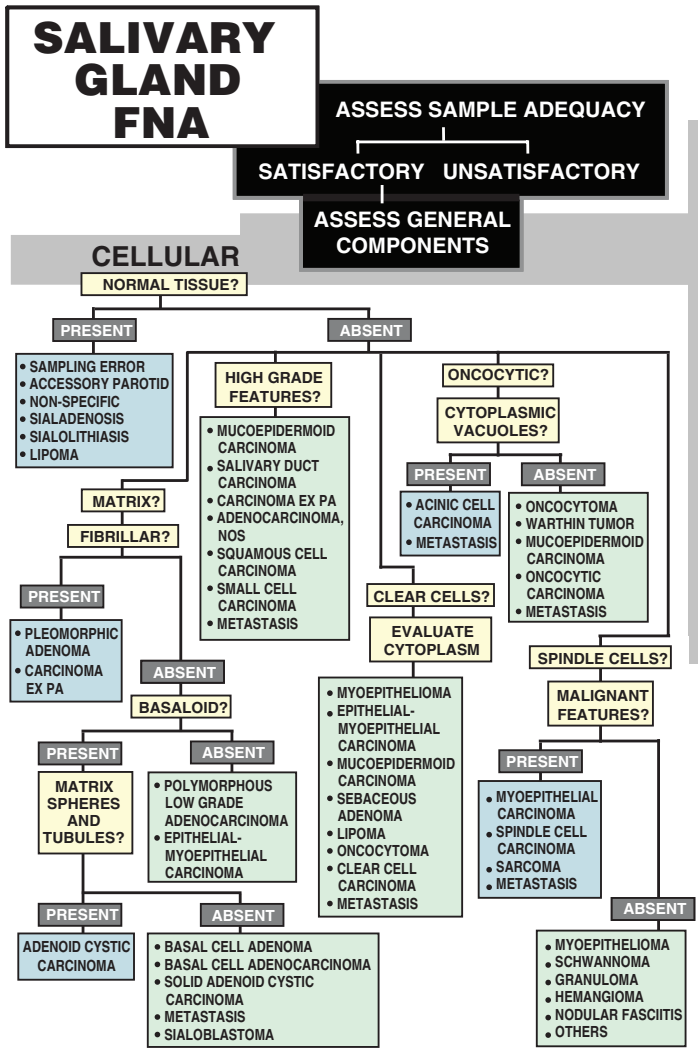
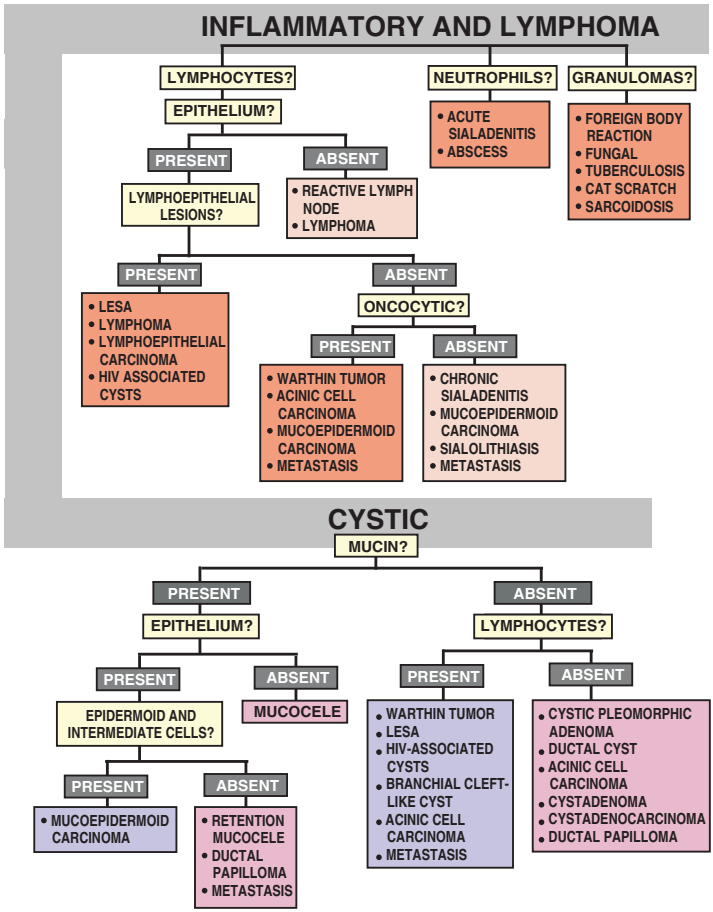


FIG. 3.1. An algorithimic approach to the evaluation of salivary gland FNAs.


INFLAMMATORY AND LYMPHOMA
LYMPHOCYTES?
NEUTROPHILS?
GRANULOMAS?
EPITHELIUM?
• ACUTE SIALADENITIS
• ABSCESS
• FOREIGN BODY REACTION
• FUNGAL
• TUBERCULOSIS
• CAT SCRATCH
• SARCOIDOSIS
PRESENT
ABSENT
LYMPHOEPITHELIAL LESIONS?
• REACTIVE LYMPH NODE
• LYMPHOMA
PRESENT
ABSENT
• LESA
• LYMPHOMA
• LYMPHOEPITHELIAL CARCINOMA
• HIV ASSOCIATED CYSTS
ONCOCYTIC?
PRESENT
ABSENT
• WARTHIN TUMOR
• ACINIC CELL CARCINOMA
• MUCOEPIDERMOID CARCINOMA
• METASTASIS
• CHRONIC SIALADENITIS
• MUCOEPIDERMOID CARCINOMA
• SIALOLITHIASIS
• METASTASIS
CYSTIC
MUCIN?
PRESENT
ABSENT
EPITHELIUM?
LYMPHOCYTES?
PRESENT
ABSENT
PRESENT
ABSENT
EPIDERMOID AND INTERMEDIATE CELLS?
MUCOCELE
• WARTHIN TUMOR
• LESA
• HIV-ASSOCIATED CYSTS
• BRANCHIAL CLEFT-LIKE CYST
• ACINIC CELL CARCINOMA
• METASTASIS
• CYSTIC PLEOMORPHIC ADENOMA
• DUCTAL CYST
• ACINIC CELL CARCINOMA
• CYSTADENOMA
• CYSTADENOCARCINOMA
• DUCTAL PAPILLOMA
PRESENT
ABSENT
• MUCOEPIDERMOID CARCINOMA
• RETENTION MUCOCELE
• DUCTAL PAPILLOMA
• METASTASIS

obscuring blood, inflammation, necrosis, or debris, fixation and staining artifacts, and preparatory artifacts such as severe crushing of cells. The precise number of cells required for a salivary gland FNA to be adequate for evaluation has not been well studied and remains a subjective measure. Diagnosing an aspirate as "satisfactory for evaluation" rather than "unsatisfactory" or "nondiagnostic" when there are inadequate cells or material present to explain a clinical mass should be avoided as this can lead to a false negative diagnosis. Importantly, the presence of atypical cells, even if very few in number, should always be mentioned in the body of the report to prompt further evaluation. In effect, the presence of atypical cells outweighs other aspects of the specimen that might otherwise lead to an "unsatisfactory" designation; this approach is analogous to that taken in the evaluation of cervicovaginal specimens.

Among the more common salivary gland entities resulting in an unsatisfactory FNA sample are cystic lesions, both benign and malignant. In some instances, where only cyst contents are aspirated, some cytologists prefer to use the term "nondiagnostic" rather than "unsatisfactory." This is analogous to diagnosing FNA's of thyroid cysts containing cyst contents only as "nondiagnostic, cyst contents only." Either approach is acceptable provided the clinician understands the implications of the diagnosis. In addition to cystic lesions, inadequate sampling of a lesion, particularly small nodules less than 1.0 cm or deep nodules, also accounts for many "unsatisfactory" samples. For those cases where only normal salivary gland tissue is aspirated (see Chapter 4), careful clinicoradiologic correlation is necessary to determine when such an FNA specimen represents a sampling error and is designated as either "nondiagnostic" or "unsatisfactory," or if it accurately reflects the nature of the apparent lesion and is thus adequate for evaluation (e.g., sialadenosis). In either case, an explanatory note to the clinician is useful for correlation and planning for the clinical follow-up evaluation. In the absence of clinicoradiologic information, a hypocellular specimen is more likely to be interepreted as "unsatisfactory" or "nondiagnostic," since the cytologist is relying heavily upon the cellularity of the sample for evaluation. Hence, encouraging the clinician to provide adequate clinical information is extremely important, and may help in reducing the rate of unsatisfactory samples.

Common Features of Unsatisfactory Salivary Gland FNAs

- Acellular and hypocellular specimens
- Fixation and staining artifacts
- Preparation artifacts (e.g., severe crushing of cells)
- Obscuring blood, inflammation, or debris

Assessment of the FNA Sample Components

Once the specimen is considered adequate for evaluation, the FNA can then be assessed microscopically for general components by viewing the slides at low- to-intermediate magnifications. Using the algorithm for salivary gland FNA presented in this book, each FNA is assigned to 1 of 3 major diagnostic categories: (1) predominantly cellular (epithelial, myoepithelial, or mesenchymal elements), (2) inflammatory and lymphoma, or (3) cystic (Fig. 3.1). The diagnostic criteria described in the subsequent chapters of this book will then be applied to arrive at a specific diagnosis or at a focused differential diagnosis within each of these categories.

Cellular Lesions (Discussed in Chapers 4, 6–8, 10–12)

This major category within the algorithm is by far the largest, encompassing a majority of the salivary gland lesions routinely encountered by FNA. This category is further divided into 7 subcategories that are discussed in separate chapters. Most of the salivary gland FNA specimens within this category will be comprised of either epithelial cells or a combination of epithelial and myoepithelial cells, although rare cellular cases comprised of mesenchymal cells (e.g., nodular fasciitis) are also included. After deciding that a case belongs to the “cellular” arm of the algorithm, the next step is to examine the cells to determine the overall cytomorphologic pattern and thus further subcategorize them. As previously mentioned, some lesions will exhibit cytomorphologic features common to more than one subcategory. Within the algorithm, the subcategories of “cellular” specimens (corresponding to subsequent separate chapters) are: normal tissue, matrix-containing tumors, basaloid

tumors, oncocytic tumors, high-grade tumors, spindle cell lesions, and clear cell tumors. In addition, features of metastatic disease, which is included in the differential diagnosis of many different lesions, will be addressed in Chapter 12.

Inflammatory Patterns and Lymphoma (Discussed in Chapter 5)

This category includes aspirates that are characterized by a prominent population of inflammatory cells or, in the case of lymphoma, neoplastic lymphocytes. Depending upon the type of inflammation and epithelial components present, this category encompasses a range of entities from lymphoepithelial sialadenitis (LESA) to acute sialadenitis to sarcoidosis. A subset of salivary gland epithelial tumors can also present with a predominant lymphoid component. Also, in the case of the parotid gland, intraparotid lymph nodes are sometimes sampled by FNA, and these are subject to the same disease processes affecting lymph nodes in other anatomic sites (e.g., reactive changes, infection, metastasis, and lymphoma). For salivary gland FNAs comprised of lymphocytes, the possibility of lymphoma should be excluded. Immunophenotyping may be accomplished by applying ancillary studies such as immunochemistry or flow cytometry.

Cystic Lesions (Discussed in Chapter 9)

Cystic lesions in the salivary gland include the more common entities that contain mucin and muciphages, such as mucocele and low-grade mucoepidermoid carcinoma, and other less common lesions that do not. The latter consist of a combination of cystic neoplasms such as cystadenoma and non-neoplastic cysts such as ductal cysts.

Reporting of Salivary Gland FNA Diagnoses

As recommended in the Guidelines of the Papanicolaou Society of Cytopathology (1997) and in conjunction with the NCI Consensus Conference on “The Uniform Approach to Breast Fine-Needle

Aspiration Biopsy," a fine-needle aspiration report should include statements regarding sample adequacy and limitations (satisfactory or unsatisfactory), an interpretation category (unsatisfactory, nondiagnostic, negative for malignant cells, atypical, suspicious for malignant cells, or positive for malignant cells), as well as a specific or descriptive diagnosis (Tables 3.1–3.4). An expanatory note or comment may also be added, and is particularly applicable to certain salivary gland FNA cases (e.g., basaloid neoplasms) where a definitive diagnosis is not always possible. We support the use of a uniform reporting system as outlined here for FNA samples, including those from the salivary gland. However, we also recognize that variations in reporting of FNA results occur at different

TABLE 3.1. Sample Salivary Gland FNA Report Template.

Patient Identification:	
	Name, age, gender
Specimen Type:	
	FNA ± radiologically sampled (U/S, CT)
Clinical History:	
	Including size of lesion, duration of symptoms, and other associated clinical findings (e.g., paresthesia)
Location:	
	Right/left/superficial/deep salivary glands: Parotid/submandibular/palate/sublingual/lip/oral/other
Adequacy:	
	Satisfactory for evaluation Satisfactory for evaluation but limited by …. Unsatisfactory for evaluation
General Inerpretation Category:	
	Unsatisfactory Nondiagnostic Negative for malignant cells Atypical Suspicious for malignant cells Positive for malignant cells
Diagnosis:	
	Specific or descriptive diagnosis
Note/Comment:	
	Additional description of the lesion, results of special studies, differential diagnosis and recommendations.

TABLE 3.2. Example of a "Negative" FNA Report.

Patient Identification:	Name, 30-year old female
Specimen Type:	Parotid gland FNA
Clinical History:	One-year history of painless, slowly enlarging, 2.0 cm right parotid mass
Location:	Right superficial parotid gland
Adequacy:	Satisfactory for evaluation
General Inerpretation Category:	Negative for Malignant Cells
Diagnosis:	Pleomorphic Adenoma.

TABLE 3.3. Example of an "Atypical" FNA Report.

Patient Identification:	Name, 45-year old male
Specimen Type:	CT-guided parotid gland FNA
Clinical History:	1.5-year history of slowly enlarging, painless, 1.0 cm left parotid gland mass.
Location:	Left deep parotid gland
Adequacy:	Satisfactory for evaluation
General Inerpretation Category:	Atypical
Diagnosis:	Basaloid neoplasm, favor basal cell adenoma. See note.

Note/Comment:
While the overall cytologic features favor a basal cell adenoma, the differential diagnosis also includes basal cell adenocarcinoma or even a more aggressive basaloid neoplasm such as adenoid cystic carcinoma.

institutions depending upon a wide range of factors. The bottom line is that the clinician and cytopathologist ***must*** communicate with each other so that the clinician fully understands the meaning and implications of the cytopathologists' FNA diagnoses.

TABLE 3.4. Example of a "Positive" FNA Report.

Patient Identification:	
	Name, 70 year-old male
Specimen Type:	
	Submandibular gland FNA
Clinical History:	
	Eight-month history of enlarging, painful, 3.0 cm right submandibular gland lesion
Location:	
	Left submandibular gland
Adequacy:	
	Satisfactory for evaluation
General Inerpretation Category:	
	Positive for malignant cells
Diagnosis:	
	High-grade mucoepidermoid carcinoma. See note.

Note/Comment:
The specimen is cellular, consisting of squamoid cells with marked cytologic atypia, and rare mucin-containing epithelial cells as confirmed using special stains for mucin and keratin in cell block material. Together with ancillary studies, the overall cytologic features are consistent with high-grade mucoepidermoid carcinoma.

Adequacy Statement

The key initial decision in the evaluation of a salivary gland FNA is whether it is adequate for evaluation. Samples are assigned to 1 of 3 adequacy statements: satisfactory for evaluation, evaluation limited by…, and unsatisfactory for evaluation.

General Interpretation Category

Next, based upon the overall features of the specimen, the salivary gland FNA should be assigned to a general interpretation category: Unsatisfactory, nondiagnostic, negative for malignant cells, atypical, suspicious for malignant cells, or positive for malignant cells.

Unsatisfactory

As discussed under the adequacy requirements above, this general category describes a specimen that for any of a variety of reasons does not lend itself to a diagnostic interpretation. Rates of

unsatisfactory salivary gland FNA samples in various series range from 5%–9%. A statement within the report providing the reason for an unsatisfactory sample is suggested for both quality assurance purposes and to provide feedback to the sampling physician for quality improvement. Any FNA sample classified as unsatisfactory should receive additional investigation by the referring clinician since an underlying clinically significant lesion has not been excluded.

Nondiagnostic

This category overlaps with the "unsatisfactory" category, and the decision to use one or the other will vary depending upon individual circumstances and/or preferences. Our approach is to utilize the nondiagnostic category for those FNA specimens that contain cellular or noncellular elements of a lesion but do not contain sufficient material for a specific diagnosis. An example of this would be an FNA consisting of cyst contents only without sufficient epithelial cells to further classify the cyst. Such a case would be classified as "nondiagnostic" and would further be described as "cyst contents only." These patients should receive careful follow-up and possibly reaspiration of any solid or complex areas that might yield better diagnostic material. Ultrasound-guided FNA is often useful for such cases.

No Malignant Cells Identified

This salivary gland FNA category is used for those samples that are satisfactory for evaluation and consist of benign elements, either neoplastic (e.g., pleomorphic adenoma) or non-neoplastic (e.g., lymphoepithelial sialadenitis). In parotid specimens, this is the most common diagnostic category, representing approximately 60%–85% of all FNAs. Within this group, pleomorphic adenoma is the most commonly aspirated lesion. The false negative rate ranges from 4% to 10% depending upon factors such as the quality of the sample and the experience of the cytopathologist. Unless the lesion is non-neoplastic, most benign neoplasms diagnosed by FNA are surgically excised. A diagnosis of "no malignant cells identified" can be followed by either a specific diagnosis (e.g., Warthin tumor), or by a descriptive interpretation (e.g., fibrous tissue, benign ductal elements, and mild chronic inflammation) with a differential diagnosis when appropriate.

Atypical

This indeterminate category is used for those FNA samples containing a subset of cells with atypical features, but where the possibility of malignancy is considered low. Caution is urged in using this category, since there is the possibility to overuse it. In various salivary gland FNA series, the atypical/indeterminate category was used in 1%–9% of cases. Because there is an increased risk of malignancy in this category, the majority of patients will be closely monitored and most will be referred for surgical excision. In some cases, the clinician may decide to perform a repeat FNA (possibly using radiologic guidance) to obtain a more diagnostic sample.

Suspicious for Malignant Cells

Salivary gland FNAs within this category, like FNAs from other anatomic sites, have a high probability of being malignant, but lack sufficient cytologic features to warrant a definitive "positive" diagnosis. The rate of malignancy of lesions diagnosed as "suspicious" tends to be greater than 50%. Close clinical follow-up and/or excision are absolutely indicated in these patients.

Positive for Malignant Cells

Salivary gland FNAs diagnosed as malignant represent approximately 10%–24% of cases. As discussed previously, the rate of malignant salivary gland tumors is lowest in the parotid gland, and greatest in the minor salivary glands. While many FNAs with a positive diagnosis will be signed-out as a definitive type of malignancy, approximately one-third or more of cases will not be specifically subtyped, especially when high-grade cytologic features are present (see Chapter 10). Regardless of specific tumor subtype, for clinical management purposes, distinction between low-grade and high-grade tumors is one of the most important distinctions a cytopathologist can make since treatment options will usually differ significantly.

Diagnostic Category

When possible, a specific diagnosis should be rendered. Fortunately, the most common salivary gland tumors by far are

pleomorphic adenoma and Warthin tumor, where FNA can be definitive. In our experience and that of others, between 60% and 70% of salivary gland FNAs will provide a specific diagnosis. However, keep in mind the converse: over 30% of diagnoses will be descriptive or less than definitive. Because of the challenging features of salivary gland cytopathology, many lesions will not be diagnosed as a specific entity; therefore, it is very important that the cytopathologist's report use clear wording that the clinician will understand. Verbal consultation between pathologist and clinician is also prudent in difficult cases.

Notes and Recommendations

As alluded to above, because of the broad range of salivary gland lesions, many with overlapping features, FNA of salivary gland, perhaps more than any other site, lends itself to the frequent use of an explanatory note or comment. This note can be used to better describe the limitations in diagnosis, the inclusion of other entities in the differential diagnosis, and/or a recommendation about follow-up and further evaluation.

Suggested Reading

Das DK, Petkar MA, Al-Mane NM, Sheikh ZA, Mallik MK, Anim JT. Role of fine needle aspiration cytology in the diagnosis of swellings in the salivary gland regions: A study of 712 cases. Med Princ Pract 2004;13:95–106.

Geisinger KR, Weidner N. Aspiration cytology of salivary glands. Semin Diagn Pathol 1986;3:219–226.

Solomon D, Nayar R. The Bethesda System for Reporting Cervical Cytology: Definitions, Criteria, and Explanatory Notes, 2nd ed. New York: Springer, 2004.

Stewart CJR, MacKenzie K, McGarry GW, Mowat A. Fine-needle aspiration cytology of salivary gland: A review of 341 cases. Diagn Cytopathol 2000;22:139–146.

Suen KC, et al. Guidelines of the Papanicolaou Society of Cytopathology for the Fine-Needle Aspiration Procedure and Reporting. Mod Pathol 1997;10(7):739–747.

The Uniform Approach to Breast Fine-Needle Aspiration Biopsy, September 1996, Bethesda, MD. Recommendations presented in Diagn Cytopathol 1997; 16:295–311.

4
The Normal Salivary Gland Aspirate

General Diagnostic Approach

Using the algorithm (Fig. 4.1), specimens of normal salivary gland tissue and its mimics should meet minimal specimen adequacy requirements and be composed of a predominance of benign acinar cells and occasional other normal salivary gland elements. Depending upon the nature of the underlying lesion, aspirates of normal salivary gland tissue and its mimics are usually mildly to moderately cellular. Features suggestive of a neoplasm, cyst, or inflammatory lesion are absent (Fig. 4.1). Using this approach, both normal salivary glands as well as a variety of pathologic entities are included in the differential diagnosis. **Beware: sampling error is one of the more common causes of an aspirate of normal salivary gland tissue!**

Cytologic Features of Normal Salivary Gland

Aspirates of normal salivary gland tissue are variably cellular, composed of small groups of acinar cells, occasional ductal cells, and admixed fibroadipose tissue (Fig. 4.2). Acinar cells are present in cohesive grapelike clusters with polarization of the constituent cells and with associated inconspicuous tubules and small honeycomb sheets of ductal cells (Fig. 4.3). It may be necessary to search carefully at high magnification to discern the small groups of intercalated ductal cells tightly associated with the acinar cell

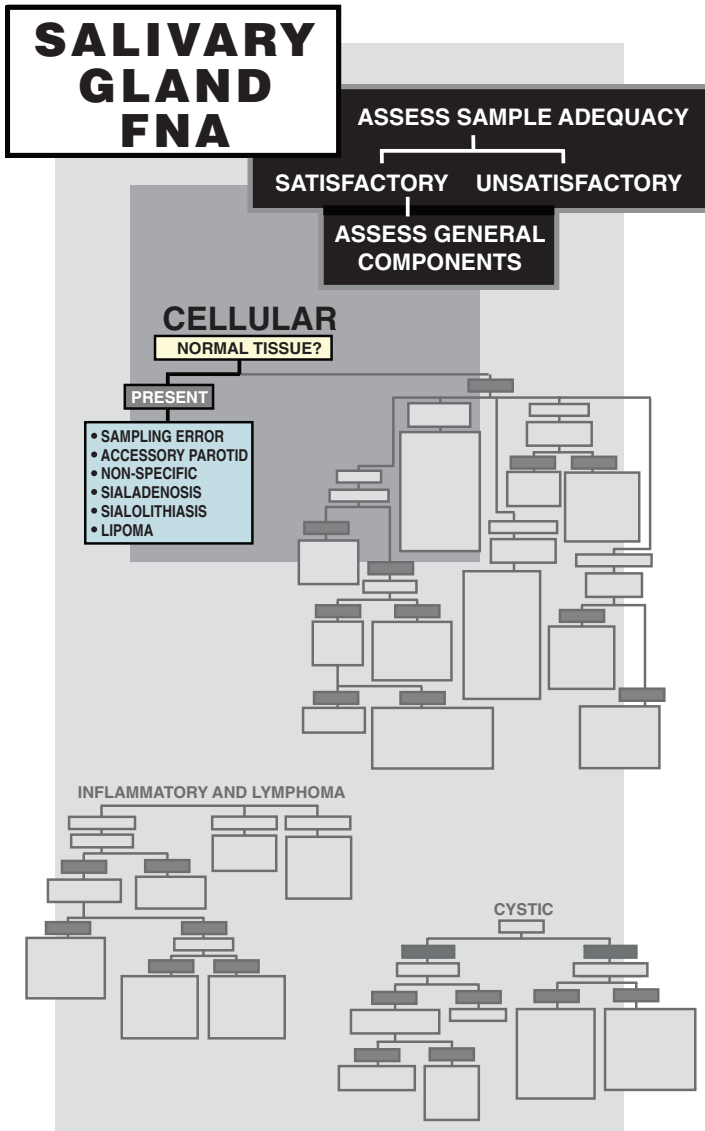


FIG. 4.1. Algorithmic approach to aspirates containing normal salivary gland tissue.

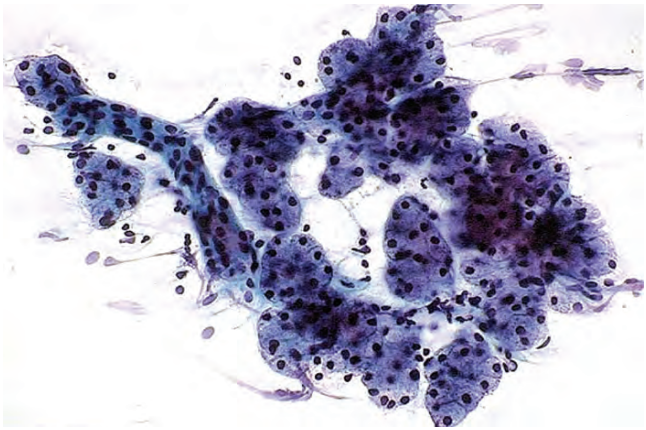


FIG. 4.2. Normal parotid gland. Low-magnification view demonstrating an admixture of acinar cells within lobules, small ducts, and fibroadipose tissue (Smear, Papanicolaou.)

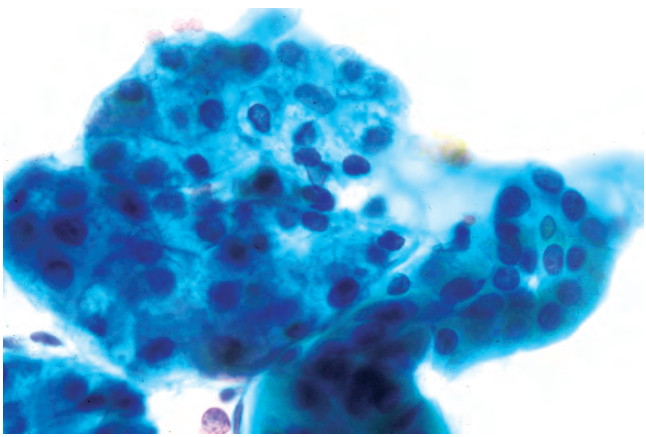


FIG. 4.3. Normal parotid gland. High-magnification view demonstrating polarization of serous-type acinar cells within lobules and associated intercalated duct cells. (Smear, Papanicolaou.)

clusters. The fragility of the acinar cell cytoplasm often results in disruption during sample preparation, resulting in stripped nuclei from these disrupted acinar cells scattered in the background of the

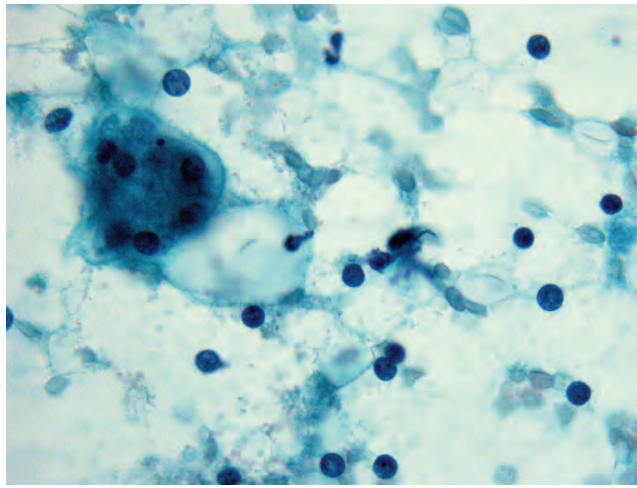


FIG. 4.4. Normal salivary gland. Numerous background stripped nuclei are typically present owing to the fragile nature of the acinar cell, and should be distinguished from lymphocytes. (Smear, Papanicolaou.)

specimen (Fig. 4.4). Myoepithelial cells, while present as a minor component, are generally not a feature readily identified in the normal salivary gland aspirate. Because of the presence of intraparotid and periparotid lymph nodes, lymphocytes can occasionally be seen as a component of a normal parotid gland aspirate, but significant numbers of lymphocytes without features consistent with a lymph node (germinal centers, tingible body macrophages, etc.) are considered abnormal in aspirates.

Cytologic Features of the Normal Salivary Gland Aspirate

- Acinar cells in grapelike clusters
- Small tubules or honeycomb fragments of ductal epithelium
- Firoadipose tissue
- Background stripped acinar cell nuclei

Rarely, individual clusters of sebaceous cells can be seen as a minor component of normal salivary glands, but these cells should be associated with other normal elements and should not be a significant component of the aspirate. The presence of sebaceous cells requires

a differential diagnosis between a rare sebaceous adenoma or sebaceous lymphadenoma of the salivary gland. Particularly in older individuals, oncocytic changes of the ductal epithelial cells can be present. The oncocytic changes are manifested by increased amounts of densely granular cytoplasm, but the normal cytoarchitectural arrangement between ductal cells and acinar clusters is maintained. Recognizing the admixture of acinar cells and oncocytic ductal cells in a "normal" architectural pattern will avoid any suspicion that the changes might represent an oncocytic neoplasm (see Chapter 8).

Uncommon Cytologic Features of the Normal Aspirate

- Lymphocytes from periparotid lymph node
- Rare sebaceous cells
- Oncocytic changes within ductal epithelial cells (especially in older individuals)

Depending upon the site of aspiration (i.e., parotid, submandibular, sublingual, or minor salivary gland), the acinar cells can be of either serous, mucinous, or seromucinous type. Especially in the submandibular and sublingual gland, an admixture of acinar cell types can be seen. Serous acinar cells predominate in the parotid gland and form a significant proportion of submandibular gland acinar cells. Mucinous cells predominate in the sublingual gland and are present in the submandibular gland and in many minor salivary glands. Seromucinous-type acinar cells are seen in the sublingual gland and in many minor salivary glands.

The acinar cell is the principal component of any normal salivary gland aspirate, making up over 90% of the cells present. Serous-type acinar cells, which are by far the most commonly encountered due to their predominance in the parotid gland, are large pyramidal cells with abundant coarse, slightly basophilic cytoplasm containing variable numbers of PAS-positive, diastase-resistant zymogen granules (Fig. 4.5). The latter are approximately the size of lysozomes and appear microscopically as dark purple granules using Papanicolaou and modified H&E stains. The cytoplasm of acinar cells also contains variable numbers of small clear vacuoles that are best appreciated in Diff-Quik preparations (Fig. 4.6). Nuclei are round and basally located with indistinct nucleoli. Mucinous-type acinar cells are larger and more columnar than the serous

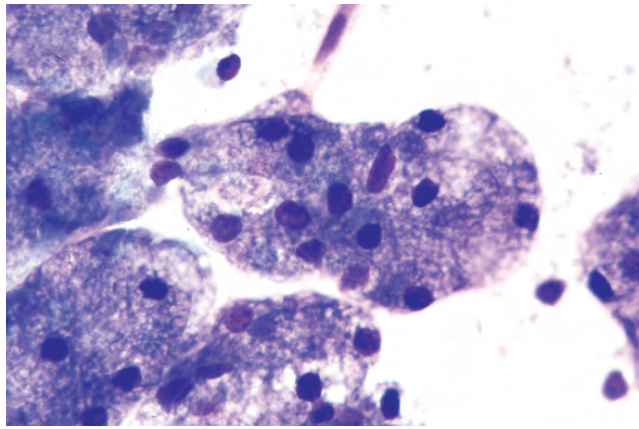


FIG. 4.5. Normal serous-type acinar cells. These cells are characterized by abundant lightly basophilic cytoplasm containing scatterd coarse zymogen granules and vacuoles. (Smear, Diff-Quik.)

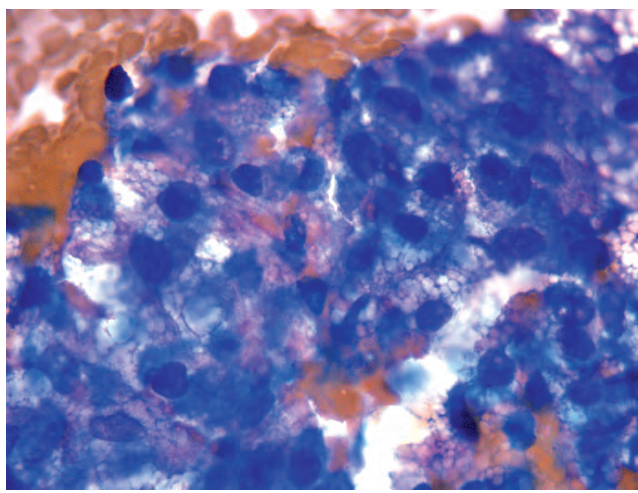


FIG. 4.6. Normal serous-type acinar cells. The cytoplasm typically contains numerous vacuoles that are best appreciated in Diff-Quik-stained preparations. (Smear, Diff-Quik.)

type and are filled with abundant clear vacuolated cytoplasm. They have flattened basal nuclei. Because of the delicate nature of the acinar cell, stripped nuclei are usually present within the background, where they can resemble lymphocytes if not examined carefully. A clue that the structures in question are indeed stripped acinar nuclei and not lymphocytes is the absence of a small rim of delicate basophilic cytoplasm surrounding the nuclear membrane. In addition, lymphoglandular bodies, small background fragments of lymphoid cell cytoplasm, are a feature of aspirates containing abundant lymphocytes. They are not seen in aspirates of normal salivary gland tissue (with the exception of an aspirate of a periparotid or intraparotid lymph node).

Cytologic Features of Normal Serous Acinar Cells

- Pyramidal shape
- Abundant lightly basophilic cytoplasm
- Small cytoplasmic vacuoles (best seen using Diff-Quik)
- PAS-positive, diastase-resistant zymogen granules
- Round eccentric nuclei

The ductal components of the normal salivary gland include intercalated ducts, striated ducts, and excretory ducts. Usually, ductal cells comprise only a small fraction of the identifiable cells within a normal salivary gland aspirate, but careful inspection will always identify at least a few small groups (Fig. 4.7). For diagnostic purposes, it is not important to distinguish between the 3 types of ductal cells present, but their detection is considered a characteristic component of the normal aspirate. Regardless of type, the ductal cells are arranged sometimes in close apposition to the clusters of acinar cells or separately as small honeycomb fragments. Intercalated ductal cells are low cuboidal cells with very scant dense cytoplasm that usually are detected within aspirates as small tubules; they are often directly adjacent to a cluster of acinar cells (Fig. 4.3). Striated duct cells are columnar cells with abundant dense eosinophilic cytoplasm, while excretory duct cells are pseudostratified columnar cells, sometimes with squamoid features. Both the striated duct and excretory duct cells are more commonly found in small sheetlike arrangements within aspirates, and have oval nuclei with a small distinct nucleolus.

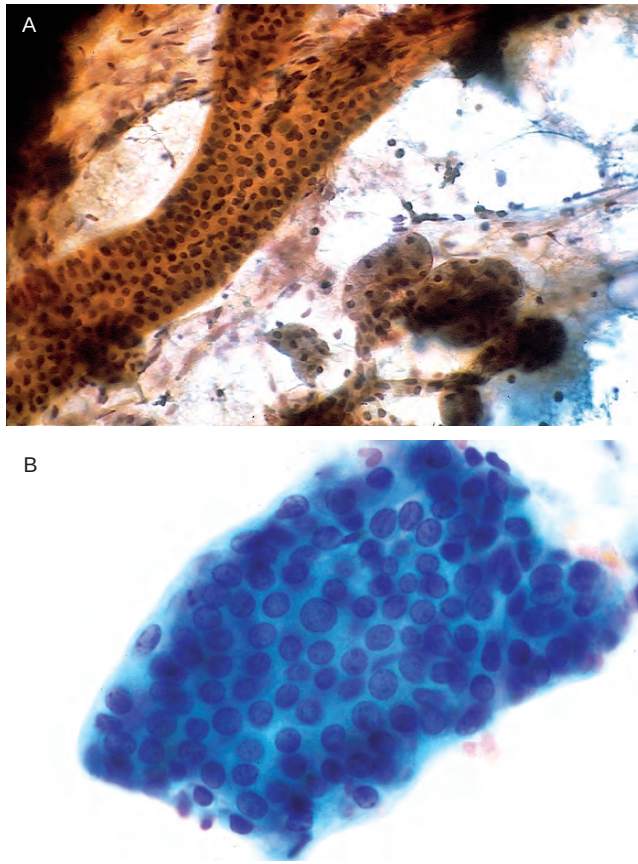


FIG. 4.7. Normal ductal cells. These cells are often detected as small flat sheets of bland epithelial cells in a honeycomb arrangement with oval to round nuclei and dense cytoplasm (A and B). (Smear, Papanicolaou.)

Cytologic Features of Normal Ductal Cells

- Tubules and honeycomb sheets
- Evenly spaced arrangement of cells
- Variable amounts of dense eosinophilic cytoplasm
- May exhibit low-cuboidal, columnar, or squamoid features
- Small oval nuclei
- Small distinct nucleolus

Differential Diagnosis and Pitfalls

While as many as 20% of salivary gland aspirates contain only normal salivary gland elements, the presence of normal salivary gland as the only cytologic finding warrants careful clinicoradiologic correlation to exclude the significant possibility of a sampling error. For aspirates containing only normal-appearing salivary gland, the cytopathologist can either place the diagnosis into the general category of "Negative for malignant cells" or into the category designated "Nondiagnostic;" we prefer the former since some aspirates containing only normal salivary gland tissue are in fact representative of the "lesion" (e.g., accessory parotid gland). In either case, a note or comment explaining that the sample may not be representative of the lesion is strongly recommended. If there is a defined palpable mass present, then repeat FNA may resolve the problem if a sampling error is suspected. In cases where the mass is small or deep and difficult to palpate, then ultrasound-guided FNA may be a useful approach. Other explanations for an aspirate containing only normal-appearing salivary gland elements include a prominent but normal salivary gland, sialadenosis, sialolithiasis, and lipoma.

Differential Diagnosis of a Normal Salivary Gland Aspirate

- Sampling error: missed neoplasm
- Accessory parotid gland tissue
- Non-neoplastic, idiopathically enlarged salivary gland
- Sialolithiasis
- Sialadenosis
- Lipoma
- Hamartoma

A pitfall for the cytologic diagnosis of normal salivary gland tissue is mistaking it for acinic cell carcinoma (and vice versa).

Occasionally, a salivary gland aspirate that yields a predominance of acinar cells with background stripped acinar nuclei can be

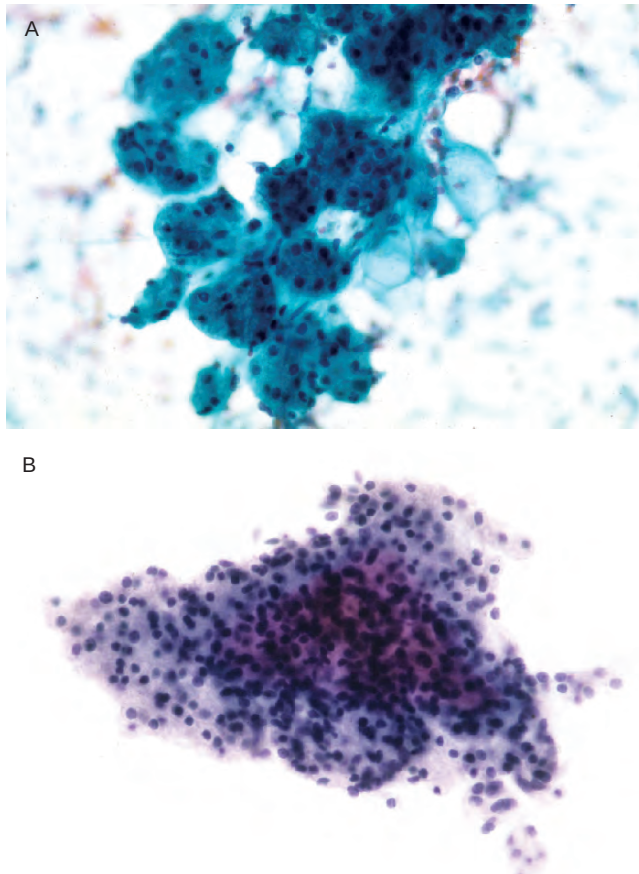


FIG. 4.8. Normal salivary gland acinar cells (A) are organized within lobules, whereas aspirates of acinic cell carcinoma (B) contain acinar cells in crowded 3-dimensional groups. (Smear, Papanicolaou.)

misinterpreted as an acinic cell carcinoma (Fig. 4.8). In our experience, these benign cases tend to consist mostly of single acinar cells and contain few actual groups of cells. The presence of single cells and stripped acinar cell nuclei within the background is seen in aspirates of both normal salivary gland tissue and acinic cell carcinomas. A reliable distinction between normal tissue and acinic cell carcinoma cannot be made on the basis of single cells

alone. One of the most helpful clues favoring normal tissue over acinic cell carcinoma comes through identification and careful examination of any acinar cell groups. Acinic cell carcinomas will invariably contain groups of cells that are crowded together in a haphazard arrangement relative to each other (see Chapter 8). In addition, a subset of acinic cell carcinomas will exhibit background lymphocytes and cell groups with a papillary cytoarchitectural arrangement. In contrast, groups of normal acinar cells lack crowding and exhibit polarization of individual cells relative to each other, the latter resembling a cluster of grapes. Of course, it is possible due to sampling that an aspirate of acinic cell carcinoma will have some normal salivary gland tissue admixed, but this is usually a minor component, and careful evaluation of the overall features of the sample will lead to a correct diagnosis. Other clues that the aspirated material is normal salivary gland rather than neoplastic include the presence of cytologically bland ductal cells, especially when present in a normal arrangement adjacent to acinar cell groups, and the presence of a fibroadipose tissue stroma.

Accessory parotid gland tissue is present in approximately 20% of individuals and presents as a mass which is anterior to and separate from the main parotid gland. Its precise location is variable along the path of Stenson's duct. Accessory parotid gland tissue usually measures less than 1.0 cm, and aspirates yield normal parotid gland elements. Clinically, accessory parotid tissue can sometimes be misinterpreted as a neoplastic condition, but care should also be taken since accessory parotid tissue is subject to development of all of the disorders affecting the parotid gland proper.

Sialoadenosis, or sialosis, is an uncommon, non-neoplastic, noninflammatory enlargement of salivary gland acinar cells. Clinically, it is most often nonpainful and manifests as an overall doughy fullness of the gland, without a well-defined palpable mass. Sialadenosis is more common in the parotid gland, where it is characteristically a bilateral process. It has been associated with a variety of systemic factors, including certain endocrine disorders such as diabetes mellitus, obesity, pregnancy, certain drugs (especially antihypertensives), alcoholism, neurogenic disorders, and cirrhosis. Aspirates of sialadenosis are cellular and contain normal salivary gland elements, with the exception that the acinar cells are hypertrophic. Acinar cell hypertrophy

can be difficult to appreciate by FNA analysis. Instead of the acinar cells being in the normal range of 30–50 µm in diameter, they are enlarged, due to increased amounts of cytoplasm. A mean acinar diameter greater than 62 µm is considered diagnostic of sialadenosis, but the acinar cells in this disorder can be much larger (over 100 µm). In the late stages of sialadenosis, the salivary gland parenchyma can actually become atrophic with diffuse fatty replacement, and aspirates at this stage would consist primarily of fibroadipose tissue. Clinical correlation of the cytologic findings and medical history are needed to arrive at a diagnosis of sialadenosis.

Sialolithiasis involves the formation of ductal calculi, often with concomitant salivary gland enlargement and pain. In chronic cases, the gland is involved by chronic inflammation, squamous metaplasia of ducts, and parenchymal atrophy (see Chapter 5). In the very early stages of the disorder, however, aspirates may yield only normal-appearing salivary gland tissue, sometimes with small calcified fragments of the salivary duct calculus in the background (Fig. 4.9).

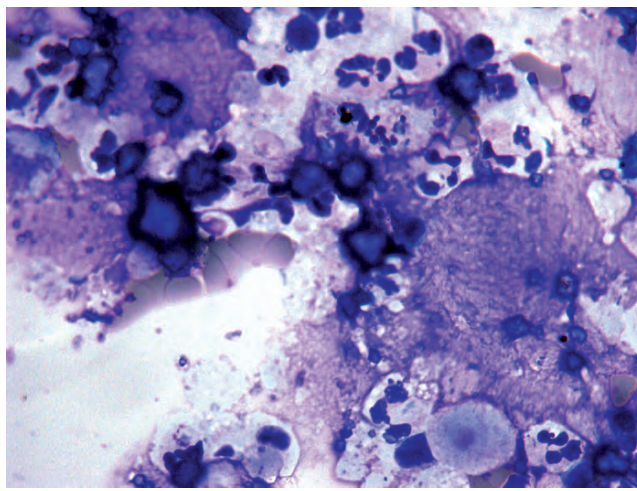


FIG. 4.9. Sialolithiasis. In addition to normal salivary elements, a diagnostic feature is the presence fragments of the salivary duct stone within the background. (Smear, Diff-Quik.)

Salivary gland lipomas have been described that involve the parotid, where they are reported to represent approximately 0.5%–1% of parotid gland neoplasms; they can be either periparotid or intraparotid. Parotid gland lipomas are more common in adult men, and present as soft, slow-growing painless masses. Occasionally, cases of lipomas entrapping normal salivary gland tissue have been termed sialolipomas. Aspirates of parotid lipomas yield a predominance of fibroadipose tissue, occasionally with small amounts of admixed normal parotid gland epithelium within the background (Fig. 4.10). Without careful clinical and radiologic correlation, aspirates of lipoma are often interpreted as "nondiagnostic." The differential diagnosis of a parotid gland lipoma includes fatty replacement of the parotid gland, which is more common in elderly individuals; FNA sampling error of a neoplasm; and as discussed above, chronic sialadenosis. Salivary gland hamartomas are very rare but can present as a discrete salivary gland mass. Histologically, they consist of an abnormal arrangement of normal salivary gland elements including acinar

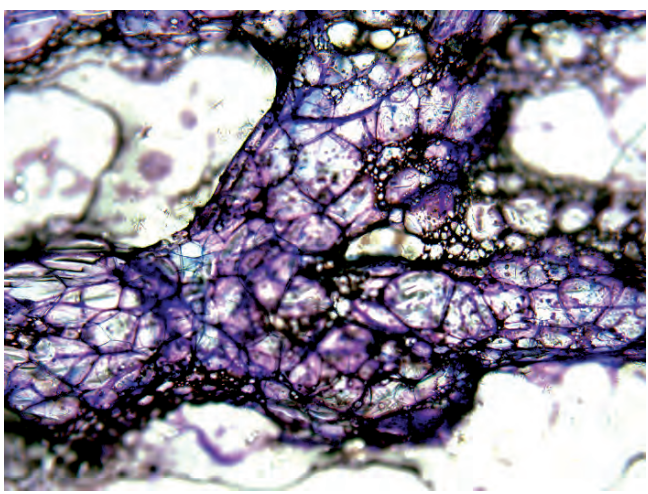


FIG. 4.10. Parotid gland lipoma. Aspirates contain a predominance of fibroadipose tissue, and clinical correlation is needed to distinguish it from other entities including a sampling error. (Smear, Diff-Quik.)

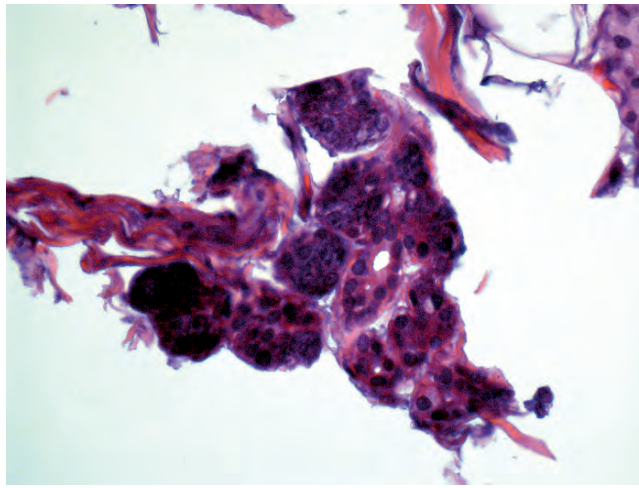


FIG. 4.11. Salivary gland hamartoma. Aspirates consist of an admixture of unremarkable salivary gland elements that are generally indistinguishable from normal salivary gland tissue (Cell block, H&E.)

cells, ductal cells, and adipose tissue (Fig. 4.11). FNA of a salivary gland hamartoma consists of an admixture of unremarkable salivary gland components and in general cannot be readily distinguished from normal tissue.

Clinical Management

A salivary gland FNA of a clinically detected mass showing only normal salivary gland tissue warrants careful clinical follow-up and correlation with clinicoradiologic features. While radiologic imaging does not necessarily distinguish benign from malignant salivary gland disease, it is effective in most cases for documenting the presence of a tumor. Depending upon the suspected nature of the lesion in question, the clinician can choose to use sialography, radionucleotide scanning, ultrasonography, computed tomography (CT) scanning, and magnetic resonance imaging (MRI) (see Chapter 2) as part of the overall workup of a salivary gland

mass. Particularly when an initial FNA does not provide an explanation for a clinically detected mass, CT or MRI is a useful tool to rule out the possibility of a salivary gland neoplasm. Furthermore, to increase the diagnostic yield of the salivary gland FNA by more accurately targeting the apparent mass, the clinician should consider repeat FNA sampling using either ultrasound or CT guidance.

Suggested Reading

Berg HM et al. Correlation of fine needle aspiration biopsy and CT scanning of parotid masses. Laryngoscope 1986;96:1357–1362.

Droese M. Cytological diagnosis of sialadenosis, sialadenitis, and parotid cysts by fine-needle aspiration biopsy. Adv Otorhinolaryngol 1981;26:49.

Ellis GL, Auclair PL. Tumors of the salivary glands. In: Rosai J (ed). Atlas of Tumor Pathology, 3rd series, Fascicle 17. Washington DC: Armed Forces Institute of Pathology, 1996.

Martinez-Madrigal F, Micheau C. Histology of the major salivary glands. Am J Surg Pathol 1989;13:879–899.

Seifert G, Michlke A, Hanubrich J et al. Diseases of the salivary glands: pathology, diagnosis, treatment, facial nerve surgery. Stuttgart: George Thieme, 1986.

Sternberg SS (ed). Histology for Pathologists. New York: Raven Press, 1992.

5
Inflammatory Patterns and Lymphoma

Background

Inflammation of the salivary glands can take several different forms: acute or chronic sialadenitis, lymphoepithelial lesions, or granulomatous disease. Acute sialadenitis is usually a bacterial inflammation of the salivary glands that most commonly affects the parotid gland. The serous nature of the parotid gland saliva, which is less antimicrobial than mucinous saliva, makes it more susceptible to this process. Acute sialadenitis tends to occur more often in individuals who are older and/or medically debilitated, and it is especially common as a postoperative complication. Other causes of acute sialadenitis include stenosis of the salivary duct system due to trauma or sialolithiasis. Importantly, neoplastic conditions can also lead to duct obstruction with accompanying acute and/or chronic sialadenitis. In contrast to older adults, children with acute sialadenitis are affected by the less clinically aggressive nonsuppurative acute parotitis secondary to infection with paramyxovirus (mumps). Most cases of acute sialadenitis in adults result from oral bacterial infection of the salivary ducts. The bacterial causes of acute sialadenitis include *Staphylococcus aureus*, *Streptococcus* spp., and *Haemophilus influenzae*. Patients with acute sialadenitis present with acute onset of pain, swelling, fever, and chills and are managed aggressively, which includes antimicrobial therapy as a cornerstone of the treatment. Untreated suppurative parotitis can be associated with several severe medical complications and even death.

Chronic sialadenitis results in recurrent postprandial swelling and pain of the salivary gland. Anything causing salivary duct obstruction can result in chronic sialadenitis, but the most common etiologic

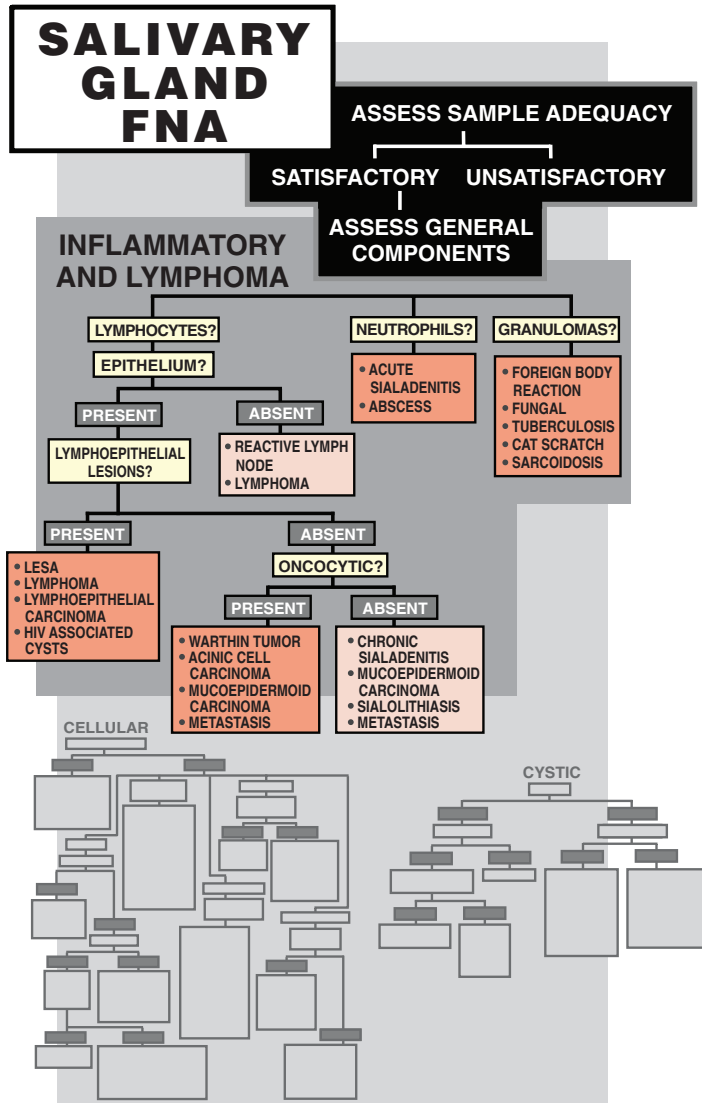


FIG. 5.1. Algorithmic approach to inflammatory conditions and lymphoma.

factor is sialolithiasis. Up to 80% of cases of sialolithiasis affect Wharton's duct of the submandibular gland, followed by approximately 20% of cases involving Stensen's duct, and only 1% in the sublingual duct. Most salivary gland stones are composed of calcium phosphate and calcium carbonate admixed with various glycoproteins, mucopolysaccharides, and salts. The etiology of sialolithiasis is uncertain, but salivary stasis and ductal inflammation are believed to be key factors. Imaging studies such as ultrasound and especially CT are very accurate at detecting salivary gland stones. Repeated episodes of chronic inflammation eventually result in destruction of the normal acinar component of the salivary gland parenchyma, which is replaced by small salivary ducts with metaplastic changes in a background of dense fibrosis. In the submandibular gland, severe chronic sclerosing sialadenitis (Kuttner's tumor) is a unilateral disorder that can mimic a neoplastic condition. Chronic sialadenitis can occur secondary to radiation therapy for head and neck squamous cell carcinoma, in which case FNA can provide a useful means to distinguish a benign reactive condition from metastatic disease.

Granulomatous inflammation of the salivary gland is uncommon, and can be due to a wide range of entities, including mucin extravasation from a mucocele, tuberculous and nontuberculous mycobacterial infection, actinomycosis, cat-scratch disease, toxoplasmosis, tularemia, and systemic diseases such as sarcoidosis. Rarely, neoplasms such as Hodgkin lymphoma, T-cell lymphoma, and certain metastatic carcinomas can have an associated granulomatous inflammation. The clinical presentation of granulomatous sialadenitis is often that of a slow-growing mass that mimics a neoplasm. While tuberculous infection of the salivary gland is more common in adults, children are more often affected by nontuberculous mycobacterial infections typically centered in the cervical lymph nodes.

Causes of Granulomatous Sialadenitis

- Extravasation mucocele
- Mycobacterial infection
- Actinomycosis
- Cat-scratch disease
- Toxoplasmosis
- Tularemia
- Sarcoidosis
- Neoplasia

Lymphoepithelial sialadenitis (LESA) has been known by a variety of names, including Mikulicz's disease, benign lymphoepithelial lesion, and myoepithelial sialadenitis. It can be systemic but primarily affects salivary and lacrimal glands. Due in part to the discovery that the cells comprising the lymphoepithelial islands of this disorder are almost entirely epithelial rather than myoepithelial, the term LESA has emerged as the preferred terminology. The vast majority of patients are women (80%–90%) with a median age in the fifth decade. LESA is believed to be an autoimmune disorder seen in nearly all patients with Sjögren's syndrome; however, approximately 50% of patients with LESA do not have Sjögren's syndrome but rather some other connective tissue disorder (especially rheumatoid arthritis) or no systemic disease whatsoever. Sjögren's syndrome is a clinical triad of keratoconjuncitivits sicca (dry eyes), xerostomia (dry mouth), and rheumatoid arthritis or other connective tissue disease. Biopsy of the labial salivary glands is the usual method for histologically confirming Sjögren's syndrome. LESA can be unilateral or bilateral, cystic or solid. The diagnosis of LESA is associated with an increased risk of over 44-fold for the development of B-cell lymphoma, especially extranodal marginal zone lymphoma of MALT type. FNA is often used to monitor patients with LESA for subsequent development of lymphoma.

Differential Diagnosis of Lymphoid Lesions

- Acute sialadenitis (suppurative and nonsuppurative)
- Chronic sialadenitis
- Granulomatous sialadenitis
- Lymphoepithelial sialadenitis (LESA)
- Non-Hodgkin B-cell lymphoma (MALT, DLBCL)

Primary lymphoma is rare and accounts for 2% of all salivary gland neoplasms and 11% of all primary major salivary gland cancers. Because of the presence of intraparotid lymph nodes, secondary involvement of the parotid gland parenchyma by nodal lymphomas can also occur. Over 75% of primary salivary gland lymphomas involve the parotid gland, and most patients are women presenting in their sixth decade. The disease usually presents as a slowly enlarging, firm, palpable mass without significant pain or tenderness. Approximately 10% of patients have

bilateral or multigland involvement. The majority of primary salivary gland lymphomas are non-Hodgkin B-cell lymphomas, and approximately 85% of these are extranodal marginal zone lymphomas of mucosa-associated lymphoid tissue (MALT) type. The remaining primary lymphomas, approximately 15%, are diffuse large B-cell lymphomas (DLBCL), and some of these represent transformation from an underlying MALT lymphoma. Rare cases of Warthin tumor have been reported with a synchronous primary lymphoma. Follicular lymphomas were previously cited as a major primary salivary gland lymphoma, but most of these probably were nodal lymphomas or MALT lymphomas with prominent follicular colonization. As mentioned above, most MALT lymphomas of the salivary gland arise in a background of LESA, over a period of months to 25 years. The role of FNA in the evaluation of salivary gland lymphoma is to exclude carcinoma and to monitor patients with LESA by obtaining cytologic material for immunophenotypic studies such as flow cytometry.

Clinicopathologic Features of Primary Salivary Gland Lymphoma

- 2% of all salivary gland neoplasms
- Women in sixth decade
- History of LESA
- Firm, diffuse salivary gland enlargement
- May be bilateral
- Two subtypes:
  - MALT lymphoma (85%)
  - Diffuse large B-cell lymphoma (15%)

General Diagnostic Approach

Using the algorithm (Fig. 5.1), salivary gland FNAs containing a predominance of lymphocytes, neutrophils, or granulomatous inflammation are divided into 3 major branches. For lymphocyte-predominant aspirates, a key branch point is the presence or absence of an epithelial component. An epithelial component prompts a differential diagnosis that includes LESA as well as

lymphoma, the latter requiring immunophenotypic analysis. In the absence of lymphoepithelial lesions, chronic sialadenitis is among the more common conditions, but a number of epithelial neoplasms, including Warthin tumor, can also present as lymphoid-rich aspirates. Acute sialadenitis is usually a clinical diagnosis and presents as an aspirate with abundant neutrophils, while granulomatous inflammation raises a broad differential diagnosis that includes both infectious and noninfectious entities. Microbiologic studies and special stains help in the evaluation of the latter.

Diagnostic Criteria

Acute Sialadenitis

Acute sialadenitis is a painful inflammatory process that is usually treated clinically with antibiotics and is rarely aspirated. When an aspirate is performed, it is often to exclude the presence of an underlying neoplastic condition. Bacteria may be seen in smears (especially in Romanowsky stained smears) or by special stains, and sometimes the most clinically useful information will result from culture and sensitivity testing of the aspirated material. Aspirates of acute suppurative sialadenitis consist of abundant neutrophils and histiocytes with necrotic background debris and fibrin (Fig. 5.2). These cytologic findings are nonspecific and generally reflect features of an abscess. Nonsuppurative acute sialadenitis will show fewer inflammatory cells and more admixed parenchymal fragments including ductal cells with reactive changes. It is important that the patient receives close clinical follow-up with repeat FNA sampling should clinicoradiologic findings show the presence of a residual mass after resolution of the acute sialadenitis.

Cytologic Features of Acute Sialadenitis

- Abundant neutrophils
- Histiocytes
- Necrotic debris and fibrin
- Few ductal cells with reactive changes

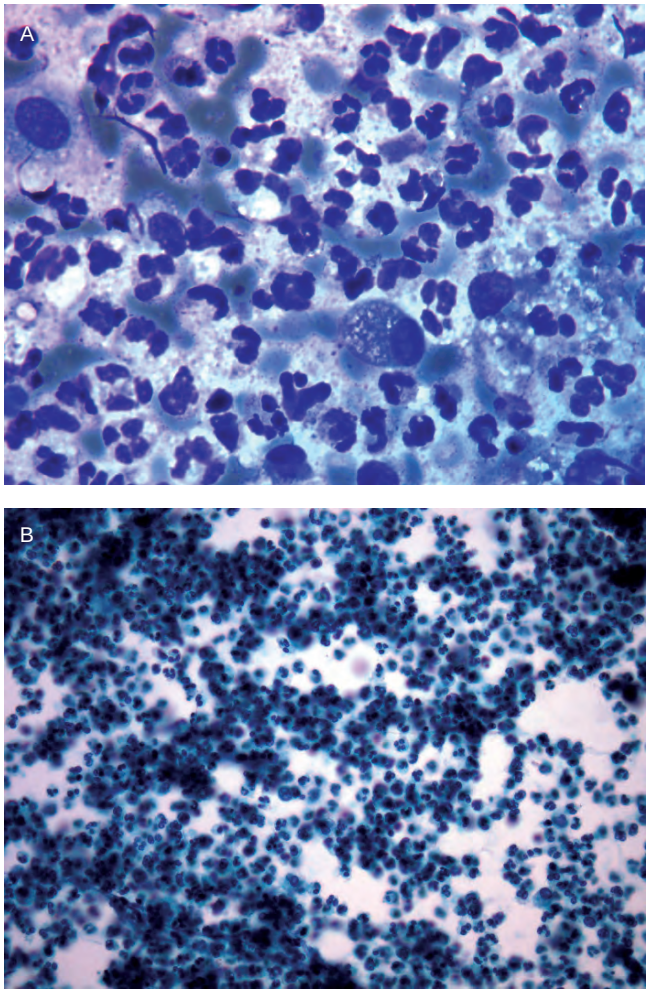


FIG. 5.2. Acute sialadenitis. Aspirates consist of abundant acute inflammation, background debris and histiocytes. Acinar cells are absent. (A, Smear, Diff-Quik; B, Thin-layer preparation, Papanicolaou.)

Chronic Sialadenitis

Aspirates of chronic sialadenitis are characteristically hypocellular specimens that are sometimes of borderline adequacy for evaluation. Depending upon whether the chronic sialadenitis is at an early stage where ductal proliferation occurs or a later fibrotic stage as in Kuttner's tumor (often nondiagnostic due to marked hypocellularity), the aspirate will contain variable amounts of ductal epithelial cells. The ductal epithelium consists of small cuboidal cells with scant dense cytoplasm, small bland round nuclei, and an overall basaloid quality. The ductal cells are in tightly cohesive groups that are usually small to intermediate size with sharp angulated borders (Figs. 5.3–5.4). In some cases, particularly in patients who have been treated with radiation for head and neck cancer, the ductal epithelium can exhibit mild nuclear atypia and/or metaplastic changes. Acinar cells are scant to absent. The background contains variable numbers of mature lymphocytes and occasional plasma cells. In some cases, background mucoid material,

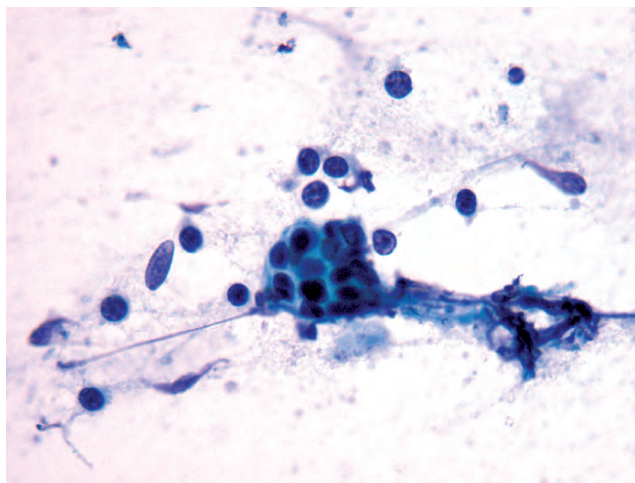


FIG. 5.3. Chronic sialadenitis. Hypocellular specimen with background lymphocytes and small cohesive group of ductal cells. (Smear, Papanicolaou.)

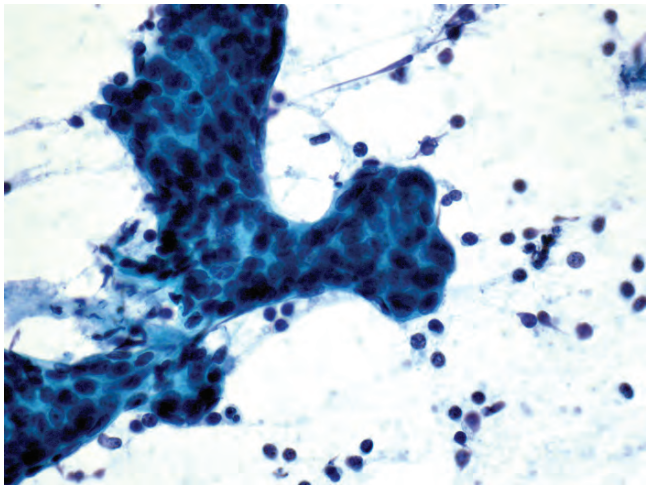


FIG. 5.4. Chronic sialadenitis. Groups of ductal cells are small to intermediate size with angulated borders and basaloid features in a background of lymphocytes. (Smear, Papanicolaou.)

histiocytes, and even psammoma bodies will be present. Small fragments of acellular fibrous tissue are often seen, and in cases of sialolithiasis, a well-sampled lesion may contain fragments of the stone. In some cases of chronic sialadenitis as well as cystic lesions, amylase crystalloids can be seen. Amylase crystalloids are nonbirefringent polygonal, needle- and plate-like structures that stain orange using Papanicoloau stains and deep blue by Diff Quik (Fig. 5.5). They range in size from 5–200 μm

Cytologic Features of Chronic Sialadenitis

- Hypocellular aspirate
- Small, cohesive groups of cuboidal ductal cells
- Scant to absent acinar cells
- Scattered background lymphocytes
- Small fragments of fibrous tissue
- Other findings: crystalloids, mucoid background, stone fragments

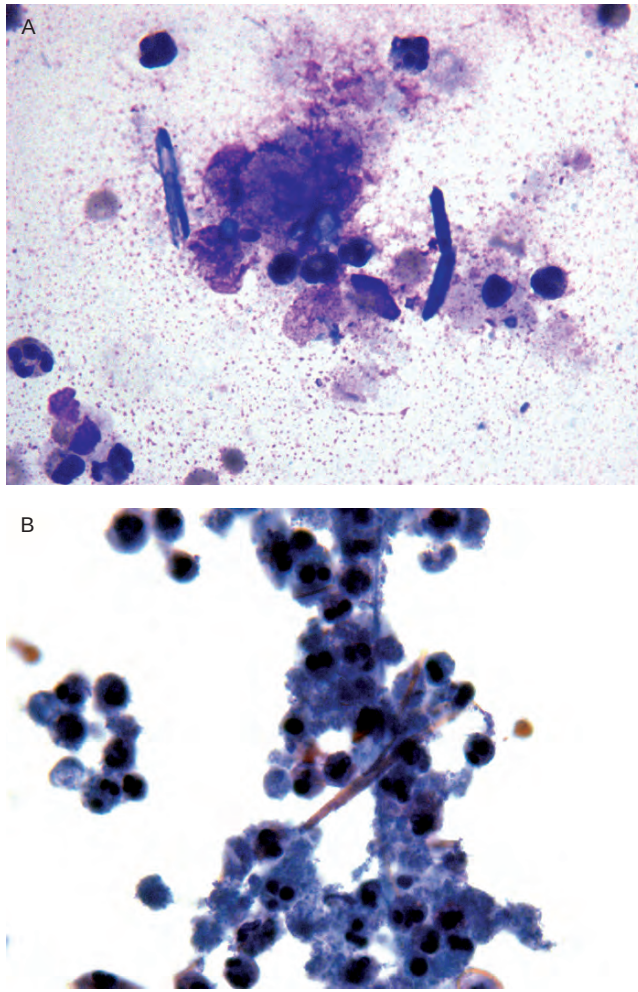


FIG. 5.5. Amylase crystalloids. (A and B) These nonbirefringent crystalloids are most often found in reactive conditions such as chronic sialadenitis and cystic lesions. (A, Smear, Diff-Quik; B, Smear, Papanicolaou.)

Granulomatous Sialadenitis

Aspirates of granulomatous sialadenitis can result from a wide variety of causes that have a similar microscopic appearance. Aspirates are usually hypocellular, and consist of loose collections

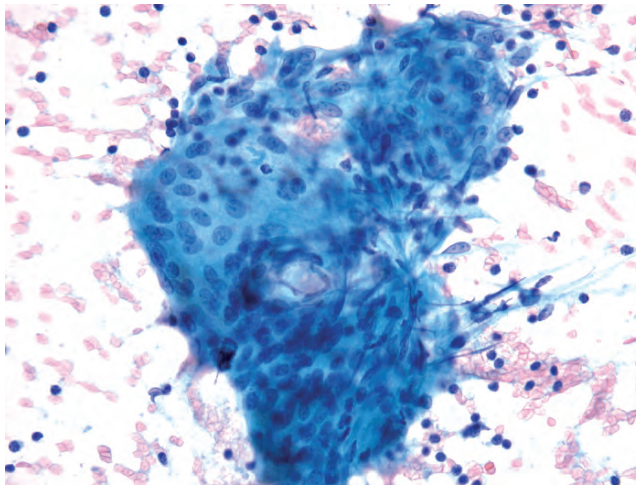


FIG. 5.6. Granulomatous sialadenitis. Aspirates consist of an admixture of epithelioid histiocytes, multinucleated giant cells, and lymphocytes. (Smear, Papanicolaou.)

of epithelioid histiocytes (granulomas), occasional scattered multinucleated giant cells and background lymphocytes (Fig. 5.6). An admixture of other background inflammatory cells, including plasma cells, eosinophils, and neutrophils, can also be seen. Some cases, such as those due to mycobacterial infection, can have background necrotic debris. Care should be taken not to misinterpret the epithelioid histiocytes with their elongate curved nuclei and abundant granular cytoplasm as an epithelial neoplasm. Salivary gland epithelial cells are usually sparse and, when present, consist of small groups of ductal cells similar to those seen in chronic sialadenitis. Special stains and microbiologic cultures should be performed, and, when positive, provide valuable clinical information. Sarcoidosis is a diagnosis of exclusion. Aspirates of sarcoidosis tend to be moderately cellular and comprised of a more uniform population of epithelioid histiocytes than granulomatous lesions due to infectious agents (Fig. 5.7).

Care should be taken not to misinterpret the epithelioid histiocytes as an epithelial neoplasm.

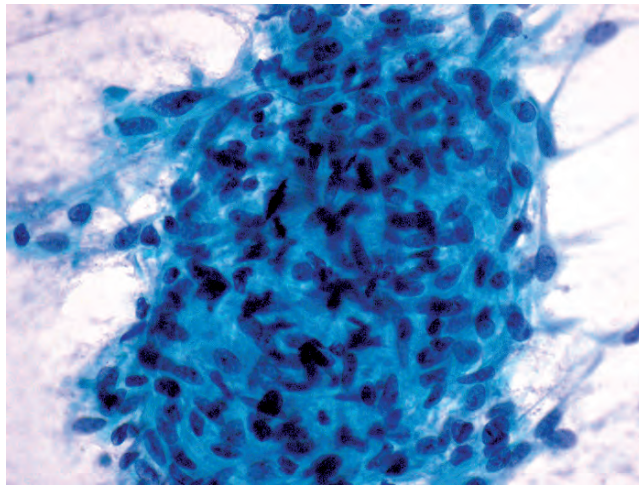


FIG. 5.7. Sarcoidosis. Loose clusters of epithelioid histiocytes (granulomas) are present. (Thin-layer preparation, Papanicolaou.)

Cytologic Features of Granulomatous Sialadenitis

- Hypocellular
- Loose clusters of epithelioid histiocytes
- Multinucleated giant cells
- Mixed chronic inflammation
- Few small groups of ductal cells

Lymphoepithelial Sialadenitis (LESA)

Aspirates of LESA are typically cellular and show an abundance of lymphocytes admixed with plasma cells, lymphohistiocytic aggregates (germinal center fragments), and occasional tingible body macrophages (Fig. 5.8). The lymphocytes exhibit a spectrum of maturation with a predominance of small mature-appearing cells as well as scattered intermediate-sized and larger lymphocytes. The hallmark cytologic feature distinguishing LESA from other salivary gland lesions with lymphocytes is the presence of lymphoepithelial complexes (Fig. 5.9). The latter are tightly cohesive sheets of pale overlapping epithelial cells of ductal origin, often with squamous

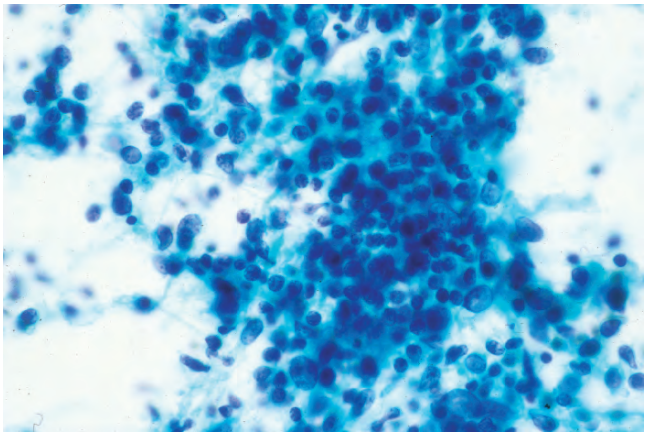


FIG. 5.8. LESA. Aspirates show a mixture of lymphocytes, plasma cells, and lymphohistiocytic aggregates. (Smear, Papanicolaou.)

metaplastic changes, including dense cytoplasm and well-defined cell borders. Although reactive atypia can be seen, the epithelial cells have a uniform monotonous reparative appearance. Individual small mature lymphocytes are present percolating through the sheets of epithelial cells. Acinar cells are generally absent. Some cases of LESA can be cystic, in which case, the aspirate will be hypocellular and more challenging to recognize as LESA. Because LESA is virtually indistinguishable from MALT lymphoma by FNA, immunophenotypic studies such as flow cytometry are essential to exclude a lymphoproliferative disorder.

LESA can be virtually indistinguishable from MALT lymphoma in fine needle aspirates.

Cytologic Features of LESA

- Cellular aspirate
- Mixed population of lymphocytes, plasma cells, tingible body macrophages
- Germinal center fragments
- Lymphoepithelial complex

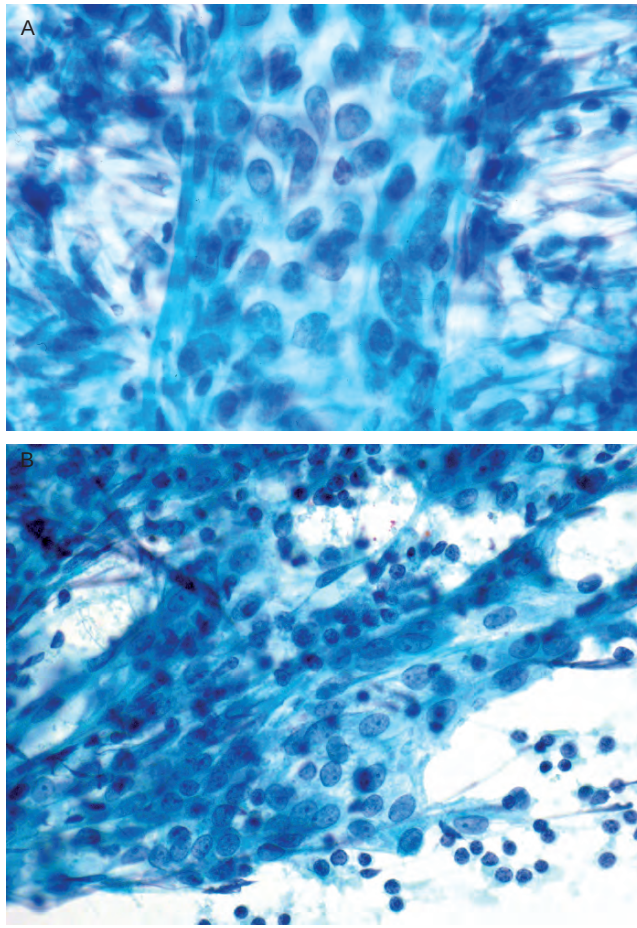


FIG. 5.9. LESA. (A and B) Lymphoepithelial complexes are the hallmark feature of LESA. (Smear, Papanicolaou.)

Primary Salivary Gland Lymphoma

MALT lymphoma is the most common primary lymphoma occurring in the salivary gland, typically arising within a background of LESA. It is a low-grade B-cell lymphoma comprised of small- to intermediate-size lymphocytes. Cytologically, the features of

MALT lymphoma overlap significantly with those of a reactive lymph node and with LESA; therefore, immunophenotypic studies are essential for accurate cytologic diagnosis! Aspirates contain a heterogeneous population of lymphoid cells with an increased number of intermediate-size lymphocytes with abundant pale cytoplasm (monocytoid B-cells) as well as plasmacytoid cells, scattered immunoblasts, and plasma cells (Fig. 5.10–5.11). A subset of cases contains a predominance of plasmacytoid cells. Nuclei of the monocytoid B-cells are slightly irregular with condensed chromatin and indistinct nucleoli. Lymphohistiocytic aggregates representing germinal center fragments are present, but the numbers of tingible body macrophages and large activated follicle center cells are decreased. MALT lymphomas show light chain restriction and exhibit an immunoprofile that is generally CD19+, CD20+ and CD45+, but CD5−, CD10−, and CD23−. MALT lymphomas are negative for bcl-1 (cyclin D1) and bcl-6 but positive for bcl-2.

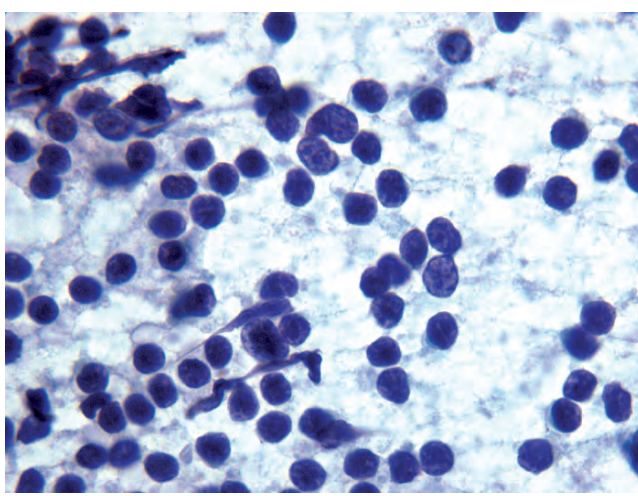


FIG. 5.10. MALT lymphoma. The aspirate contains an increased number of small to intermediate-size lymphocytes resembling a reactive condition. (Smear, Papanicolaou.)

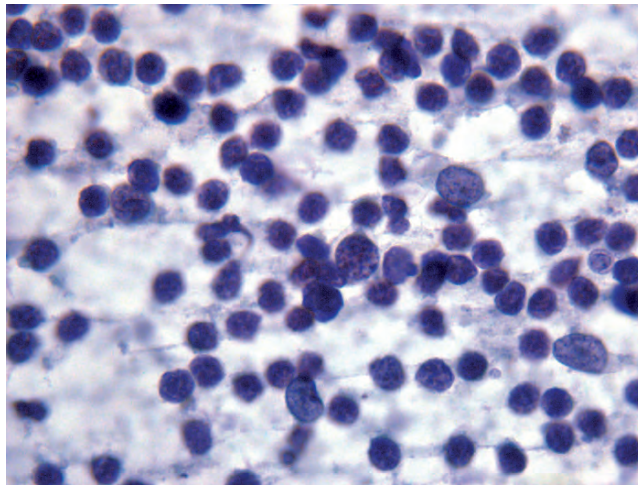


FIG. 5.11. MALT lymphoma. The monocytoid B-cell, with its moderate to abundant delicate pale cytoplasm, is often a prominent component of MALT lymphomas. (Thin-layer preparation, Papanicolaou.)

Cytologic Features of MALT Lymphoma

- Cellular aspirate
- Increased small- to intermediate-size lymphocytes
- Monocytoid B-cells
- Scattered immunoblasts and plasma cells
- Lymphohistiocytic aggregates ± tingible body macrophages
- Monotypic light chain restriction
- CD20+, CD45+, CD5−, CD10−, CD23−
- Bcl-1−, bcl-6−, bcl-2+

In contrast to MALT lymphoma, DLBCL is easily recognized by FNA due to its malignant nuclear features. When diagnostic difficulties arise in the diagnosis of DLBCL, it is usually due to confusion with nonlymphoid malignancies. Aspirates of DLBCL are cellular and comprised of large, atypical immature lymphoid cells that include centroblast-like cells or immunoblasts in a background of lymphoglandular bodies, a cytologic clue that the aspirate is lymphoid (Figs. 5.12–5.13). The malignant lymphoid cells are 2–3 times larger than a small mature lymphocyte, and have irregular nuclei

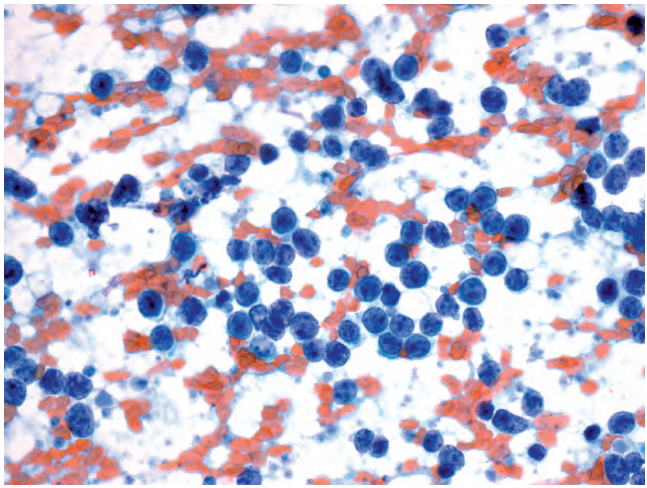


FIG. 5.12. Diffuse large B-cell lymphoma. The aspirate shows a single cell pattern of large atypical lymphoid cells in a background with lymphoglandular bodies. (Smear, Papanicolaou.)

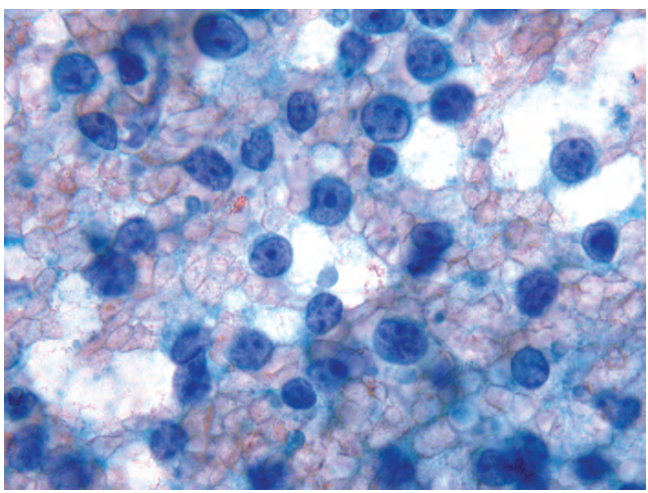


FIG. 5.13. Diffuse large B-cell lymphoma. Immunoblastic forms are 2-3 times the size of mature lymphocytes and such cells contain a prominent central nucleolus. (Smear, Papanicolaou.)

with vesicular chromatin and basophilic cytoplasm. The centroblast-like cells have 1–3 peripheral nucleoli and scant cytoplasm, while the immunoblastic cells have a prominent central nucleolus and moderate to abundant cytoplasm. Using flow cytometry or some other method of immunophenotypic analysis, the typical profile for DLBCL shows light chain restriction and expression of CD45 and pan-B cell markers such as CD20; other markers including CD5, CD10, and CD23 are variable but often negative.

Cytologic Features of Diffuse Large B-Cell Lymphoma

- Cellular aspirate
- Large, atypical immature lymphoid cells
- Background lymphoglandular bodies
- Monotypic light chain restriction
- CD20+, CD45+, CD5−, CD10±

Differential Diagnosis and Pitfalls

The differential diagnosis of chronic sialadenitis includes LESA, Warthin tumor, a basaloid neoplasm, lymphoma, and mucoepidermoid carcinoma. In contrast to the first 4 of these entities, which yield cellular aspirates, FNA of chronic sialadenitis is hypocellular, sometimes even bordering on being nondiagnostic. In addition, chronic sialadenitis lacks both the lymphoepithelial complexes of LESA and the cohesive groups of oncocytes and cystic background that characterize Warthin tumor. The ductal cells in chronic sialadenitis can show metaplastic squamoid changes, raising the possibility of a mucoepidermoid carcinoma; however, the 3 cellular components (mucous cells, epidermoid cells, and intermediate cells) and the prominent cystic mucoid background of mucoepidermoid carcinoma are not present. The most common pitfall in the evaluation of chronic sialadenitis is mistaking it for a basal cell neoplasm since the ductal epithelium of the former can be basaloid. The groups of epithelial cells in chronic sialadenitis are generally sparse as well as small, compact, and angulated, in contrast to the cellular aspirates of basal cell tumors comprised of large and crowded clusters of basaloid epithelial cells. In patients who are postradiation for head and neck squamous cell carcinoma, the ductal cells of chronic sialadenitis can exhibit radiation atypia, but

the marked hyperchromasia and angulated nuclei of squamous cell carcinoma are absent. Keep in mind that chronic sialadenitis is often superimposed as a secondary reaction to an underlying problem, especially sialolithiasis and/or mucocele. Sclerosing sialadentitis (Kuttner's tumor) of the submandibular gland is usually hypocellular by FNA, but aspirates can occasionally contain large numbers of lymphoid cells, raising the possibility of lymphoma. Flow cytometry is useful in these cases to exclude the latter.

Differential Diagnosis of Chronic Sialadenitis

- Basaloid neoplasm
- LESA
- Lymphoma
- Mucoepidermoid carcinoma
- Squamous cell carcinoma
- Warthin tumor

The differential diagnosis of LESA includes a variety of lymphocyte-containing entities - reactive, benign, and malignant. The most important and difficult lesion to exclude from the differential diagnosis is MALT lymphoma. **Therefore, ancillary studies for immunophenotyping, such as flow cytometry or immunohistochemistry on cell block material, should be employed liberally to avoid this pitfall!** Although they can be quite subtle, cytologic features favoring LESA over MALT lymphoma include a normal spectrum of lymphocytes in all stages of maturation rather than the shift towards intermediate-size monocytoid B-cells in MALT. In addition, tingible body macrophages and activated follicle center cells are more commonly seen in LESA than in MALT lymphoma.

Cytologic Features Favoring LESA Over MALT Lymphoma

- Spectrum of lymphocytes in all stages of maturation
- Lymphohistiocytic aggregates with tingible body macrophages
- Activated follicle-center cells
- Polytypic light chain expression

LESA differs from a reactive lymph node by the presence of an epithelial component in the form of a lymphoepithelial complex. Aspirates of LESA raise the differential diagnosis of an epithelial neoplasm with a lymphoid component. It is important to be aware

of the salivary gland tumors that can have background lymphocytes: acinic cell carcinoma, lymphadenoma, lymphoepithelial carcinoma, mucoepidermoid carcinoma, Warthin tumor, and metastatic carcinoma to an intraparotid lymph node. None of these tumors exhibit the features of lymphocytes intimately associated with sheets of epithelial cells that form the lymphoepithelial complex of LESA.

Epithelial Salivary Gland Tumors That Can Have Background Lymphocytes

- Acinic cell carcinoma
- Lymphadenoma
- Lymphoepithelial carcinoma
- Mucoepidermoid carcinoma
- Warthin tumor
- Metastatic carcinoma

In a subset of cases, LESA can be cystic. thus necessitating the differential diagnosis of other cystic lesions of the salivary gland. HIV-associated cystic lymphoid hyperplasia is cytologically identical to cystic LESA; however, this disorder is usually a clinical diagnosis in a patient with HIV infection and multiple bilateral small parotid gland cysts. An FNA of this disorder is not usually performed except for rare cases to exclude other primary cystic salivary gland neoplasms or lymphoma. Other cystic salivary gland lesions with a lymphoid background in the differential diagnosis of LESA include a simple lymphoepithelial cyst. Simple lymphoepithelial cysts (branchial cleftlike cysts) are unrelated to HIV infection or to Sjögren's syndrome. Aspirates contain abundant mature squamous cells and anucleate squames singly and in small clusters in a background of lymphocytes and lymphohistiocytic aggregates. The lymphoepithelial complexes seen in LESA are not present.

Differential Diagnosis of LESA

- Reactive lymph node
- Chronic sialadenitis
- Lymphoma
- HIV-associated cystic lymphoid hyperplasia
- Simple lymphoepithelial cyst
- Epithelial neoplasm with lymphocytes

The differential diagnosis of primary salivary gland lymphoma, particularly MALT lymphoma, includes intrapartotid lymph nodes, LESA, and other reactive lymphoid conditions. The distinguishing hallmark of non-Hodgkin B-cell lymphomas is the demonstration of light chain restriction. Other features favoring MALT lymphoma over a benign lymphoid condition include a more homogeneous population of lymphocytes, cytologic atypia, or the presence of plasma cells with intranuclear inclusions known as Dutcher bodies. Once a diagnosis of lymphoma is made, further immunophenotyping and molecular analysis combined with cytomorphology are used to subtype the lymphoma based upon current WHO criteria. Because MALT lymphoma consists of small- to intermediate-size lymphocytes, it is very important to distinguish it from mantle cell lymphoma, which has a much more aggressive clinical behavior. MALT lymphomas are negative for cyclin D1 (bcl-1) and CD5, while mantle cell lymphomas are positive. Although a subset of follicular lymphomas are CD10 negative, most are positive for both bcl-6 and CD10, while MALT lymphomas are negative for these markers. In the evaluation of MALT lymphomas, it is important to exclude the possibility of a DLBCL component. Therefore, if increased numbers of large atypical centroblastic or immunoblastic cells are present, the possibility of transformation to DLBCL should be considered, and excisional biopsy is warranted for further histologic evaluation.

Differential Diagnosis of MALT Lymphoma

- Reactive lymph node
- LESA
- Chronic sialadenitis
- Other small cell lymphomas (e.g. mantle cell and follicular)
- Transformation to DLBCL

DLBCL is easily distinguished from MALT lymphoma and other small cell lymphomas; however, it can sometimes be difficult to exclude a nonlymphoid malignancy with a single cell pattern such as metastatic malignant melanoma and certain carcinomas (e.g., small cell carcinoma, medullary carcinoma, and lymphoepithelial carcinoma). The presence in the background of small cytoplasmic lymphoid fragments known as lymphoglandular bodies is a characteristic cytologic feature of lymphoid

aspirates, both benign and malignant. But the most definitive evidence that the lesion is a lymphoma is through the application of immunohistochemical studies, which are best performed using cell block material. A panel of antibodies, including cytokeratin, HMB-45, S-100, CD45, and CD20, is appropriate for evaluating aspirates of DLBCL where the differential diagnosis of a nonlymphoid malignancy is considered. DLBCL will be CD45+ and CD20+ as well as showing light chain restriction, and negative for nonlymphoid markers.

It can sometimes be difficult to distinguish DLBCL from a nonlymphoid malignancy with a single cell pattern.

Ancillary Techniques

The most important ancillary study for the evaluation of inflammatory conditions of the salivary gland is light chain restriction analysis to exclude the possibility of lymphoma. In fact, analysis for light chain restriction is very critical for all cases involving LESA, where MALT lymphoma is a key differential diagnostic entity. While immunohistochemical studies for kappa and lambda light chains can be performed by in situ hybridization or by immunohistochemistry using cell block material or cytospins, flow cytometry is probably the most accurate and effective means for obtaining diagnostic information. Material obtained by FNA is also suitable for the application of fluorescent in situ hybridization (FISH) and other molecular and cytogenetic analyses. When combined with cytomorphologic findings, the results of these ancillary studies can be used to accurately subclassify salivary gland lymphomas according to the current WHO system. While no specific oncogene has been shown to be specific for MALT lymphomas, trisomy 3 has been found in up to 60% of cases, and t(11:18) (q21;q21) has been identified in 25% to 50% of cases. For patients with LESA in the setting of Sjögren's syndrome, patients should be evaluated for the presence of an autoimmune process; this can be accomplished by ELISA testing for autoantibodies (ribonuclear proteins Ro and La) and by biopsy of the labial minor salivary glands.

Clinical Management and Prognosis

Acute suppurative sialadenitis is a potentially life-threatening illness secondary to septicemia that is managed in its initial stages by antimicrobial therapy and in some cases by surgical drainage. A major factor in the clinical response of patients with acute suppurative sialadenitis is the advanced age and debilitated condition of many patients. Chronic sialadenitis is an indolent disease that can be refractory to conservative treatment regimens and surgical intervention is sometimes required to remove the involved salivary gland. It is clinically important in all cases of sialadenitis to exclude the possibility of an underlying malignancy.

LESA involving Sjögren's syndrome is characterized by dry eyes and dry mouth, but patients can also experience low-grade fevers, myalgias, dysphagia, renal tubular acidosis, vasculitis, and peripheral neuropathy. Treatment usually consists of directed symptomatic therapies as well as preventive strategies to avoid damage to eyes and teeth. As mentioned previously, an important clinical consequence of LESA is the potential for the development of lymphoma. MALT lymphomas of the salivary gland are usually stage 1E (localized disease) at presentation and are associated with an overall excellent clinical prognosis. A primary mode of treatment for salivary gland MALT lymphomas is surgical excision, but the tumors are also sensitive to radiation therapy. DLBCL is a significantly more aggressive and clinically complex disease than MALT lymphoma, but it is potentially curable using combined modes of therapy including multiagent chemotherapy.

Suggested Reading

Allen EA, Ali AZ, Mathew S. Lymphoid lesions of the parotid. Diagn Cytopathol 1999;21:170–173.

Chan ACL, Chan JKC, Abbondanzo SL. Haematolymphoid tumors. In: Barnes L, Eveson JW, Reichart P, Sidransky D (eds). World Health Organization Classification of Tumours: Head and Neck Tumours.. Lyon: IARC Press, 2005: 277–280.

Chhieng DC, Cangiarella JF, Cohen J-M. Fine-needle aspiration cytology of lymphoproliferative lesions involving the major salivary glands. Am J Clin Pathol 2000;113:563–571.

Droese M. Cytological diagnosis of sialadenosis, sialadenitis, and parotid cysts by fine-needle aspiration biopsy. Adv Otorhinolaryngol 1981;26:49–96.

Jaffe ES, Harris NL, Stein H, Vardiman JW (eds). World Health Organization Classification of Tumours: Tumours of Haematopoietic and Lymphoid Tissues. Lyon: IARC Press, 2001.

Pantanowitz L, Goulart R, Cao JQ. Salivary gland crystalloids. Diagn Cytopathol 2006 ;34 :749–750.

Quintana PG, Kapadia SB, Bahler DW, Johnson JT, Swerdlow SH. Salivary gland lymphoid infiltrates associated with lymphoepithelial lesions: a clinicopathologic, immunophenotypic, and genotypic study. Hum Pathol 1997;21:850–861.

Remstein ED, Dogan A, Einerson RR, Paternoster SF, Fink SR, Law M, Dewald GW, Kurtin PJ. The incidence and anatomic site specificity of chromosomal translocations in primary extranodal marginal zone B-cell lymphoma of mucosa-associated lymphoid tissue (MALT lymphoma) in North America. Am J Surg Pathol 2006;30:1546–1553.

Rice DH. Chronic inflammatory disorders of the salivary glands. Otolaryngol Clin North Am 1999;32:813–834.

Rice DH. Non-neoplastic disorders of the salivary glands. Otolaryngol Clin North Am 1999;32:835–843.

6
Matrix-Containing Tumors: Pleomorphic Adenoma and Adenoid Cystic Carcinoma

Background

There are many tumors of the salivary gland that produce matrix, including pleomorphic adenoma, carcinoma *ex* pleomorphic adenoma, adenoid cystic carcinoma, basal cell adenoma and adenocarcinoma, epithelial-myoepithelial carcinoma, and polymorphous low-grade adenocarcinoma. By far, however, the most commonly encountered among these tumors is pleomorphic adenoma. And one of the more common and clinically important differential diagnoses that the cytopathologist faces is between pleomorphic adenoma and adenoid cystic carcinoma. Because the difference in clinical prognosis and management between these two neoplasms is dramatically different, it is imperative not to confuse these two lesions cytologically. In their most classic and frequently encountered forms, FNA can readily distinguish the two. However, in a subset of cases, both pleomorphic adenoma and adenoid cystic carcinoma can exhibit significant cytomorphologic variation and overlap microscopically. The differential diagnosis for these two lesions can encompass a variety of tumors. In view of this, both lesions will also be discussed for comparative purposes in other chapters, particularly with regard to basaloid tumors (Chapter 7) and spindle cell lesions (Chapter 11).

Pleomorphic adenoma, also known as "benign mixed tumor," is the most common salivary gland tumor in both children and adults. Over 75%–80% of parotid tumors and 60% of all salivary gland tumors are pleomorphic adenoma. It can also occur in nonsalivary gland sites, including skin, soft tissue, and breast.

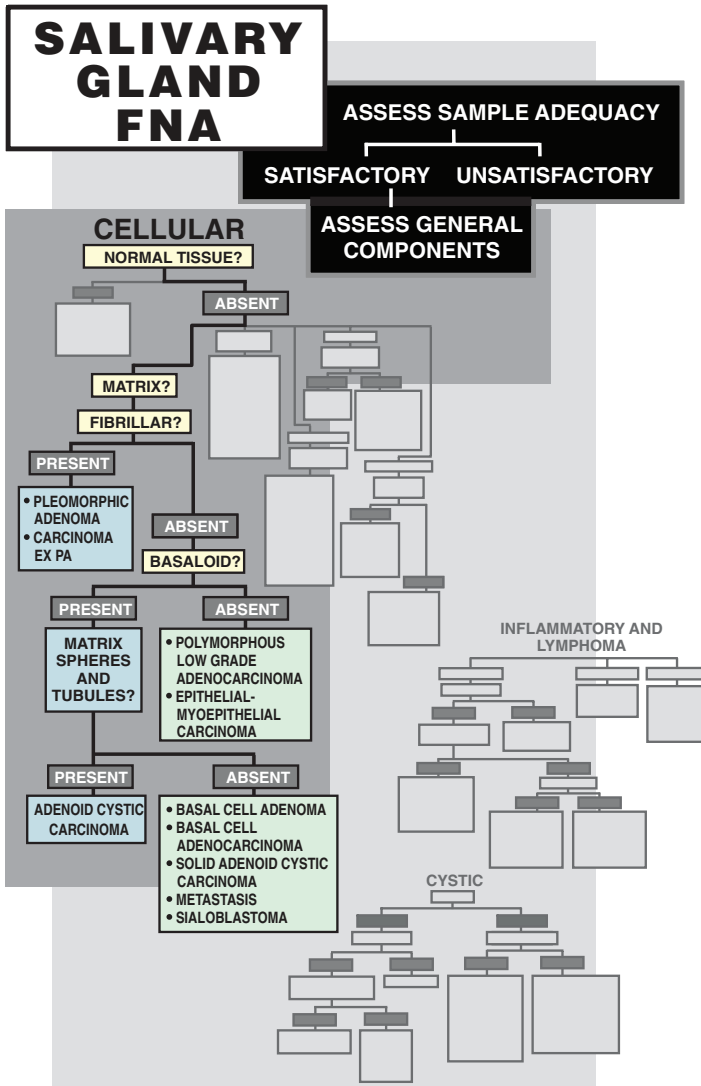


FIG. 6.1. Algorithm showing the differential diagnosis of matrix-containing salivary gland tumors.

The term "pleomorphic" refers to the tumor's admixture of epithelial and mesenchymal elements, and it is believed to be derived from a pluripotent reserve cell of the intercalated duct. A broad age range of individuals is affected – from children to adults. In adults, women are more commonly affected, and the mean age at presentation is approximately 46 years (4th to 6th decade). Patients with pleomorphic adenoma usually present with a painless, slow-growing mass which on examination is solitary, well-defined, firm, and somewhat mobile. In many instances, the tumor has been present for several years and can attain large size, making surgical excision more difficult. Clinical problems in the management of these tumors include recurrent pleomorphic adenoma, malignant transformation into carcinoma *ex* pleomorphic adenoma, and the rare metastasizing pleomorphic adenoma. Even more uncommon is the development of carcinosarcoma *ex* pleomorphic adenoma (aka true malignant mixed tumor).

Within the parotid gland, 90% of pleomorphic adenomas occur lateral to the facial nerve within the superficial lobe, and most of these are in the tail of the parotid gland at the angle of the jaw. It is not uncommon for tumors within the distal parotid tail to be misinterpreted clinically as a lymph node or other nodule within the neck. In such cases, the clinician may be surprised to learn that the suspected lymph node aspiration yielded a pleomorphic adenoma! In approximately 10% of cases, the tumor is present medial to the facial nerve within the deep parotid lobe, and in a minority of cases it extends into the parapharyngeal space. When viewed by MRI, pleomorphic adenomas have a characteristic post-Gadolinium contrast enhancement and high signal intensity on T2-weighted imaging. FNA is an exceptionally accurate tool for the evaluation of pleomorphic adenomas. The majority of cases are easily recognized and distinguished from other salivary gland entities. In our experience, FNA is over 90% accurate in the diagnosis of pleomorphic adenoma. Only a small subset of pleomorphic adenomas with unusual metaplastic or cellular features will pose a diagnostic challenge, as will be discussed (see Diagnostic Criteria).

In contrast to pleomorphic adenoma, adenoid cystic carcinoma is a clinically aggressive tumor representing 4%–10% of all salivary gland neoplasms and 20% of salivary gland malignancies. It is the most common salivary gland malignancy of the submandibular

and minor salivary glands. It occurs most often in the 4th to 6th decade of life, and is slightly more common in women than men. In addition to the parotid gland, submandibular gland, and palate, it also can be found in other sites, such as the nasal cavity and sinuses, tongue and floor of mouth, buccal mucosa, and lip, as well as unusual sites such as skin, breast, lung, prostate, and the lower female genital tract. Patients usually present with a slowly enlarging mass, which, with time, will often become painful and fixed, or result in other symptoms associated with nerve involvement. The tumors typically measure 2–4 cm at presentation; surface ulceration, bony invasion, and marked neurotropism are other clinicopathologic findings.

Although the clinical course of adenoid cystic carcinoma is usually protracted, with good short-term prognosis, the long-term (10–20 year) survival rate is poor (approximately 30%–40% overall). Three histologic subtypes of adenoid cystic carcinoma are recognized: cribriform, tubular, and solid. The most common subtype is the cribriform pattern, which is also the most easily recognized in both cytologic and histologic samples. It is often found in combination with the tubular subtype. In contrast, the solid form is considered by some as a "high-grade" subtype of adenoid cystic carcinoma. It often follows a more clinically aggressive course than the cribriform and tubular subtypes, and it is the most difficult to diagnose in FNA samples because of its lack of matrix material. The strong neurotropism of adenoid cystic carcinoma manifests itself clinically as a painful mass and/or as pain during the FNA procedure. Such symptoms should increase the clinical suspicion of malignancy.

General Diagnostic Approach

Pleomorphic adenoma and adenoid cystic carcinoma are evaluated using the matrix-containing arm of the algorithm (Fig. 6.1). The first step in the evaluation is to recognize the presence within the aspirate of a combination of both cells and matrix material. Next, it is important for salivary gland aspirates in general, but especially for the evaluation of matrix-containing salivary gland tumors, that both air-dried and alcohol-fixed preparations be available. The

matrix material is probably the single most important feature used to distinguish pleomorphic adenoma and adenoid cystic carcinoma from each other as well as from other matrix-containing salivary gland entities. Therefore, following the algorithm, the presence of a predominantly fibrillar matrix material will lead one to the diagnosis of pleomorphic adenoma, while the presence of basaloid cells accompanied by homogenous extracellular matrix spheres and tubules will lead to the diagnosis of adenoid cystic carcinoma. The solid subtype of adenoid cystic carcinoma presents a diagnostic challenge that includes several other basaloid tumors in the differential diagnosis (see Chapter 7)

Diagnostic Criteria

Cytologic Features of Pleomorphic Adenoma

Because pleomorphic adenomas are the predominant tumor occurring in the salivary gland, it is important to be familiar with their classic cytologic appearance as well as with unusual variations that can be seen. As suggested by its name, pleomorphic adenoma exhibits remarkable cytomorphologic diversity. Pleomorphic adenomas are characterized by an admixture of two cell types: myoepithelial cells and epithelial cells along with a unique fibrillar background matrix (Fig. 6.2–6.4). The ratio of matrix to myoepithelial cells and epithelial cells is variable - there is a spectrum of pleomorphic adenomas from matrix-predominant to myoepithelial- or epithelial-predominant (aka cellular pleomorphic adenoma). In our experience, the cells of pleomorphic adenomas are most often myoepithelial cells, but epithelial-predominant lesions do occur and can pose a diagnostic problem.

The myoepithelial cells of pleomorphic adenoma often appear plasmacytoid and are sometimes referred to as hyaline cells due to the presence of eosinophilic cytoplasm (Fig. 6.5). Myoepithelial cells can also have a variety of patterns that include epithelioid, polygonal, stellate, clear, and spindled (Figs. 6.6–6.7). Unlike the epithelial cells, which invariably appear as small cohesive honeycomb groups of uniform cuboidal ductal cells, myoepithelial cells are found individually, embedded within surrounding matrix

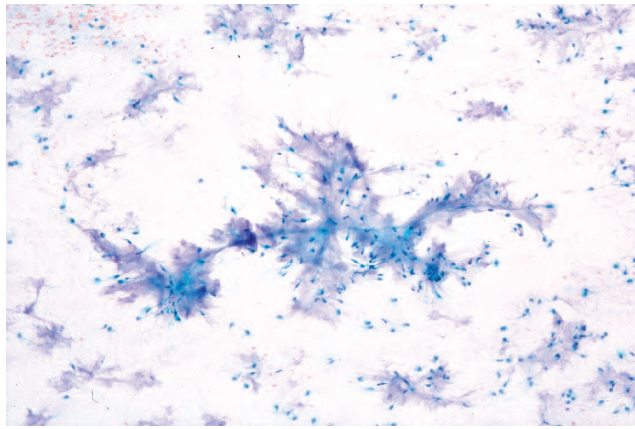


FIG. 6.2. Pleomorphic adenoma. Aspirates are comprised of a combination of myoepithelial cells and myxoid matrix. (Smear; Papanicolaou).

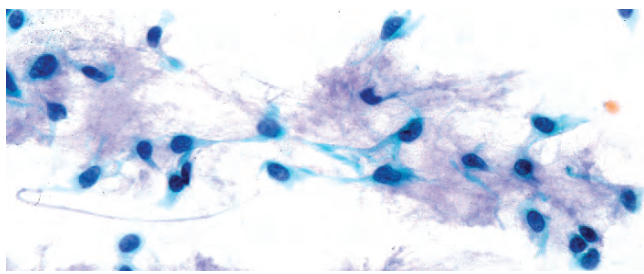


FIG 6.3. Pleomorphic adenoma. Individual stellate and epitheliod myoepithelial cells are embedded within a pale-staining fibrillar matrix. (Smear; Papanicolaou).

material, in small loose clusters, or arranged haphazardly in large groups (Fig 6.8). Myoepithelial-predominant tumors sometimes are classified separately as myoepitheliomas (See Chapters 11 and 12). The latter are rare, and are defined by their histologic absence of a ductal component and absence of chondroid matrix; reliable distinction of myoepithelioma from pleomorphic adenoma requires histologic rather than cytologic evaluation, but this distinction is not clinically significant.

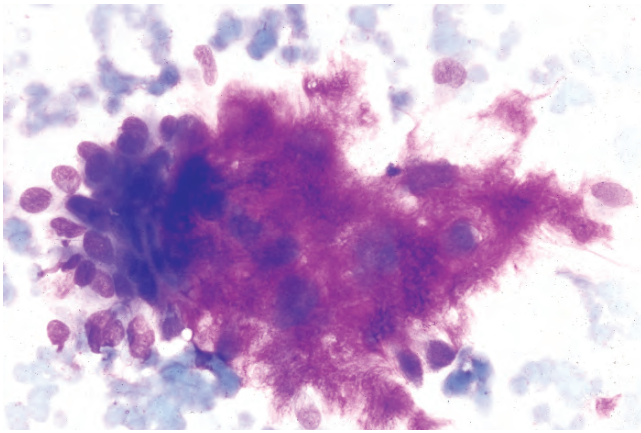


FIG. 6.4. Pleomorphic adenoma. In Romanowsky-stained preparations, the matrix is magenta with myoepithelial cells present individually and in clusters. (Smear; Diff-Quik).

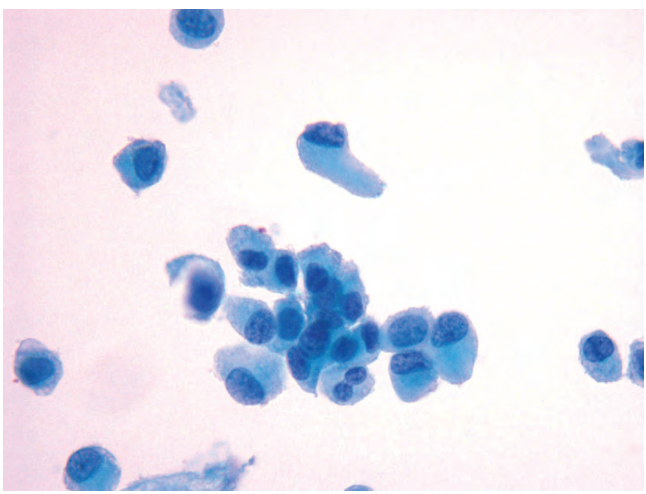


FIG. 6.5. Hyaline cells. The myoepithelial cells in pleomorphic adenomas are often plasmacytoid with dense cytoplasm; some cells are multinucleated. (Thin-Prep; Papanicolaou).

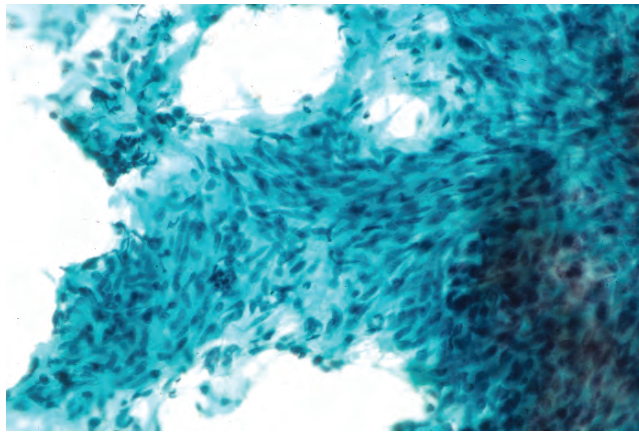


FIG. 6.6. Myoepithelial cells. In some pleomorphic adenomas, the myoepithelial cells are predominantly spindled. (Smear; Papanicolaou).

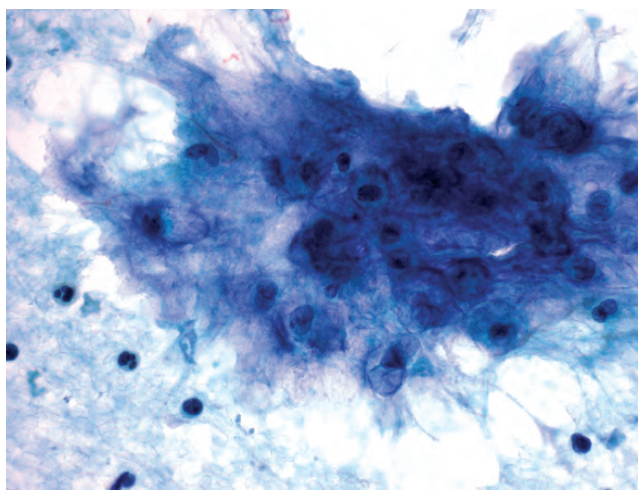


FIG. 6.7. Pleomorphic adenoma. In this example, the myoepithelial cells have abundant clear cytoplasm. (Smear; Papanicolaou).

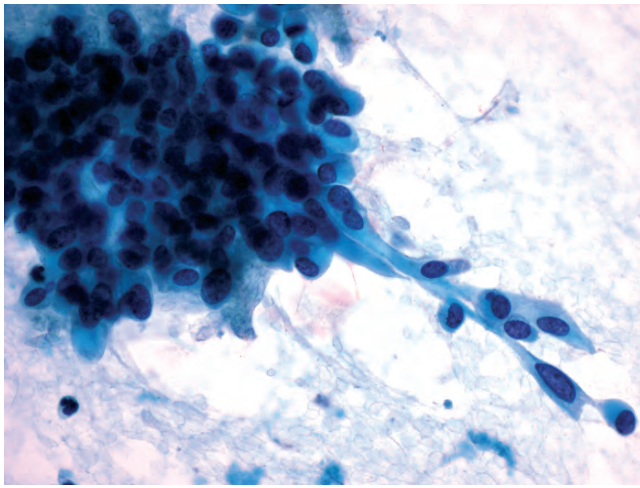


FIG. 6.8. Cellular pleomorphic adenoma. The myoepithelial cells in this cellular pleomorphic adenoma are haphazardly arranged in groups. (Smear; Papanicolaou).

Major Cytologic Features of Pleomorphic Adenoma

- Chondromyxoid matrix material
- Myoepithelial cells
- Cohesive groups of epithelial cells

Cytologic Features of Myoepithelial Cells

- Uniform, bland oval nuclei with indistinct nucleolus
- Dense cytoplasm
- Present singly and in loosely cohesive haphazardly-arranged clusters
- Multiple patterns:
  - Plasmacytoid
  - Epithelioid
  - Polygonal
  - Spindled
  - Clear
  - Stellate

Cytologic Features of the Epithelial Component

- Cuboidal ductal cells
- Bland oval nucleus with small nucleolus
- Moderate amount of dense cytoplasm
- Small honeycomb sheets with distinct cell borders

In addition to the myoepithelial and epithelial components of pleomorphic adenoma, there is a characteristic matrix material which is best appreciated in air-dried Romanowsky-stained preparations, where it is bright magenta (metachromatic) and has a fibrillary pattern; it is pale blue-green, or even colorless in Papanicolaou stained preparations (Figs. 6.9–6.10). The distinctive fibrillary nature of the matrix material with its frayed, indistinct margins and embedded myoepithelial cells is the most important cytologic feature in the cytologic evaluation of pleomorphic adenomas. It is characteristic enough to distinguish a pleomorphic adenoma from other lesions that may mimic it, especially adenoid cystic carcinoma. The matrix can vary in gross and microscopic appearance from mucoid to myxoid to myxochondroid, but it is uncommon

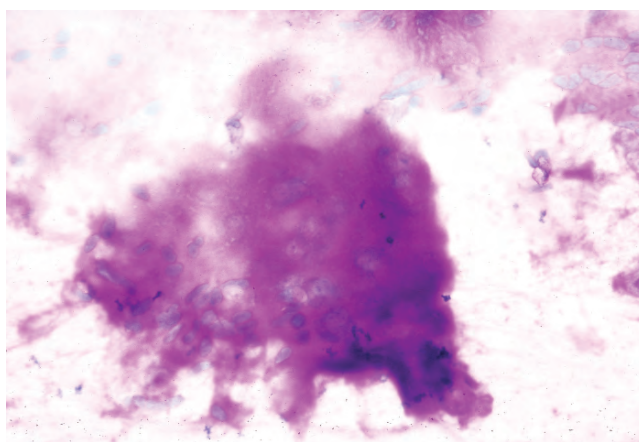


FIG. 6.9. Pleomorphic adenoma. The fibrillar metachromatic matrix is the most characteristic feature of pleomorphic adenomas. (Smear; Diff-Quik).

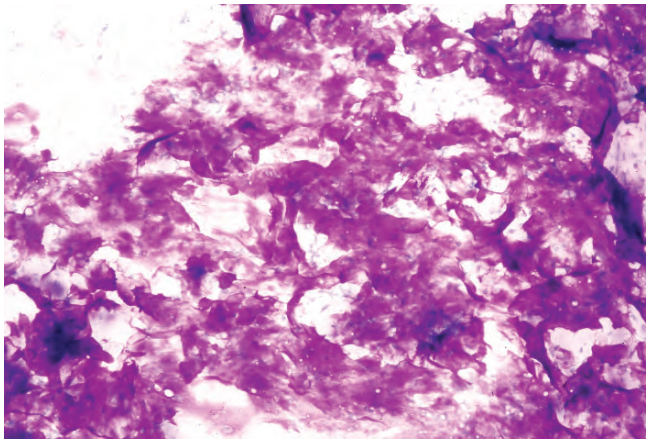


FIG. 6.10. Matrix-predominant pleomorphic adenoma. (Smear; Diff-Quik).

in cytologic preparations to see the chondroid component. Occasionally the chondroid matrix can ossify, forming frank bone, or in other cases, there can be associated adipose tissue within the tumor matrix (pleomorphic adenoma with lipomatous change). Some pleomorphic adenomas consist predominantly of matrix material with very few myoepithelial or epithelial cells, but these are easily recognized and do not cause any diagnostic problems.

The matrix of pleomorphic adenomas is the most important cytologic feature distinguishing it from other salivary gland tumors.

Cytologic Features of the Matrix Component

- Mucoid, myxoid, or myxochondroid
- Fibrillar with frayed indistinct edges
- Embedded single myoepithelial cells
- Magenta (metachromatic) in Romanowsky preparations
- Pale-staining or colorless in Papanicolaou preparations

Pleomorphic Adenomas with Metaplastic and Other Changes

While the majority of pleomorphic adenomas exhibit classic diagnostic features in fine needle aspirates, a very wide range of cytologic variation can be seen among a subset of these tumors. As alluded to previously, some cases of pleomorphic adenoma are cellular, due to a predominance of either myoepithelial or epithelial cells. Such cases have a concurrent decrease in matrix material (Fig 6.11). When myoepithelial cells predominate, they can impart a clear cell appearance or sometimes a spindled appearance to the tumor; when epithelial cells predominate, the cellular pleomorphic adenoma will often appear basaloid. Because of their lack of matrix material, cellular pleomorphic adenomas can pose a diagnostic challenge.

Cytologic atypia in the form of occasional myoepithelial cells with enlarged nuclei, nuclear grooves, inclusions, or multinucleation is acceptable (Fig. 6.12), and does not warrant concern that the tumor is a carcinoma *ex* pleomorphic adenoma. However, atypia which is diffuse, including marked nuclear pleomorphism,

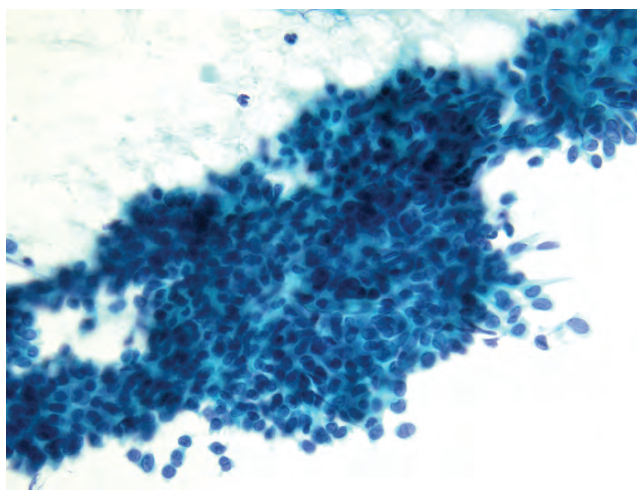


FIG. 6.11. Cellular pleomorphic adenoma. Some pleomorphic adenomas are cellular and contain only scant matrix. (Smear; Papanicolaou).

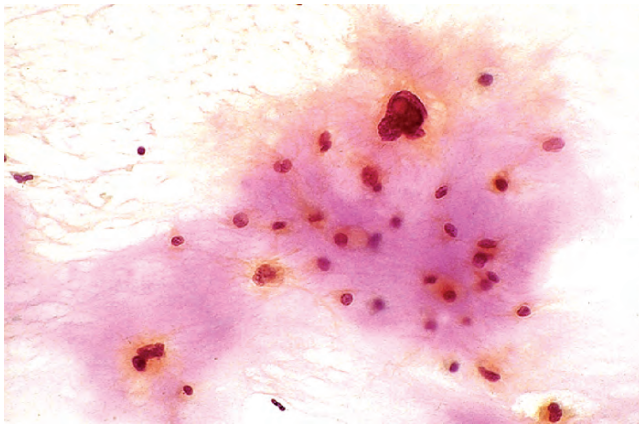


FIG. 6.12. Pleomorphic adenoma. Occasional enlarged "atypical" or multinucleate cells are acceptable in pleomorphic adenomas. (Smear; Papanicolaou).

hyperchromasia, and prominent nucleoli, is not acceptable and warrants caution when interpreting the aspirate. For cases that are atypical but not overtly malignant, it is appropriate to make a diagnosis of "pleomorphic adenoma with atypical features." The malignant component of most carcinoma *ex* pleomorphic adenomas is a high-grade malignancy (usually salivary duct carcinoma) which is easily recognized in cytologic preparations (see Chapter 10).

Metaplastic changes in pleomorphic adenomas are most often squamous, but can also include mucinous, sebaceous, oncocytic, and clear cell features (Fig. 6.13). All can create diagnostic difficulties, but the squamous and mucinous metaplasias are the most problematic. Up to 25% of pleomorphic adenomas have been reported to exhibit squamous metaplasia. This feature encompasses a range of cytologic findings that most often are evidenced by the presence of cytologically bland squamoid cells in cohesive sheets with well-defined cell borders and dense waxy cytoplasm (Fig 6.14). Just as the ductal epithelial cells of the normal salivary gland can undergo squamous metaplastic changes in the setting of chronic inflammation, so the epithelial component of pleomorphic adenoma can undergo the same transformation. In some cases, there is marked

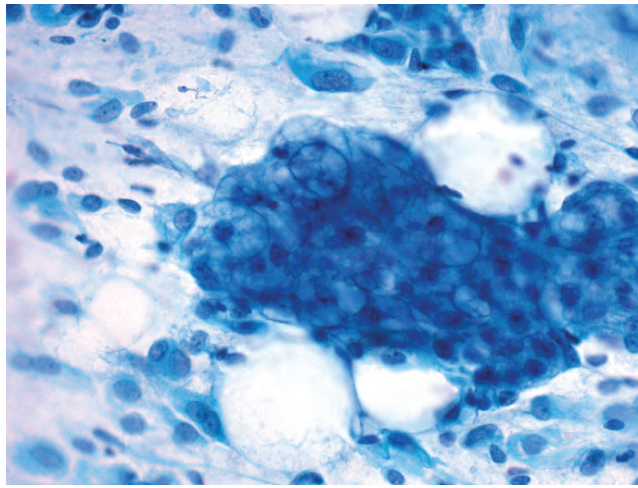


FIG. 6.13. Pleomorphic adenoma with sebaceous metaplasia. (Smear; Papanicolaou).

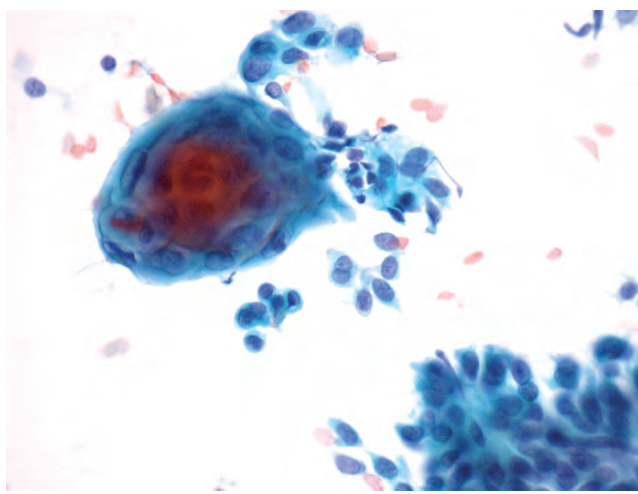


FIG. 6.14. Pleomorphic adenoma with squamous metaplasia. The epithelial cells have dense squamoid cytoplasm and well defined cell borders. (Smear; Papanicolaou).

keratin production and even keratin pearl formation with cystic changes that in effect represent skin adnexal differentiation within the pleomorphic adenoma (Fig. 6.15). When metaplastic changes are extensive, it can be difficult to reliably recognize the lesion as a pleomorphic adenoma unless, through extensive sampling, there is sufficient matrix material present to be diagnostic.

Cytologic Variations in Pleomorphic Adenoma

- Cellular (matrix poor)
- Matrix-predominant
- Pleomorphic adenoma with atypia
- Squamous metaplasia ± keratin pearls
- Mucinous metaplasia
- Sebaceous metaplasia
- Spindled with palisading
- Clear cell changes
- Oncocytic changes
- Cystic degeneration
- Tyrosine-rich crystalloids
- Ossification
- Lipomatous changes

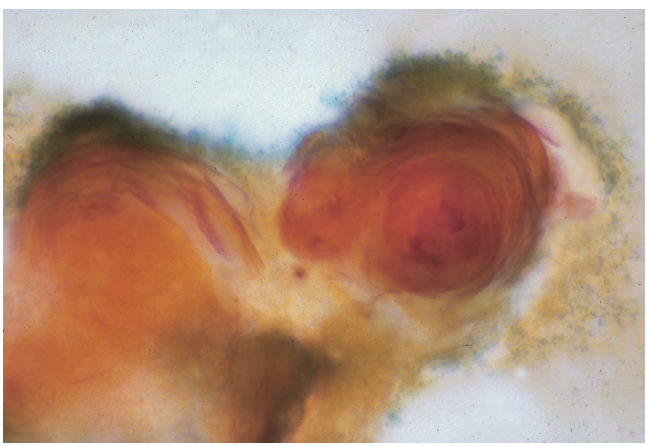


FIG. 6.15. Pleomorphic adenoma with keratin pearls. In some cases with squamous metaplasia, keratin pearls are present. (Smear; Papanicolaou).

Some pleomorphic adenomas with extensive squamous metaplastic changes will exhibit cystic features that include abundant histiocytes and debris, and some are cystic due to degenerative changes. In such instances, it is important to avoid sampling only the cyst contents which would result in a nondiagnostic or misleading sample. Tyrosine-rich and collagenous crystalloids are also occasionally seen in pleomorphic adenomas, but usually only the former are encountered in aspirates. The tyrosine-rich crystalloids are nonbirefringent with a floret-shaped structure and, are most commonly associated with pleomorphic adenomas (Fig 6.16). They are not, however, entirely specific for pleomorphic adenoma, having also been reported in other tumors such as adenoid cystic carcinomas and polymorphous low-grade adenocarcinomas.

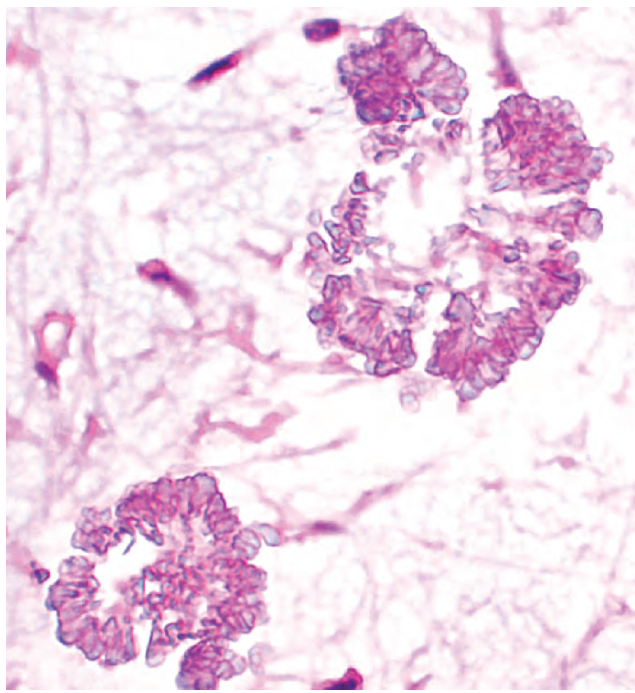


FIG. 6.16. Tyrosine-rich crystalloids. Floret-shaped non-birefringent crystalloids are occasionally found in aspirates of pleomorphic adenoma (Smear; Papanicolaou).

Cytologic Features of Adenoid Cystic Carcinoma

Three forms of adenoid cystic carcinoma can be encountered in aspirates. The cribriform subtype, the most frequent form, with its classic histologic "Swiss-cheese" appearance, and the tubular subtype with its small tubules of basaloid cells are easily recognized in well-sampled aspirates that include both Romanowsky and Papanicolaou stains (Fig. 6.17). Aspirates of the cribriform and tubular subtypes of adenoid cystic carcinoma are characterized by basaloid cells, which are small and oval, with scant clear cytoplasm, dark oval to angulated nuclei, and indistinct intercellular borders (Fig. 6.18). The nuclei are generally uniform in appearance and tend to lack atypia, mitotic activity, or apoptosis. As previously mentioned, the matrix pattern is the characteristic feature that distinguishes adenoid cystic carcinoma from other matrix-containing and basaloid neoplasms. The matrix is present as metachromatic spheres, cylinders, and branching tubules of hyalinized to myxoid material that is homogeneous and acellular - lacking the embedded myoepithelial cells of pleomorphic adenoma (Fig. 6.19). Occasionally the matrix can be mucoid. The matrix has sharply defined borders, and the basaloid epithelial cells surround the matrix spheres with a well demarcated interface. In

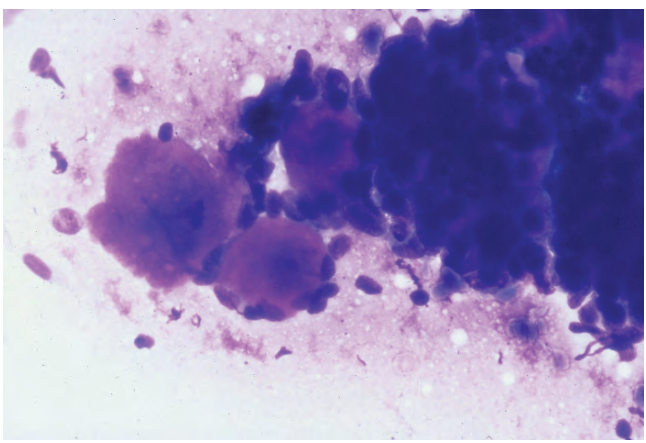


FIG. 6.17. Adenoid cystic carcinoma. Characteristic spheres of metachromatic matrix are surrounded by basaloid cells. (Smear; Diff-Quik).

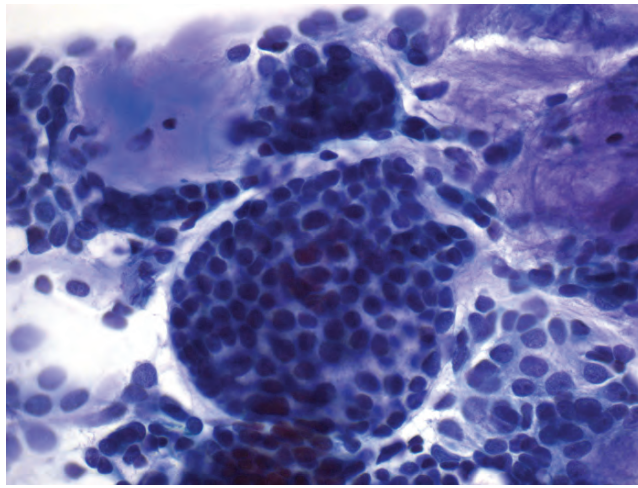


FIG. 6.18. Adenoid cystic carcinoma. The basaloid cells have scant pale-staining cytoplasm and oval to angulated dark nuclei. (Smear; Papanicolaou).

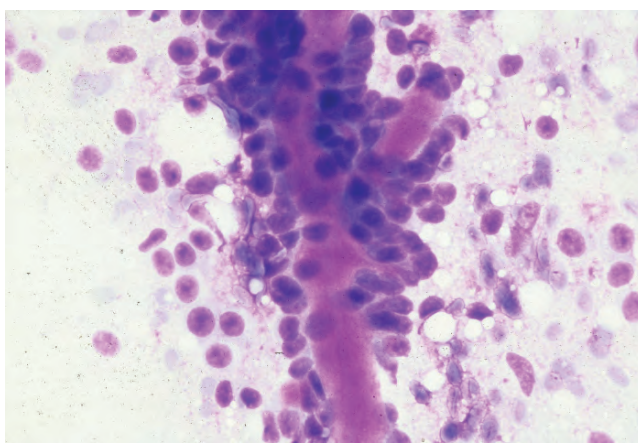


FIG. 6.19. Adenoid cystic carcinoma. In this example, the metachromatic matrix is a branching tubular structure. (Smear; Diff-Quik).

Papanicolaou-stained preparations, the matrix often appears pale blue-green or even colorless, and in some cases, can be very difficult to appreciate (Figs. 6.20–6.21). Smear artifacts can also result in the matrix of adenoid cystic carcinoma having a focal fibrillary appearance, reminiscent of pleomorphic adenoma, but the predominant matrix pattern is that of large acellular metachromatic geometric structures (Fig. 6.22).

Cytologic Features of the Cribriform and Tubular Subtypes of Adenoid Cystic Carcinoma

- Uniform basaloid cells
- Scant clear cytoplasm
- Dark angulated nuclei
- Metachromatic matrix:
  - Spheres, cylinders, branching tubules
  - Hyalinized to myxoid
  - Acellular
  - Homogeneous
  - Sharp well-defined edges

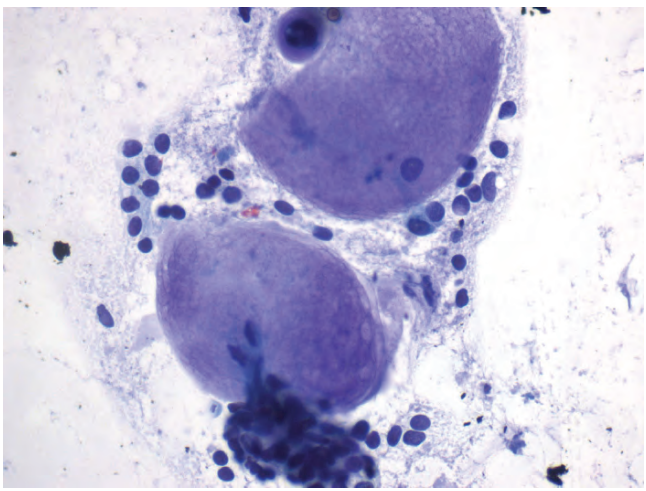


FIG. 6.20. Adenoid cystic carcinoma. In some Papanicolaou-stained preparations, the matrix is easily detected. (Smear; Papanicolaou).

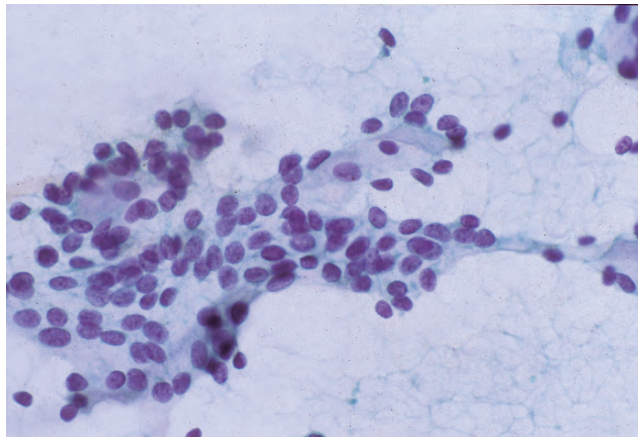


FIG. 6.21. Adenoid cystic carcinoma. In this example, the matrix is pale to colorless using a Papanicolaou stain. (Smear; Papanicolaou).

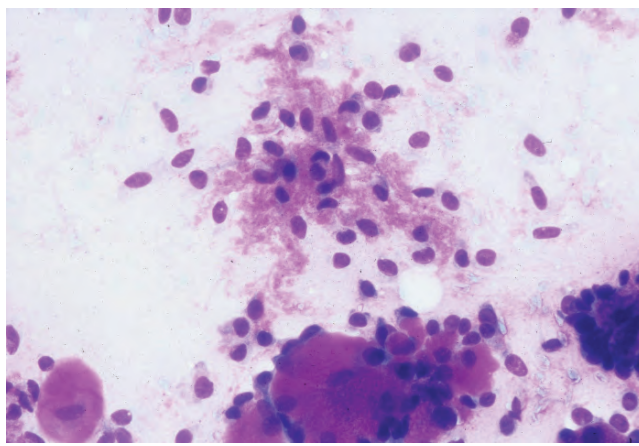


FIG. 6.22. Adenoid cystic carcinoma. Occasionally, the matrix can focally resemble that of pleomorphic adenoma. (Smear; Papanicolaou).

The solid or high-grade form of adenoid cystic carcinoma is the most challenging to recognize in aspirates and is comprised of atypical basaloid cells without significant amounts of the characteristic

metachromatic matrix material (Figs. 6.23–6.24). Aspirates contain crowded 3-dimensional groups of small basaloid cells with scant cytoplasm and dark angulated nuclei that can show significant degrees of nuclear membrane irregularities and pleomorphism. The

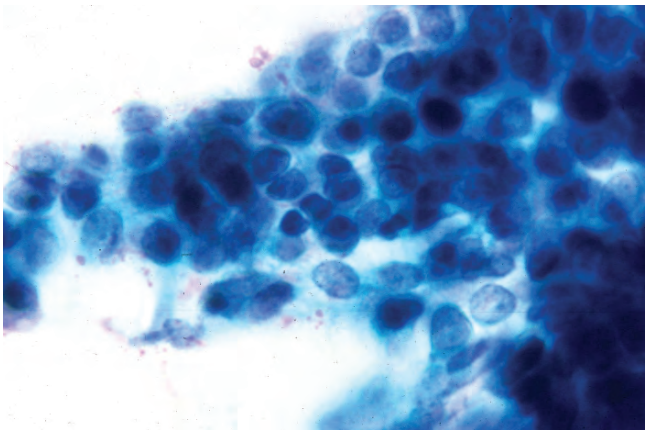


Fig. 6.23. Solid subtype of adenoid cystic carcinoma. The basaloid cells are moderately atypical and form a 3-dimensional crowded group without associated matrix. (Smear; Papanicolaou).

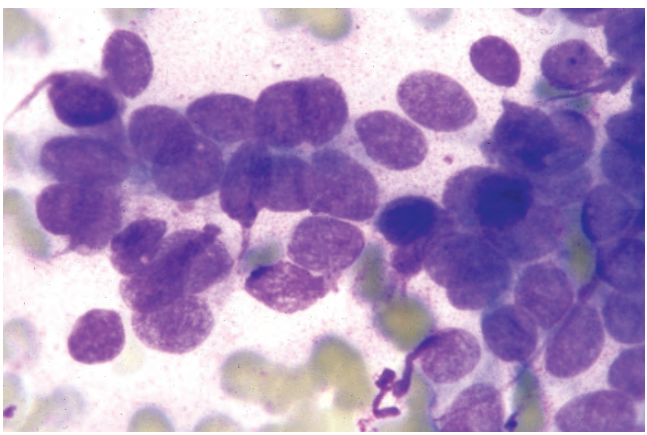


Fig. 6.24. Solid subtype of adenoid cystic carcinoma. The cells are basaloid but lack significant atypia. (Smear; Diff-Quik).

chromatin is coarsely granular, and mitotic activity, apoptotic cells, and background necrosis can also occasionally be seen.

Cytologic Features of the Solid Subtype of Adenoid Cystic Carcinoma

- Crowded 3-D groups of basaloid cells
- Scant clear cytoplasm
- Coarse chromatin
- Variable nuclear pleomorphism
- Variable mitotic activity and apoptotic cells
- Scant to absent matrix

Differential Diagnostic Considerations

The differential diagnosis of matrix-containing tumors includes a number of entities, but the most important differential is between pleomorphic adenoma and adenoid cystic carcinoma since the implications for clinical management and prognosis are critical. When dealing with classic forms of these two entities, FNA is highly accurate; however, for lesions that are poorly sampled, for those where both Romanowsky and Papanicolaou stained preparations are not available, and for cellular or solid (i.e., matrix-poor) cases, the distinction can be more difficult.

Differential Diagnosis of Matrix-Containing Tumors

- Pleomorphic adenoma
- Adenoid cystic carcinoma
- Basal cell adenoma & adenocarcinoma
- Myoepithelioma
- Polymorphous low grade adenocarcinoma
- Epithelial-myoepithelial carcinoma
- Carcinoma ex pleomorphic adenoma

The cytologic diagnosis of the most frequent and conventional forms of pleomorphic adenoma and adenoid cystic carcinoma is generally straightforward (Table 6.1), provided the sample is adequate and well prepared. The most important distinction lies

TABLE 6.1. Cytologic comparison of conventional pleomorphic adenoma and adenoid cystic carcinoma.

	Pleomorphic Adenoma	Adenoid Cystic Carcinoma
Matrix	Fibrillar, indistinct frayed edges, embedded myoepithelial cells	Homogeneous spheres and tubules, sharp well-defined edges, acellular
Cells	Often predominantly myoepithelial cells	Basaloid with dark angulated nuclei
Atypia	Minimal	Variable

in the nature of the matrix material - fibrillar with frayed edges and embedded cells in pleomorphic adenoma versus the matrix of adenoid cystic carcinoma which forms acellular spheres and tubules with sharply defined edges. Both are metachromatic using Romanowsky stains. Although more subtle as a feature, pleomorphic adenomas are more often comprised predominantly of myoepithelial cells, while adenoid cystic carcinomas are characteristically comprised predominantly of basaloid cells with dark angulated nuclei. Rarely, pleomorphic adenomas can contain focal adenoid cystic-type matrix and in such instances, caution is warranted to avoid making a false positive diagnosis (Fig. 6.25).

The real difficulty in diagnosing either pleomorphic adenoma or adenoid cystic carcinoma occurs when dealing with the cellular and solid forms respectively. When there is insufficient matrix material present, caution is warranted in interpreting the aspirate, and a descriptive diagnosis should be used that includes a differential diagnosis. To make matters worse, cellular pleomorphic adenomas comprised predominantly of epithelial cells can appear basaloid, closely mimicking the solid subtype of adenoid cystic carcinoma and possibly resulting in a false positive diagnosis. If the pleomorphic adenoma is well sampled, at least focal amounts of fibrillar matrix material may be present to aid in the recognition of the tumor, but unequivocal distinction is sometimes not possible. In contrast to cellular pleomorphic adenomas, solid adenoid cystic carcinomas are comprised of cells that are more hyperchromatic, and angulated, and as mentioned previously, will often exhibit at least moderate degrees of nuclear atypia, sometimes with mitoses, apoptosis, and necrosis, which are not characteristics of pleomorphic adenoma.

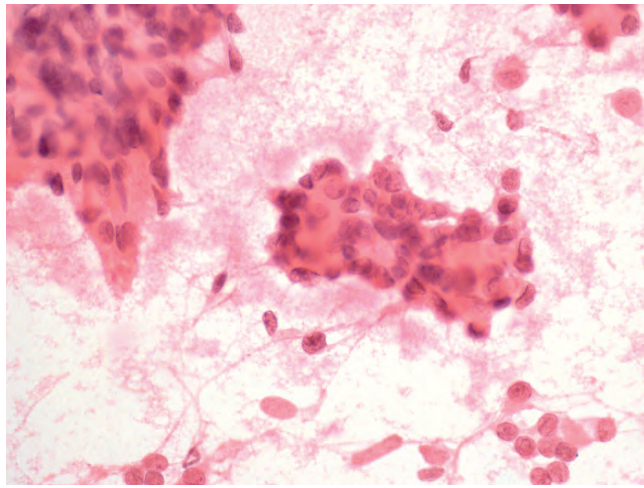


FIG. 6.25. Pleomorphic adenoma with focal adenoid cystic-like matrix. (Smear; Papanicolaou).

When there is insufficient matrix material present, caution is warranted in interpreting the aspirate; a descriptive diagnosis is used that includes a differential diagnosis.

The differential diagnosis of salivary tumors with basaloid features will be discussed in more detail in Chapter 7, but a brief summary of distinguishing features will be mentioned here. In contrast to both cellular pleomorphic adenoma and adenoid cystic carcinoma, basal cell adenoma and adenocarcinoma (which are indistinguishable based upon cytologic criteria alone) are characterized by peripheral bands or ribbons of metachromatic matrix, as well as intercellular droplets of matrix material, a dual population of basaloid cells, and focal squamous morules (Fig. 6.26). Other basaloid tumors to consider in the differential diagnosis with solid adenoid cystic carcinoma include basal cell carcinoma of the skin and metastatic basaloid squamous carcinoma. In both cases, the patient will almost invariably have a history of overlying cutaneous basal cell carcinoma, or of head and neck squamous cell carcinoma of the hypopharynx or glottic region. This is where clinical correlation can be very helpful! Basal cell carcinoma is moderately atypical and

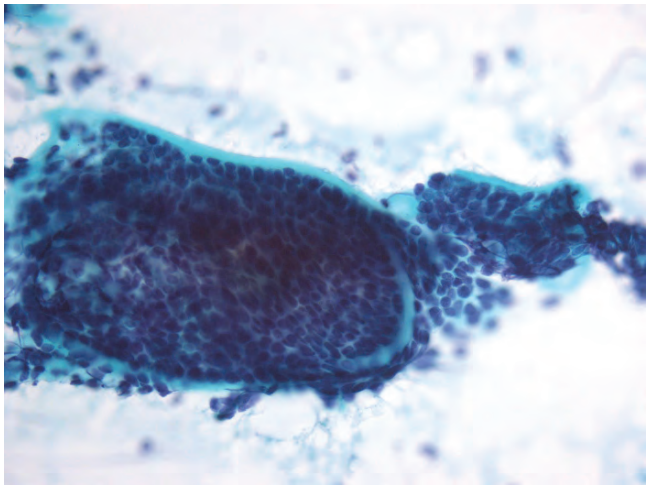


FIG. 6.26. Basal cell adenoma. The aspirate is comprised of groups of basaloid cells with peripheral palisading and a surrounding ribbon of basement membrane material. (Smear; Papanicolaou).

features peripheral palisading of basaloid cells. Basaloid squamous carcinoma is a high-grade malignancy that will often exhibit at least focal squamous features if well sampled (see Chapter 7). Squamous differentiation is not a feature of adenoid cystic carcinomas and can be used to distinguish adenoid cystic carcinoma from several of the above-mentioned basaloid tumors.

Squamous differentiation is not a feature of adenoid cystic carcinoma, and can be used to distinguish it from other basaloid salivary gland tumors.

Myoepithelial-predominant pleomorphic adenomas raise the question of myoepithelioma; however, for clinical management, the distinction is really one of semantics. A more clinically important entity to consider in the differential diagnosis is epithelial-myoepithelial carcinoma (see Chapter 12). Epithelial-myoepithelial carcinomas have larger clear myoepithelial cells than pleomorphic adenomas, as well as acellular myxoid matrix, but the best distinguishing feature is the characteristic biphasic population of cells

that can be further highlighted using immunohistochemical stains for keratin or smooth muscle actin in cell block material. Some epithelial-myoepithelial carcinomas will exhibit spheres of acellular proteincaceous material that in Papanicolaou-stained preparations resemble the matrix spheres of adenoid cystic carcinoma, but the latter is more basaloid than clear and also lacks a prominent biphasic pattern.

Some cases of myoepithelial-predominant pleomorphic adenoma are composed of spindled cells, suggesting other spindled cell tumors of the head and neck region, particularly schwannoma (see Chapter 11). An immunohistochemical profile indicative of myoepithelial differentiation (keratin+, smooth muscle actin+, calponin+) versus strong diffuse staining for S-100 can be used to distinguish these two entities. The presence of focal palisading also favors a schwannoma, although palisading can also rarely be seen in pleomorphic adenomas.

Pleomorphic adenomas with any combination of cystic change or squamous or mucinous metaplasia can suggest the differential diagnosis of a low-grade mucoepidermoid carcinoma. In some cases, it will not be possible to exclude the latter, and a descriptive diagnosis should be made. Distinguishing features that are not found in mucoepidermoid carcinomas include the presence of myoepithelial cells, chondromyxoid matrix material, or keratin pearls.

Cytologic Features of Polymorphous Low-Grade Adenocarcinoma

A matrix-containing tumor that is often confused with pleomorphic adenoma and adenoid cystic carcinoma is polymorphous low-grade adenocarcinoma (PLGA). Formerly known as terminal duct carcinoma, PLGA is a low-grade malignant tumor predominantly of the subepithelial minor salivary glands in the oral cavity and particularly the palate, where over 60% of cases occur. While it is encountered over a broad age range, the majority of patients are between 50 and 70 years old. Unlike adenoid cystic carcinoma, it has an excellent clinical prognosis.

In histologic samples, PLGA is neurotropic and its histomorphologic pattern is often compared with lobular carcinoma of the breast. Cytologically, PLGA is characterized by a monomorphic

population of cuboidal to columnar cells with moderate amounts of pale eosinophilic cytoplasm. Nuclei are oval with open, finely stippled chromatin and indistinct nucleoli (Figs. 6.27–6.28).

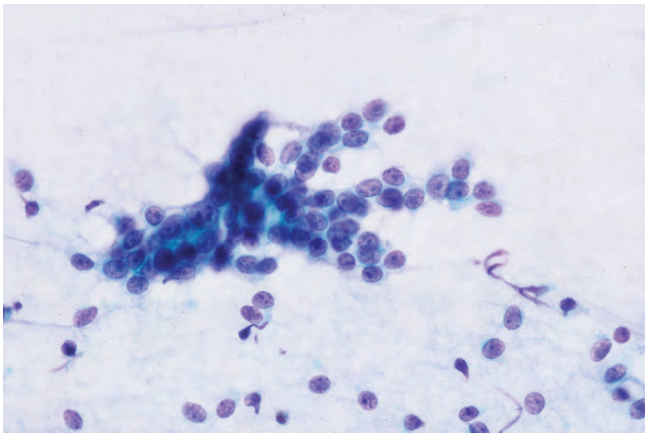


FIG. 6.27. Polymorphous low grade adenocarcinoma. The cells have oval nuclei with stippled chromatin, and are arranged in papillary groups. (Smear; Papanicolaou).

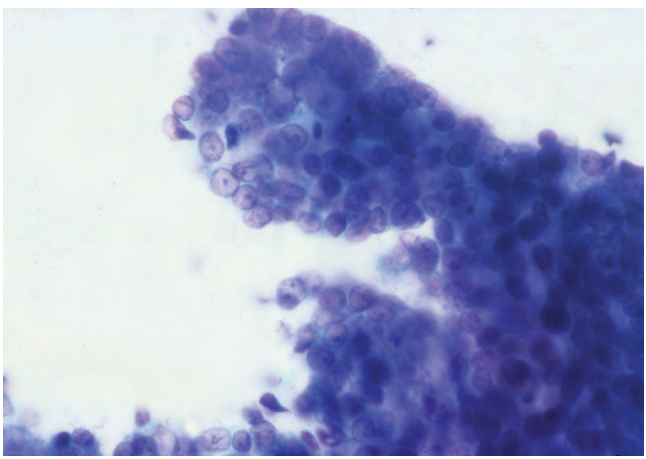


FIG. 6.28. Polymorphous low grade adenocarcinoma. Large crowded group of monomorphic cells with open chromatin and small nucleoli. (Smear; Papanicolaou).

Variable cytomorphologic patterns are seen, including single cells, papillary groups, small cohesive ductal groups, and trabeculae. Extracellular matrix material is usually scant but can include fibrillar arrays of myxoid material similar to pleomorphic adenoma as well as focal acellular spheres similar to those in adenoid cystic carcinoma (Fig. 6.29). In contrast to adenoid cystic carcinoma, the cells of PLGA are not basaloid; they are more cuboidal to columnar with moderate amounts of cytoplasm and with pale vacuolated nuclei. The variety of cytomorphologic patterns, especially the presence of papillary groups, helps to distinguish PLGA from both pleomorphic adenoma and adenoid cystic carcinoma. Nonetheless, cytologic overlap between these 3 lesions can be seen, and while PLGA may be strongly favored, a definitive diagnosis of PLGA may not be possible. Even in histologic samples (especially small biopsies), the distinction between PLGA and pleomorphic adenoma or adenoid cystic carcinoma can be challenging!

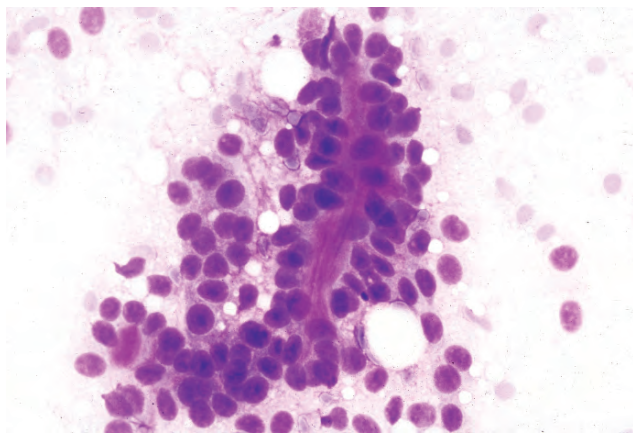


Fig 6.29. Polymorphous low grade adenocarcinoma. In this example, the cells surround metachromatic matrix material reminiscent of adenoid cystic carcinoma. (Smear; Diff-Quik).

Cytologic Features of Polymorphous Low-Grade Adenocarcinoma

- Monomorphous population of cuboidal to columnar cells
- Plump oval nuclei with indistinct nucleoli
- Open stippled chromatin
- Pale eosinophilic cytoplasm
- Scant myxoid matrix
- Variable cytomorphologic patterns:
  - Papillary
  - Single cell
  - Trabecular
  - Ductal

In contrast to adenoid cystic carcinoma, the cells of PLGA are not basaloid; they are more cuboidal to columnar with moderate amounts of cytoplasm and with pale vacuolated nuclei.

Ancillary Techniques

Standard immunocytochemical markers are generally not very useful in the evaluation of salivary gland tumors, particularly for those that are matrix-containing. This group of neoplasms share a similar cell composition of epithelial and myoepithelial cells, and hence, they also share a similar immunohistochemical profile. In our experience, immunohistochemical studies can be performed on destained aspirates, cytospins, and thin-layer preparations, but the best approach is to use paraffin-embedded cell block material for ancillary marker studies. Pleomorphic adenomas and other matrix-containing salivary gland tumors exhibit immunoreactivity for vimentin and cytokeratin, and are variably positive for smooth muscle actin, calponin, GFAP, p63, CD10, muscle specific actin, and S-100. This immunoprofile reflects the component myoepithelial cell. Immunohistochemical markers are more commonly used in the small subset of cases where the differential diagnosis is between a salivary gland tumor and a mesenchymal or metastatic

tumor, or for distinguishing myoepithelial-containing tumors from other salivary gland tumors that lack myoepithelial cells (e.g., mucoepidermoid carcinoma, acinic cell carcinoma, oncocytoma). Ductal cells of the matrix-containing tumors are positive for pan-cytokeratin, EMA, and CEA. Interestingly, some pleomorphic adenomas are also positive for prostate-specific antigen. Most pleomorphic adenomas have a low proliferation index and thus are not very immunoreactive with Mib-1 (low Ki-67 index) or proliferating cell nuclear antigen (PCNA), while adenoid cystic carcinoma has been reported to have a moderate Ki-67 index of 20% and higher.

Immunoprofile of Myoepithelium in Matrix-Containing Tumors

- Vimentin +
- Keratin +
- Smooth muscle actin ±
- Calponin ±
- S-100 ±
- Muscle specific actin ±
- GFAP ±
- P63 ±
- CD10 ±

While a majority of pleomorphic adenomas have karyotypic abnormalities, cytogenetic changes involving 8q12 are the most frequent, reflecting the dysregulation of PLAG1, a zinc finger gene. The various cytogenetic changes that have been detected leading to altered expression of PLAG1 are believed to play a major role in the tumorigenesis of pleomorphic adenomas. It is not surprising that rearrangements of 8q12 have also been identified as a frequent finding in carcinoma *ex* pleomorphic adenomas which probably result from the acquisition by pleomorphic adenomas of additional genetic mutations leading to the malignant phenotype.

Research efforts to identify a specific molecular change driving the carcinogenesis of adenoid cystic carcinomas have been largely unsuccessful to date. Within the past several years, several groups have demonstrated an unusual feature of adenoid cystic carcinomas – the majority overexpress CD117, also known

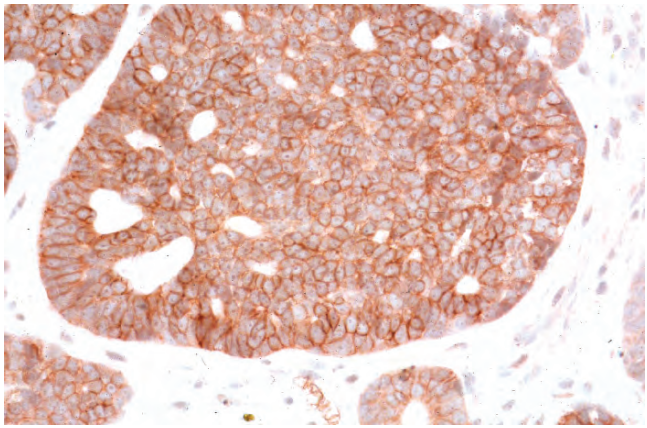


FIG. 6.30. Adenoid cystic carcinoma. Strong immunoreactivity for CD117 (kit). (Biopsy; immunohistochemical reaction).

as KIT, a transmembrane receptor-type tyrosine kinase, and immunohistochemical stains can be used to demonstrate CD117 immunoreactivity (Fig. 6.30). This marker is probably best known for its association with gastrointestinal stromal tumors where mutations in the KIT gene have led to specific successful treatment strategies. Unfortunately, a specific mutation of KIT, similar to that in GISTs, has not been identified in adenoid cystic carcinomas, and CD117 immunoreactivity is employed simply as an ancillary marker but is not considered to have biological significance. Since CD117 immunoreactivity is not specific for adenoid cystic carcinoma and can in fact be seen focally within pleomorphic adenomas, it is probably best used with caution, and may be better suited to larger resection specimens than to small biopsies or cell block material. Some studies indicate that polymorphous low-grade adenocarcinomas are negative for CD117 immunoreactivity so that this marker may have limited value for distinguishing it from adenoid cystic carcinoma. Recent investigations have identified deletions on chromosome 6q in over half of adenoid cystic carcinoma cases, raising the possibility that a tumor suppressor might be found in this region, but further research is needed.

Clinical Management and Prognosis

Malignant transformation to carcinoma *ex* pleomorphic adenoma occurs in an estimated 5%–10% of pleomorphic adenomas and is the driving force for surgical excision in most cases. Because pleomorphic adenomas present primarily within the superficial lobe of the parotid gland, surgical management consists of a superficial parotidectomy with dissection and preservation of the facial nerve. For the 10% of pleomorphic adenomas occurring within the deep lobe of the parotid gland, a total parotidectomy with facial nerve preservation is required. Within the submandibular gland or within a minor salivary gland, the pleomorphic adenoma is surgically excised with a cuff of normal surrounding tissue to help prevent the possibility of recurrence.

A major clinical problem associated with the management of pleomorphic adenomas is recurrence in 3%–7% of cases, which is more common in the stromal-rich variants. The explanation for recurrences is related in part to the fact that pleomorphic adenomas are variably encapsulated and frequently exhibit small buds of tumor protruding into surrounding salivary gland parenchyma. Recurrent pleomorphic adenomas often present as multifocal tumor nodules that can defy surgical management. For those tumors which are not completely excised at initial surgery, recurrent pleomorphic can result in significant morbidity for the patient, as well as present a lifelong risk for malignant transformation to carcinoma *ex* pleomorphic adenoma. Postoperative radiotherapy is sometimes used for pleomorphic adenomas that are "spilled" during excision and for recurrent pleomorphic adenomas. FNA is sometimes used to monitor patients who are not surgical candidates or who have recurrent pleomorphic adenoma for evidence of malignant transformation. The latter is typically high grade (see chapter 10) and easily recognized by FNA. A very rare occurrence that has been associated with multiply recurrent pleomorphic adenomas is the metastasizing pleomorphic adenoma (aka metastasizing benign mixed tumor). Cytologic and histologic evaluation of these tumors does not reveal microscopic evidence predictive of a metastatic behavior.

Clinical Problems Associated with Pleomorphic Adenoma

- Recurrent pleomorphic adenoma (3%–7% of cases)
- Malignant transformation to carcinoma *ex* pleomorphic adenoma (5–10%)
- Rare metastasizing pleomorphic adenoma
- Carcinosarcoma *ex* pleomorphic adenoma

For adenoid cystic carcinoma, the standard treatment is radical excision of the local tumor followed by postoperative radiotherapy to reduce the local recurrence rate. The radical surgical excision usually includes sacrifice of the facial nerve and/or other associated nerves. For surgically unresectable tumors, radiotherapy alone is used; thus far, there is no proven role for chemotherapy as a treatment option. Prognosis for the cribriform and tubular subtypes is 60%–90% at 5 years, but drops markedly to 25%–40% at 15 years. Thus while initially behaving as an indolent tumor, the long-term survival rate is very poor with late local recurrences, base of skull involvement, and distant metastases to sites such as lung, bone, brain, and liver. Lymph node involvement is uncommon. Even worse, tumors that are comprised of greater than 30% of the solid subtype have a 5-year survival of less than 20% and a 15-year survival rate of approximately 5%. Other features that have been associated with poor prognosis include advanced clinical stage, large tumor size, perineural and bone invasion, positive surgical margins, and high proliferation index. In a small subset of adenoid cystic carcinomas, dedifferentiation into a more rapidly aggressive high grade malignancy can also occur.

Suggested Reading

Castle JT, Thompson LD, Frommelt RA, Wenig BM, Kessler HP. Polymorphous low grade adenocarcinoma: a clinicopathologic study of 164 cases. Cancer 1999;86:207–219.

Chandon VS, Wilbur D, Faquin WC, Khurana KK. Is c-kit (CD117) immunolocalization in cell block preparations useful in the differentiation of adenoid cystic carcinoma from pleomorphic adenoma. Cancer Cytopathol 2004;102:207–209.

Chen I, Tu H. Pleomorphic adenoma of the parotid gland metastasizing to the cervical lymph node. Otolaryngol Head Neck Surg 2000;122:455–457.

Hocwald E, Korkmaz H, You GH et al. Prognostic factors in major salivary gland cancer. Laryngoscope 2001;111:1434.

Mino M, Pilch BZ, Faquin WC. Expression of KIT (CD117) in neoplasms of the head and neck: an ancillary marker for adenoid cystic carcinoma. Mod Pathol 2003;16:1224–1231.

Pantanowitz L, Goulart RA, Cao QJ. Salivary gland crystalloids. Diagn Cytopathol 2006; 34 :749–750.

Rutherford S, Yu Y, Rumpel CA, Frierson HF, Moskaluk CA. Chromosome 6 deletion and candidate tumor suppressor genes in adenoid cystic carcinoma. Cancer Letters 2006;236:309–317.

Spiro RH, Huvos AG. Stage means more than grade in adenoid cystic carcinoma. Am J Surg 1992;164:623.

Voz ML, Agten NS, Van de Ven WJ, Kas K. PLAG1, the main translocation target in pleomorphic adenoma of the salivary glands, is a positive regulator of IFG-II. Cancer Res 2000;60:106–113.

7
Basaloid Tumors: Basal Cell Adenoma and Basal Cell Adenocarcinoma

Background

Basaloid tumors of the salivary gland are among the most diagnostically challenging areas of salivary gland FNA cytopathology. The primary tumors included in this group are basal cell adenoma, basal cell adenocarcinoma, and the solid variant of adenoid cystic carcinoma. In addition, various other salivary gland tumors, such as cellular pleomorphic adenoma, can also exhibit basaloid features and will be considered as differential diagnostic entities within this section.

Basal cell adenomas are rare salivary gland tumors comprised of basaloid cells and lacking the chondromyxoid matrix material characteristic of pleomorphic adenomas. In the past, they have been classified as "monomorphic adenomas," but this nonspecific terminology is to be avoided in favor of the more specific designation recommended by the WHO - "basal cell adenoma." Basal cell adenomas represent 1%–3% of all salivary gland neoplasms, and they arise primarily in older adults, usually in the sixth to seventh decade. A somewhat histologically similar basaloid tumor that occurs in infants is known as sialoblastoma. Over 75% of basal cell adenomas occur in the parotid gland; they are rarely seen in minor salivary glands. There are 3 subtypes of basal cell adenoma: solid, tubulotrabecular, and membranous. Most patients present with a solitary firm nodule between 1 and 3 cm that is slowly enlarging. Occasionally, basal cell adenomas can be cystic.

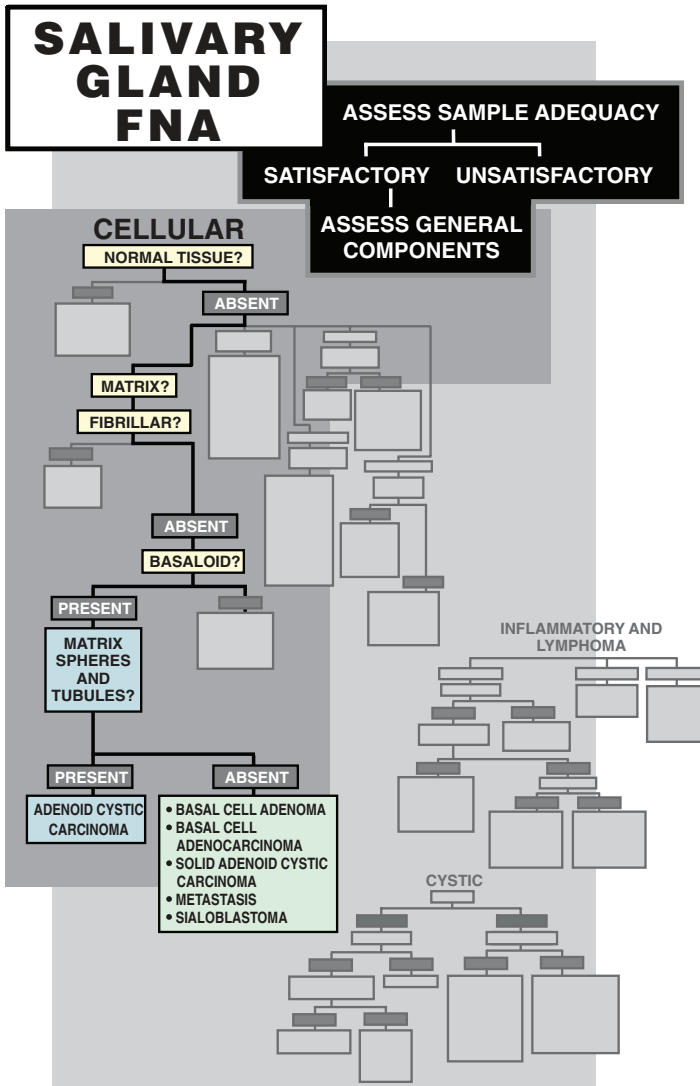


FIG. 7.1. Algorithm for basaloid tumors.

The membranous subtype of basal cell adenoma is the most cytologically and histologically distinctive of the 3 subtypes. In contrast to the tubulotrabecular and solid subtypes, the membranous subtype is often multinodular and sometimes multifocal. It is also unusual because of its occasional association with multiple synchronous dermal cylindromas, trichoepitheliomas, and spiradenomas, to which basal cell adenoma can bear a remarkable microscopic resemblance. For this reason, the membranous subtype of basal cell adenoma has also been known as "dermal analogue tumor." The condition of multiple cutaneous adnexal tumors and synchronous salivary gland basal cell adenomas, which can be disfiguring, is called Brooke-Spiegler syndrome. It is an autosomal dominant disease caused by mutations in the tumor suppressor gene that encodes the CYLD protein (an inhibitor of NF-kB).

Basal cell adenocarcinoma is a rare salivary gland neoplasm that is the malignant counterpart of basal cell adenoma. It is a low-grade malignancy with a very good clinical prognosis; although it has a tendency for local recurrence (approximately 35% of cases), metastatic disease is uncommon. Basal cell adenocarcinoma accounts for less than 2% of malignant epithelial salivary gland tumors. The majority occur in the superficial lobe of the parotid gland, although occasional cases have been reported in the submandibular gland and the minor salivary glands. The average age at diagnosis is 60 years, with a broad age range from third to tenth decade, with no gender predilection. Salivary gland enlargement is the main presenting symptom, and uncommonly, mild pain or tenderness may also be present. Like its benign counterpart, basal cell adenocarcinomas can be solid, tubulotrabecular, or the membranous subtype. The solid subtype is the most common. Most basal cell adenocarcinomas are believed to arise *de novo*, although a small subset may develop from a pre-existing basal cell adenoma.

FNA is highly sensitive at detecting basaloid neoplasms such as basal cell adenoma and adenocarcinoma, but distinction between several of the basaloid entities in the differential diagnosis is often not possible. As will be discussed, some cases of basal cell tumor can be recognized by FNA, but many will receive a descriptive signout and differential diagnosis. Most basal cell adenocarcinomas are microscopically identical to basal cell adenomas except for the presence of an invasive histologic growth pattern. Because FNA does not

detect parenchymal invasion, basal cell adenomas and adenocarcinomas are, for the most part, indistinguishable by FNA.

General Diagnostic Approach

Using the algorithm (Fig. 7.1), aspirates comprised of epithelial cells that lack the characteristic matrix of pleomorphic adenoma, and that exhibit basaloid cytologic features (scant cytoplasm and dark nuclei), lead to a differential diagnosis that includes adenoid cystic carcinoma, basal cell adenoma, basal cell adenocarcinoma, and other lesions with basaloid features. The classic cribriform type of adenoid cystic carcinoma is basaloid but can be distinguished by its acellular matrix spheres and branching matrix tubules; however, the solid form of adenoid cystic carcinoma must be considered in the differential diagnosis with the solid form of basal cell adenoma and adenocarcinoma.

Diagnostic Criteria

Basal cell adenoma and adenocarcinoma are classic basaloid tumors that in most cases exhibit identical cytomorphologic features – basal cell adenocarcinoma is distinguished from basal cell adenoma by an infiltrative growth pattern in histologic specimens. There is a small subset of basal cell adenocarcinomas that exhibits nuclear atypia and may show mitotic activity and/or necrosis. Such features are rare in basal cell adenocarcinomas, but when present, would exclude basal cell adenoma. For the majority of FNA cases, a general rule of thumb is that basal cell adenoma and basal cell adenocarcinoma cannot be reliably distinguished on the basis of cytologic features.

Basal cell adenoma and basal cell adenocarcinoma cannot be reliably distinguished on the basis of cytologic features.

The three histologic subtypes of basal cell adenoma and adenocarcinoma share certain cytologic features (Fig. 7.2, Table 7.1). Aspirates are cellular, and the cytologic diagnosis of either

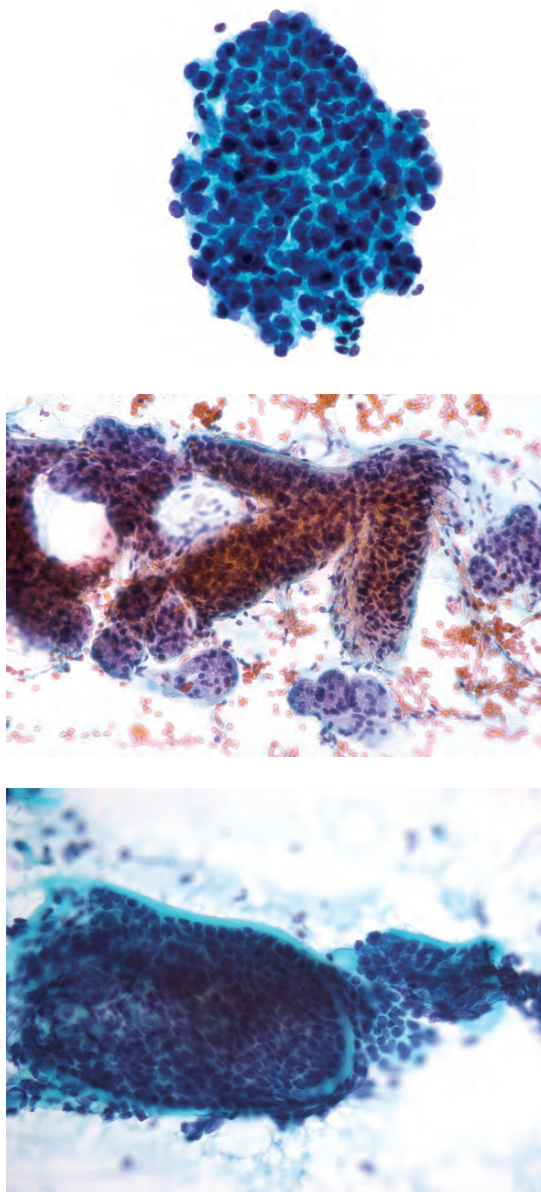


FIG. 7.2. Basaloid tumor. There are three subtypes of basal cell adenoma and adenocarcinoma: solid (A), tubulotrabecular (B), and membranous (C). (Thin-layer preparation, Papanicolaou.)

TABLE 7.1. Cytologic features of the three subtypes of basal cell adenoma and adenocarcinoma.

Subtype	Cytoarchitecture	Characteristic Cytology	FNA Diagnosis
Solid	Fragmented groups of haphazard basaloid cells	Squamous morules and intercellular matrix droplets	Usually descriptive
Tubulotrabecular	Branching tubules	Thin peripheral matrix ribbon	Sometimes diagnostic
Membranous	Cohesive trabecular and insular groups	Thick peripheral matrix ribbon	Usually diagnostic

basal cell adenoma or adenocarcinoma rests on identifying two populations of basaloid cells: a group of small oval cells with bland hyperchromatic nuclei, scant cytoplasm, and indistinct nucleoli, and a group of larger oval to polygonal cells with moderate amounts of delicate pale cytoplasm (Fig. 7.3). The basaloid cells are uniform and haphazardly arranged in variably-sized clusters or trabeculae, often with peripheral palisading of the smaller population of cells (Fig. 7.4). Squamous morules, a characteristic feature of basal cell tumors, are sometimes present in well-sampled cases (Fig. 7.5). All three subtypes can have small dense, nonfibrillary intercellular globules of acellular matrix material that is blue-green using Papanicolaou stains and metachromatic using Diff-Quik stains (Fig. 7.6).

Shared Cytologic Features of Basal Cell Adenoma and Adenocarcinoma

- Cellular aspirate
- Two populations of basaloid cells
- Haphazardly arranged cells
- Peripheral palisading
- Squamous morules
- Intercellular matrix globules

The tubulotrabecular subtype of basal cell adenoma and adenocarcinoma is characterized by branching tubules and trabeculae of basaloid cells with a thin peripheral ribbon of acellular matrix

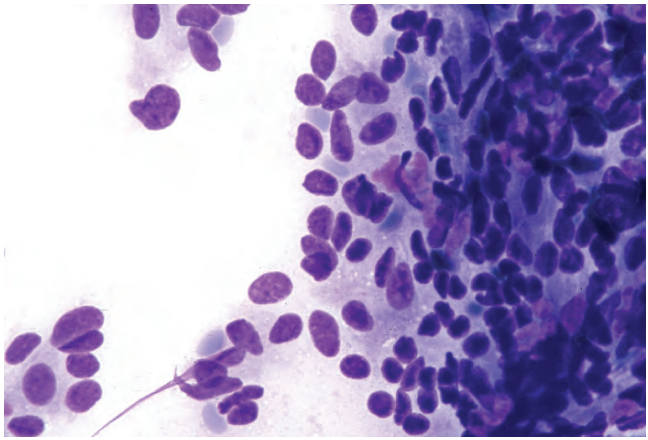


FIG. 7.3. Basal cell adenocarcinoma. Two populations of basaloid cells are present. (Smear, Diff-Quik.)

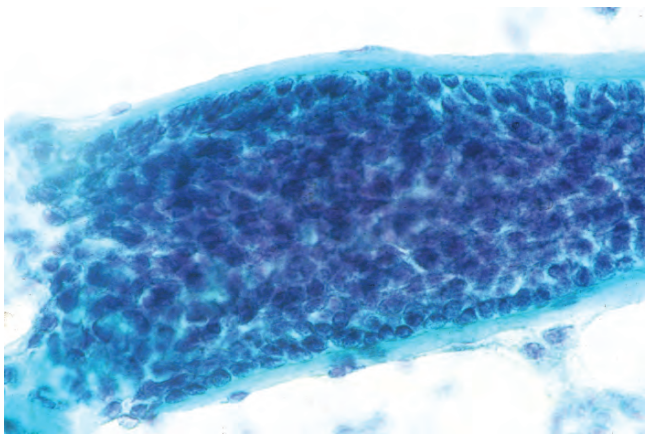


FIG. 7.4. Basal cell adenoma. Palisading of the smaller basaloid cells is often seen along the periphery of the groups. (Smear, Papanicoloau.)

material surrounding the group (Fig. 7.7). The matrix is pale and colorless to blue-green in Papanicolaou stains and is metachromatic using Diff-Quik. In a well-sampled tumor, the cytologic findings are often diagnostic. The membranous subtype exhibits a

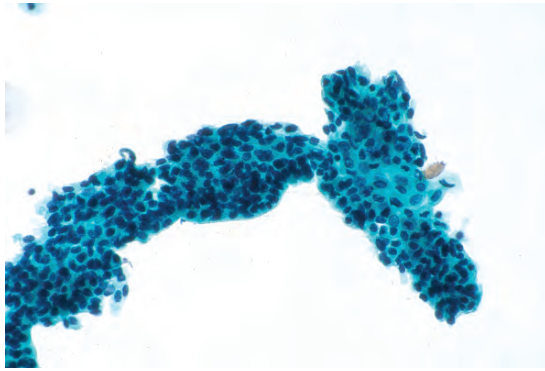


FIG. 7.5. Basal cell adenoma. Squamous morules, if present, are characteristic of well-sampled basal cell tumors. (Smear, Papanicolaou.)

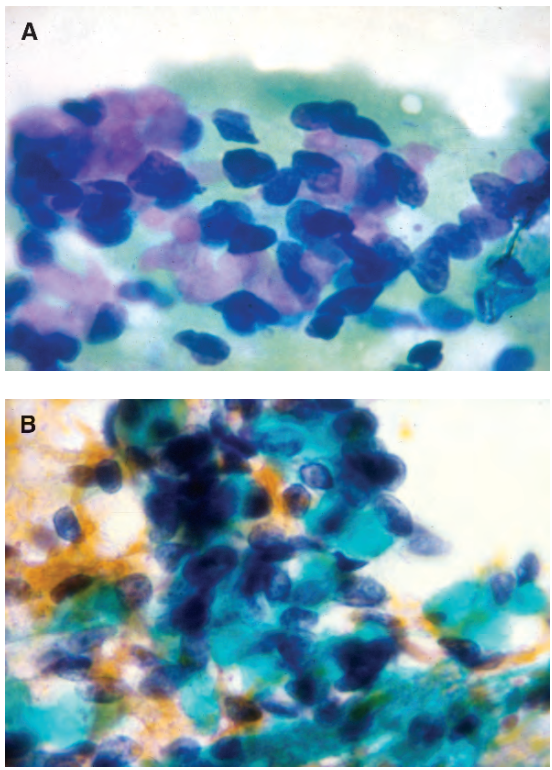


FIG. 7.6. Basal cell adenoma. (A and B) Intercellular globules of acellular matrix material are often present. (A, Smear, Diff-Quik; B, Smear, Papanicolaou.)

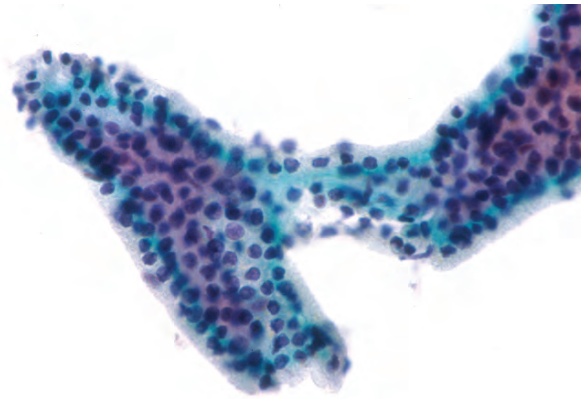


FIG. 7.7. The tubulotrabecular subtype of basal cell tumor consists of basaloid cells in cohesive groups surrounded by a thin peripheral ribbon of basement membrane material. (Thin-layer preparation, Papanicolaou.)

dramatic cytomorphologic pattern consisting of cohesive groups of basaloid cells with peripheral palisading and an impressive thick peripheral band of acellular matrix material (Fig. 7.8). This pattern is unique among salivary gland tumors, and thus the membranous subtype is readily diagnosed by FNA, although one can still not reliably distinguish benign from malignant.

The solid subtype of basal tumor is the most diagnostically problematic (Fig. 7.9). Aspirates consist of fragmented groups of haphazardly arranged basaloid cells, but the characteristic peripheral ribbons of matrix material seen in the tubulotrabecular and membranous subtypes are not present. Squamous morules and intercellular matrix droplets are more commonly found in the solid subtype. When several general cytologic features of basal cell tumors are found (e.g., two populations of basaloid cells, palisading, squamous morules, and intercellular matrix globules), the diagnosis of a basal cell tumor can be strongly suggested, but most cases of the solid subtype will require a descriptive diagnosis.

Most cases of the solid subtype of basal cell tumor will require a descriptive rather than definitive diagnosis.

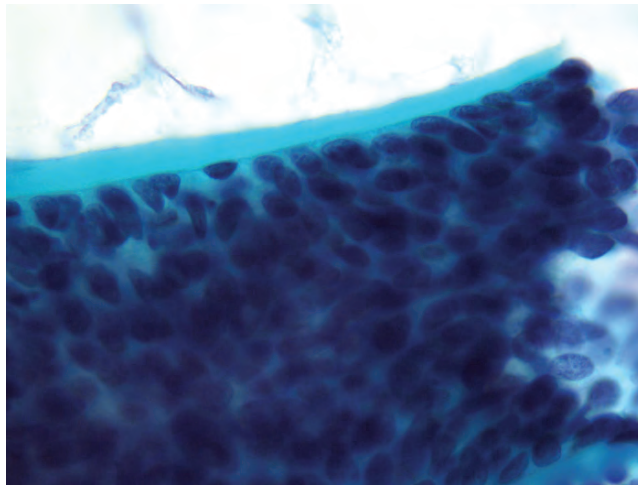


FIG. 7.8. The membranous subtype of basal cell adenoma consists of groups of cells with a very distinctive peripheral band of basement membrane material. (Smear, Papanicolaou.)

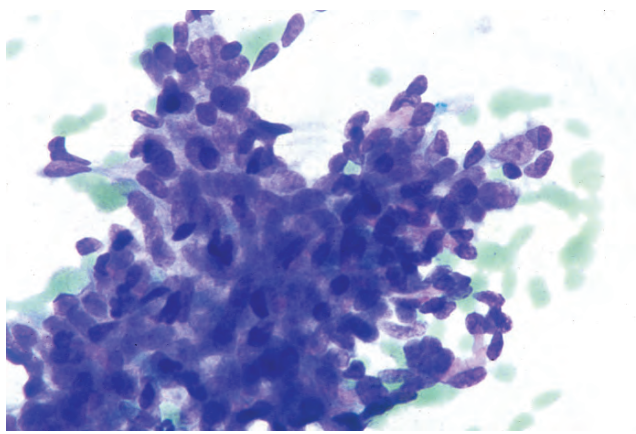


FIG. 7.9. The solid subtype of basal cell adenocarcinoma. Aspirates often exhibit a nonspecific pattern of haphazardly arranged basaloid cells. (Smear, Diff-Quik.)

Differential Diagnosis and Pitfalls

The differential diagnosis of basal cell adenoma and adenocarcinoma includes adenoid cystic carcinoma, cellular pleomorphic adenoma, chronic sialadenitis, cutaneous basal cell carcinoma, and metastatic basaloid squamous carcinoma (Table 7.2). It has been

TABLE 7.2. Cytologic differential diagnosis of selected basaloid lesions.

Cytologic Features	Basal Cell Adenocarcinoma	Basal Cell Adenoma	Pleomorphic Adenoma	Adenoid Cystic Carcinoma	Chronic Sialadenitis
Cytoarchitecture	Cohesive clusters; haphazard; peripheral palisading; squamous morules	Cohesive clusters; haphazard; peripheral palisading; squamous morules	Single cells and groups; haphazard; ductal structures	3-D cylinders and branching groups; mosaic pattern	Small angulated groups
Cells	Two basaloid cell types	Two basaloid cell types	Often myoepithelial predominant + cuboidal cells	Basaloid cells and variable numbers of myoepithelial cells	Low-cuboidal ductal cells, sparse cellularity
Nuclei	Round to oval; Dark; bland	Round to oval; dark; bland	Round to oval with fine chromatin	Oval to angulated; mild to moderate atypia	Round to oval; dark; bland
Stroma	Intercellular matrix globules; peripheral acellular matrix ribbons	Intercellular matrix globules; peripheral acellular matrix ribbons	Fibrillar myxoid matrix; frayed edges; embedded cells;	Branching tubules and spheres; acellular; sharp borders	Absent
Background	Clean with occasional stripped nuclei	Clean with occasional stripped nuclei	Single myoepithelial cells	Clean with occasional stripped nuclei	Mild chronic inflammation

suggested that the cytologic distinction of basaloid neoplasms, particularly the distinction of basal cell adenoma and adenocarcinoma from the solid variant of adenoid cystic carcinoma, may be the most difficult diagnostic problem in the salivary gland (Fig. 7.10)!

Differential Diagnosis of Salivary Gland Basaloid Lesions

- Basal cell adenoma
- Basal cell adenocarcinoma
- Adenoid cystic carcinoma (solid)
- Cellular pleomorphic adenoma
- Chronic sialadenitis
- Cutaneous basal cell carcinoma
- Metastatic basaloid squamous carcinoma

Because basal cell adenocarcinoma is a low-grade malignancy with an excellent prognosis, while adenoid cystic carcinoma is a clinically aggressive cancer that usually requires a more extensive surgical management, it is important to be cautious when diagnosing a basal cell tumor. Distinguishing the membranous and tubulotrabecular subtypes of basal cell tumors from the classic cribriform and tubular subtypes of adenoid cystic carcinoma is feasible given a well-sampled aspirate. In contrast to the three-dimensional branching tubules and cylinders of metachromatic matrix in classic adenoid cystic carcinoma, the tubulotrabecular and membranous subtypes of basal cell tumor have a characteristic peripheral ribbon of matrix. The problem arises when one compares the solid form of basal cell tumor to the solid form of adenoid cystic carcinoma. Matrix is scant to absent, and both are comprised of similar-appearing basaloid cells. Most cases will be signed out descriptively; however, there are a few characteristics which if present, would favor a basal cell tumor: two distinct populations of basaloid cells, intercellular matrix globules, peripheral palisading of cells, squamous morules, and absence of atypia. The solid subtype is the most aggressive form of adenoid cystic carcinoma. It frequently exhibits at least moderate nuclear atypia and apoptotic cells, a finding that is uncommon in the solid subtype of basal cell tumor. In addition, aspirates of adenoid cystic carcinoma are often reported as markedly painful secondary to neurotropism, while this is not typical of a basal cell adenoma or adenocarcinoma.

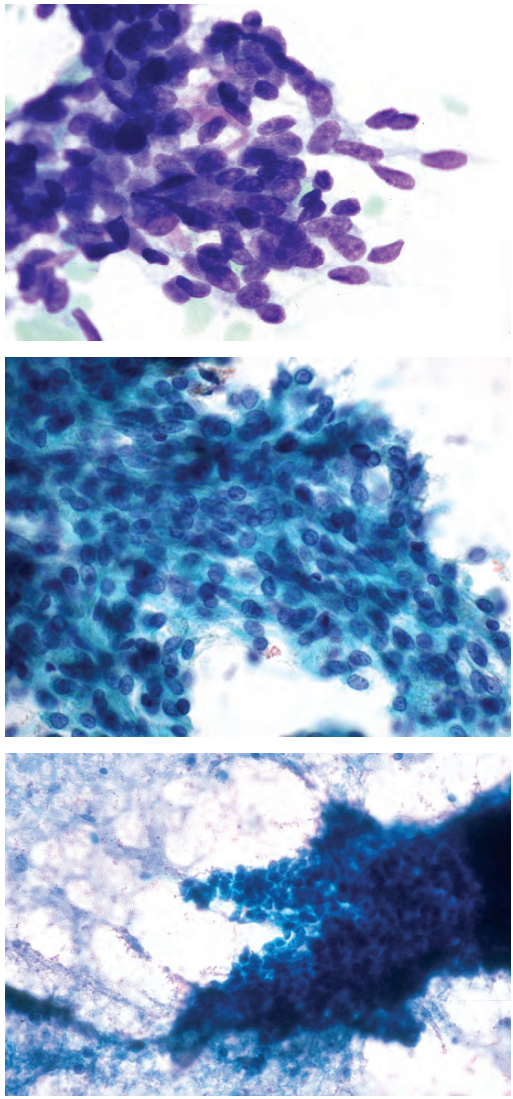


FIG. 7.10. The differential diagnosis of the solid subtype of basal cell adenoma (A), cellular pleomorphic adenoma (B), and solid adenoid cystic carcinoma (C) is among the most challenging and will often require a descriptive diagnosis. (Smears, Papanicolaou.)

Cytologic Features Favoring a Solid Subtype of Basal Cell Tumor Over the Solid Subtype of Adenoid Cystic Carcinoma

- Two distinct populations of basaloid cells
- Peripheral palisading
- Squamous morules
- Intercellular matrix globules
- Absence of atypia
- Nonpainful FNA

A small subset of cellular pleomorphic adenomas with a predominance of basaloid epithelial cells can be difficult to distinguish from other basaloid tumors because the characteristic fibrillar matrix material is sparse. Adequate sampling combined with a careful search for matrix in Diff-Quik stained preparations can be helpful, but when absent, a descriptive diagnosis will be necessary. Chronic sialadenitis is occasionally misinterpreted as a basaloid tumor because the ductal cells present have a low cuboidal basaloid appearance. In contrast to aspirates of basal cell tumors, aspirates of chronic sialadenitis are hypocellular, often bordering on nondiagnostic, cell groups are very small and angulated, and the background contains at least mild chronic inflammation. Aspirates of basal cell carcinoma involving the parotid gland can be very difficult to distinguish from a solid basal cell tumor. This is a good example of where clinical correlation is helpful. Patients with basal cell carcinoma will invariably have a clinical history of an overlying cutaneous skin tumor infiltrating into the deep subcutaneous tissue and involving the parotid gland. In contrast to the cytologically bland appearance of basal cell adenoma and adenocarcinoma, basaloid squamous carcinoma exhibits high-grade cytologic features, and most patients will have a prior history of head and neck squamous cell carcinoma. Polymorphous low-grade adenocarcinoma (PLGA) is sometimes considered in the differential diagnosis of basaloid tumors, but in fact, the cells of PLGA are not truly basaloid. In contrast to the dark nuclei and scant cytoplasm of the basaloid tumors discussed above, the cells of PLGA are more cuboidal to columnar with moderate amounts of cytoplasm and with pale vacuolated nuclei. The pitfall with PLGA is that it can sometimes contain matrix material resembling that seen in the classic form of adenoid cystic carcinoma (see Chapter 6).

Ancillary Techniques

The immunohistochemical profile of basal cell adenoma and adenocarcinoma includes reactivity with markers of both epithelial and myoepithelial differentiation such as cytokeratin, smooth muscle actin, calponin, S-100, and p63. This pattern is nonspecific, being similar to that of many of the other mixed epithelial-myoepithelial tumors of salivary gland origin, including pleomorphic adenoma and adenoid cystic carcinoma. Specific molecular markers of basal cell tumors have not been identified, except for inherited forms of the membranous subtype of basal cell tumor (Brooke-Spiegler syndrome) that contain mutations in the tumor suppressor gene encoding the CYLD protein.

Clinical Management and Prognosis

Basal cell adenomas are treated by complete surgical excision with negative margins, usually involving superficial parotidectomy. Unlike pleomorphic adenomas, which can result in a high degree of morbidity due to recurrent disease, most basal cell adenomas are nonrecurrent. The exception is that approximately one-fourth of membranous basal cell adenomas have been reported to recur; this is probably related to the multinodular nature of this particular subtype. The clinical management of basal cell adenocarcinoma is similar to that of its benign counterpart: conservative surgical resection with negative margins. Basal cell adenocarcinomas are low-grade salivary gland cancers. While they exhibit local infiltrative growth, including focal vascular and perineural invasion detectable by histologic examination, they rarely metastasize, or result in mortality; the overall prognosis is excellent. Local recurrence occurs in about one-third of cases, and is the primary complication associated with this cancer.

Suggested Reading

Choi HR, Batsakis JG, Callender DL, Prieto VG, Luna MA, El Naggar AK. Molecular analysis of chromosome 16q regions in dermal analogue tumors of salivary glands: a genetic link to dermal cylindroma? Am J Surg Pathol 2002;26:778–783.

Ellis G. Basal cell adenocarcinoma. In: Barnes L, Eveson JW, Reichart P. Sidransky D (eds).. World Health Organization Classification of Tumours: Head and Neck Tumours. Lyon: IARC Press, 2005;229–230.

Kawahara A, Harada H, Akiba J, Yokoyama T, Kage M. Fine-needle aspiration cytology of basal cell adenoma of the parotid gland: characteristic cytological features and diagnostic pitfalls. Diagn Cytopathol 2007;35:85–90.

Muller S, Barnes L. Basal cell adenocarcinoma of the salivary glands : report of seven cases and review of the literature. Cancer 1996;78:2471–2477.

Nagao T, Sugano I, Ishida Y, et al. Basal cell adenocarcinoma of the salivary glands: comparison with basal cell adenoma through assessment of cell proliferation, apoptosis, and expression of p53 and bcl-2. Cancer 1998;82:439–442.

8
Oncocytic Tumors: Oncocytoma, Warthin Tumor, and Acinic Cell Carcinoma

Background

Perhaps more than any other tissues in the body, the salivary glands exhibit a wide range of benign and neoplastic lesions that can have oncocytic or oncocyte-like features. True oncocytes have abundant densely granular eosinophilic cytoplasm due to the presence of numerous mitochrondria. In addition, they have central enlarged round nuclei with a distinct nucleolus. Sometimes the oncocytic features are a primary characteristic of the lesion such as in oncocytoma, and sometimes they are secondary to a process such as oncocytosis, a metaplastic change in the salivary glands of older adults. In the diffuse form of oncocytosis a majority of the salivary gland parenchyma, including ductal and acinar cells, is replaced by oncocytes. This process can even affect the neoplastic cells of many different salivary gland tumors. Radiologically, oncocytes concentrate technetium (99mTc), and therefore, tumors comprised of oncocytes are "hot" by radionuclide scanning. In this chapter, we will discuss the differential diagnosis of salivary gland lesions with true oncocytic features, as well as those that have eosinophilic features that can mimic those of an oncocyte.

Features of Oncocytic Lesions

- Cytoplasmic mitochondria
- May form a primary tumor (e.g., oncocytoma)
- May be a metaplastic process (e.g., oncocytosis)
- Can affect salivary gland neoplasms (e.g., pleomorphic adenoma)
- Detected by radionuclide scanning

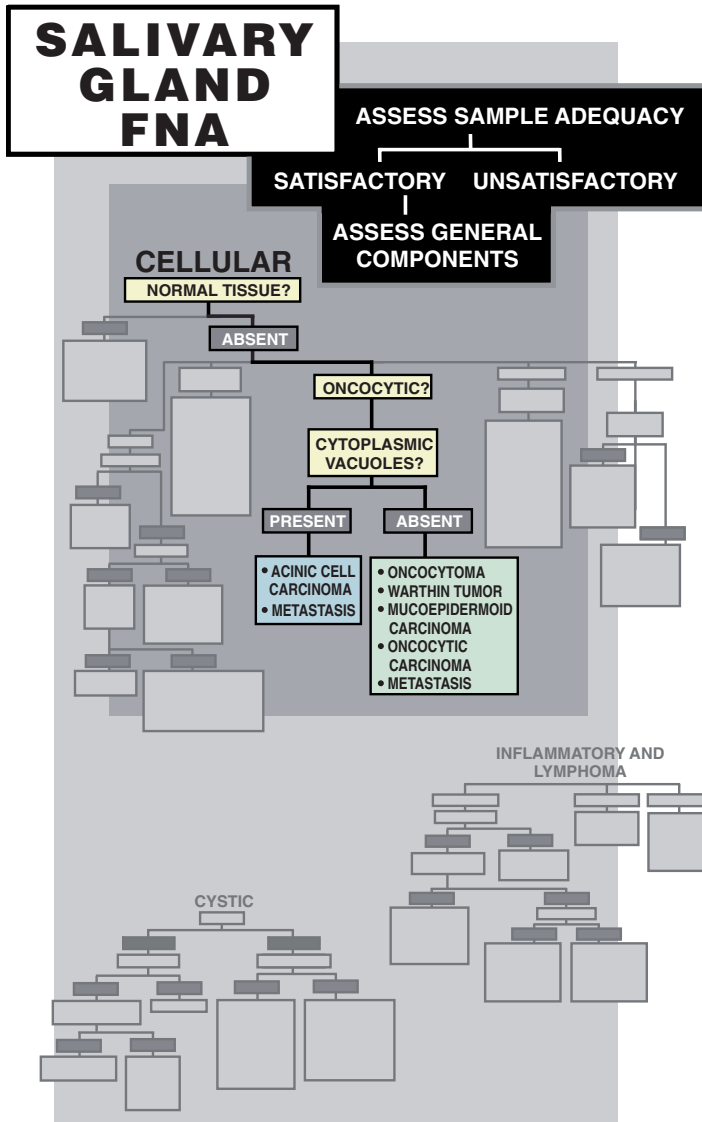


FIG. 8.1. Algorithm for oncocytic salivary gland lesions.

One of the most common diagnostic challenges among salivary gland tumors with an oncocytic microscopic appearance is differentiating the benign tumor, oncocytoma, from the low-grade carcinoma, acinic cell carcinoma. In addition, other tumors or subtypes of tumors with oncocytic features include Warthin tumor, the oncocytic variant of mucoepidermoid carcinoma, pleomorphic adenoma with oncocytic features, oncocytic carcinoma, and metastatic tumors, especially renal cell carcinoma. With the exception of pleomorphic adenoma, the most common salivary gland tumors with oncocytic features lack myoepithelial differentiation.

Salivary gland proliferations of oncocytes include nodular or diffuse hyperplasia (oncocytosis), oncocytoma, and oncocytic carcinoma. Oncocytomas are rare benign salivary gland tumors, representing less than 1% of all salivary gland neoplasms. The majority occur in the parotid gland in older adults with a mean age in the sixth to seventh decade and an equal gender predilection. A small subset of oncocytomas occurs in the submandibular and minor salivary glands. Most present as a slowly enlarging, 3–4 cm, painless mass. Approximately 5% can be multifocal, and even by histologic evaluation, it may very difficult to reliably distinguish a true oncocytoma from a hyperplastic oncocytic nodule in the setting of oncocytosis. On gross examination, oncocytomas are well circumscribed with a characteristic brown-red coloration; microscopically they are encapsulated, while the hyperplastic nodules in oncocytosis are not. Oncocytic carcinomas are clinically expansile, invasive tumors.

Warthin tumor (aka Warthin's tumor) is an interesting neoplasm with a unique pathologic appearance. It is also known by the more complex term papillary cystadenoma lymphomatosum, a name which accurately reflects its histologic appearance. Warthin tumor was first described in 1929 by Aldred Warthin, and it accounts for 5%–15% of all salivary gland tumors. In several large series, it is the second most common salivary gland tumor. Warthin tumor occurs almost exclusively within the parotid gland or rarely within adjacent lymph nodes; 5%–20% of cases can be bilateral or multifocal. While previously cited as more common in men, the ratio of Warthin tumor in women in recent studies is now similar to that in men. It is also more common in caucasians than in other racial groups. Epidemiologically,

Warthin tumor has been associated with cigarette smoking, with an 8-fold increased risk among smokers. There are several hypotheses for the pathogenesis of Warthin tumors; the proliferation of ductal elements developmentally entrapped within parotid-associated lymph nodes seems to be the most logical. This also helps to explain the nearly uniform occurrence within the parotid gland. Clonal and molecular studies indicate that Warthin tumors are not clonal proliferations, and thus are probably developmental lesions rather than true neoplasms, but this remains controversial. Patients, typically in their 5th to 7th decade, present with an enlarging painless mass, of usually less than 4 cm, in the superficial lobe of the parotid gland near the angle of the jaw. Warthin tumors are rare before the age of 40. On clinical examination, palpation of Warthin tumors has a very characteristic "doughy" feel, and aspirates yield a thick green-brown turbid fluid. In very rare cases, synchronous cancers such as squamous cell carcinoma or mucoepidermoid carcinoma, as well as malignant lymphoma can develop within a Warthin tumor. FNA is highly accurate for the diagnosis of Warthin tumors; however, as will be discussed, extensive squamous or mucinous metaplastic changes can present significant diagnostic challenges.

Clinicopathologic Features of Warthin Tumor

- 5%–15% of all salivary gland tumors
- Older adults (5th to 7th decade)
- Primarily within the parotid gland
- 5%–20% are bilateral or multifocal
- 8-fold increased risk among smokers
- Doughy feel on palpation
- FNA is highly accurate except in cases with extensive metaplasia
- Probably developmental, not neoplastic

In many series, acinic cell carcinoma is the second or third most common salivary gland malignancy, representing approximately 6% of all salivary gland tumors and up to 17% of salivary gland malignancies. The exact proportion of acinic cell carcinomas relative to other salivary gland tumors varies, in part due to the classification of some cases as "adenocarcinoma, NOS." Acinic cell carcinoma was first described by Nasse in 1892; it is defined histologically by the presence of at least focal serous acinar differentiation characterized

by PAS+diastase-resistant cytoplasmic zymogen granules. It is generally a low-grade tumor, although high-grade and dedifferentiated forms do occur. Attempts to histologically grade acinic cell carcinomas have met with limited success, and clinical stage is usually the better predictor of clinical outcome. Acinic cell carcinoma characteristically presents as a solitary, well-circumscribed, mobile, slowly growing, 1–3 cm mass which occasionally is painful. The low-grade nature of acinic cell carcinoma is evidenced by the fact that it was previously known as acinic cell tumor, with some patients presenting with a 10 or more year history of a salivary gland mass. Acinic cell carcinoma is more common in women, and there is a broad age range (mean age is 44 years) among patients, from young children to elderly adults. Seventy-five to 90% of cases present in the parotid gland, and most of the remaining cases occur in the intraoral minor salivary glands. A small subset of acinic cell carcinomas is bilateral. Tumors can be solid or cystic, with four histologic types of acinic cell carcinoma recognized: solid, microcystic, papillary-cystic, and follicular. FNA is moderately accurate at detecting acinic cell carcinomas, with approximately 75% of cases being diagnosed as suspicious or malignant. The most difficult cases to detect by FNA are those with a predominance of intercalated duct cells and lacking significant serous acinar differentiation.

Clinicopathologic Features of Acinic Cell Carcinoma

- Second to third most common salivary gland malignancy
- 6% of all salivary gland tumors
- Wide age range, from children to elderly adults
- Primarily within the parotid gland
- At least focal serous acinar differentiation
- Usually low-grade clinical behavior
- Can be solid or cystic

General Diagnostic Approach

The oncocytic arm of the algorithm is characterized by cellular aspirates containing epithelial cells with moderate to abundant eosinophilic cytoplasm (Fig. 8.1). Most cases in the differential diagnosis lack a myoepithelial component. Identifying the presence

or absence of cytoplasmic vacuoles is a very useful feature since their presence favors the diagnosis of acinic cell carcinoma (or a metastasis). Diff-Quik preparations can be used to more readily identify cytoplasmic vacuolization. The distinction between the various differential diagnostic entities in the oncocytic arm of the algorithm is important, since it includes both benign and malignant tumors. A key step is in evaluating the cytoplasmic features of the cells carefully and applying ancillary tests as needed.

Diagnostic Criteria

Oncocytoma

Aspirates of oncocytomas are variably cellular and consist of 2- and 3-dimensional cohesive groups of uniform polygonal cells with moderate to abundant amounts of densely granular eosinophilic cytoplasm (Figs. 8.2–8.3). Occasionally, the cells are arranged in trabeculae, and some single cells can be seen. Intercellular borders are very well-defined. The cytoplasm appears granular in Papanicolaou-stained preparations, and is deep blue and waxy using Diff-Quik stains. Importantly, cytoplasmic vacuoles are absent. The nuclei are centrally placed, uniform, enlarged and round to oval with a small distinct nucleolus. In some cases, the nuclei are more round and pyknotic without a discernible nucleolus. Mitotic activity is absent. The background is clean, lacking debris and lymphocytes. As mentioned previously, oncocytoma and oncocytosis are indistinguishable by FNA and can sometimes be challenging to distinguish even in excisional biopsy specimens. Clinicoradiologic correlation is sometimes helpful, but the distinction does not usually affect the clinical management since both are benign lesions.

Cytologic Features of Oncocytomas

- Cohesive 2- and 3-dimensional groups of uniform polygonal cells
- Moderate to abundant densely granular eosinophilic cytoplasm
- Well-defined intercellular borders
- Absence of cytoplasmic vacuoles
- Enlarged round nucleus with distinct nucleolus
- Clean background without lymphocytes

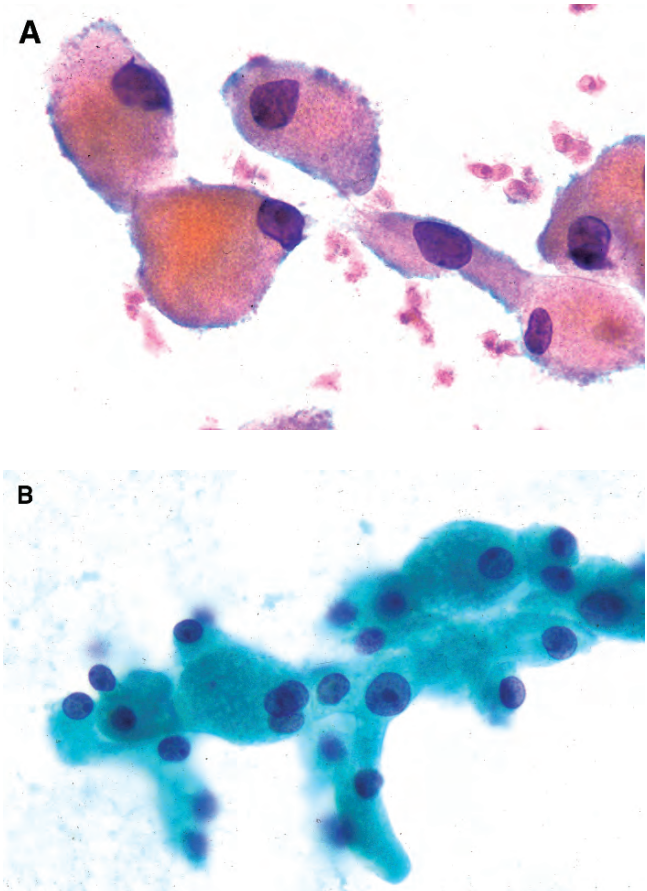


FIG. 8.2. Oncocytoma. (A and B) Cells have abundant evenly granular eosinophilic cytoplasm in a clean background. (Thin-layer preparation, Papanicolaou.)

Warthin Tumor

In addition to its characteristic "doughy" feel on palpation, and the granular opaque green-brown cyst fluid obtained by aspiration, the cytologic features of most Warthin tumors are easily recognized microscopically. There are 3 characteristic cytologic findings in aspirates of Warthin tumor: oncocytes, lymphocytes, and a "dirty"

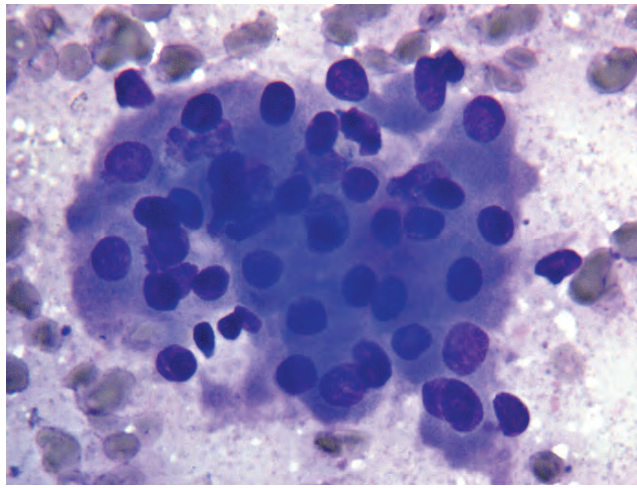


FIG. 8.3. Oncocytoma. The cells are uniform, with moderate amounts of waxy cytoplasm that lacks vacuoles. (Smear, Diff-Quik.)

granular proteinaceous background (Figs. 8.4–8.6). The oncocytes of Warthin tumor are similar to those described for oncocytoma. They are usually present in cohesive 2-dimensional sheets and have well-defined cell borders, giving a pavementing appearance. In some cases, the oncocytes can be arranged in a papillary formation. The cytoplasm is moderate to abundant, granular and eosinophilic owing to the many cytoplasmic mitochondria. The nuclei are enlarged, round to oval and centrally placed, usually with a distinct nucleolus. While the nuclei may appear atypical at first glance, they are very uniform, lacking significant nuclear pleomorphism; mitotic activity is absent. Background lymphocytes consist of a mixed pattern, usually with a predominance of small mature-appearing forms with an admixture of plasma cells, mast cells, tingible body macrophages, and occasional intermediate to larger lymphoid cells. Lymphohistiocytic aggregates representing aspirated fragments of germinal centers are sometimes also seen. The background in Warthin tumors is characteristically cystic, proteinaceous, and granular, sometimes with admixed necrotic cell debris. Occasionally, the background cystic material will contain

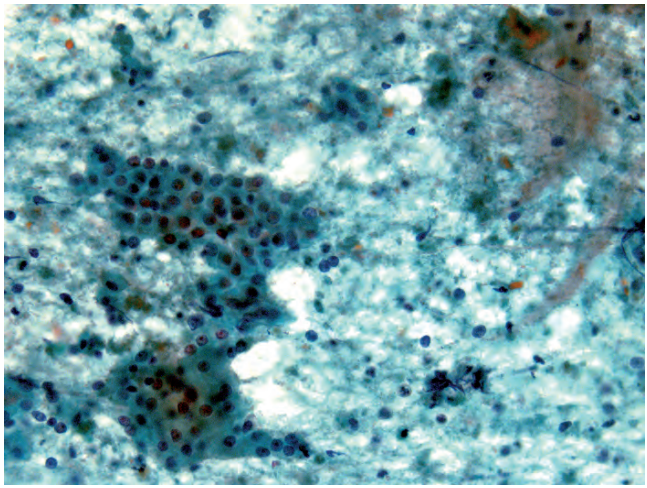


FIG. 8.4. Warthin tumor. The characteristic findings include cohesive flat sheets of oncocytes, lymphocytes, and a granular proteinaceous background. (Smear, Papanicolaou.)

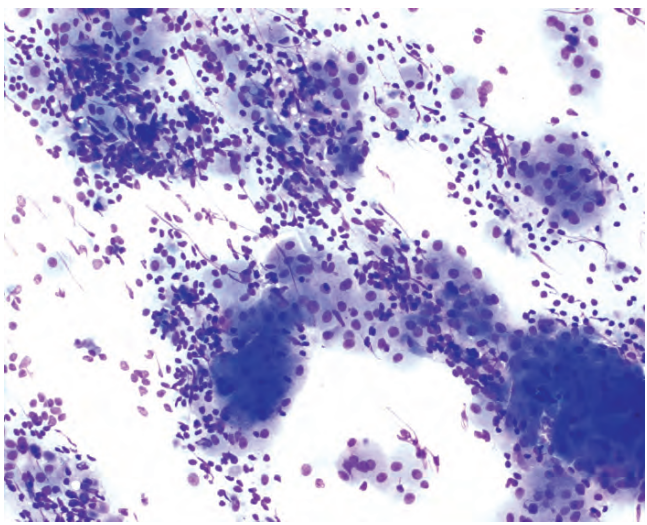


FIG. 8.5. Warthin tumor. The oncocytes have abundant densely granular to waxy cytoplasm in a background of a mixed population of lymhocytes. (Smear, Diff–Quik.)

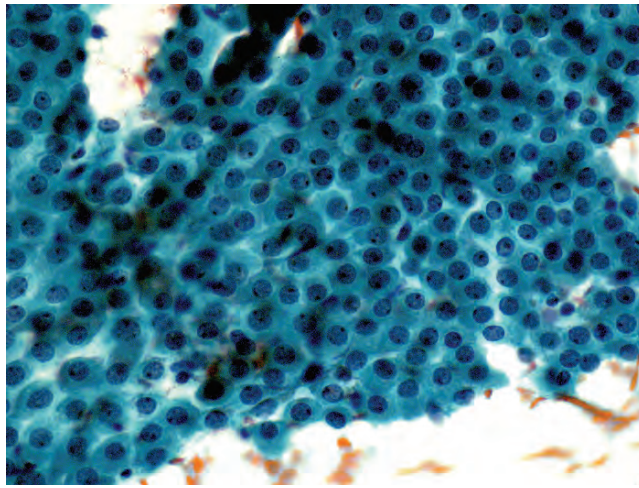


FIG. 8.6. Warthin tumor. The oncocytes form cohesive 2-dimensional groups with distinct cell borders. The cells are uniform, and have enlarged round nuclei with a distinct nucleolus. (Smear, Papanicoloau.)

abundant acute inflammation. It is important not to mistake the background of Warthin tumor for a malignant-associated tumor diathesis.

Cytologic Features of Warthin Tumor

- Cohesive 2- and 3-dimensional groups of oncocytes
- Scattered background lymphocytes
- Lymphohistiocytic aggregates
- Granular cystic background debris
- Squamous and mucinous metaplasia

While over 80% of Warthin tumors exhibit conventional cytologic features by FNA, a subset of Warthin tumors show variations that can lead to diagnostic problems. The predominant cell in most cases is the lymphocyte with occasional scattered groups of oncocytes. However, there is a wide range of cellular findings from cases that are predominantly lymphoid, resembling a lymph node aspirate, to those that have few lymphocytes and are mostly

oncocytes, resembling an oncocytoma. Such variations in cellularity and proportion of lymphocytes to oncocytes can become a diagnostic problem in poorly sampled cases. A careful search for both cell components and good clinicoradiologic correlation are important to avoid a diagnostic pitfall.

Squamous and mucinous metaplastic changes occur in over 30% of Warthin tumors, and in a subset of these, the changes can be extensive. Squamous metaplasia is the best-known change to occur in Warthin tumors (Figs. 8.7–8.8). It can vary from occasional squamoid cells to frank squamous differentiation with keratinization, parkeratotic cells, and even nuclear atypia. Warthin tumors with degenerating squamoid cells or groups of squamous cells within the background debris in the setting of oncocytes and lymphocytes are usually not a diagnostic problem. However, squamous atypia in the absence of the characteristic features of Warthin tumor may make it impossible to exclude a metastatic or primary squamous cell carcinoma. Rare cases of carcinoma arising within Warthin

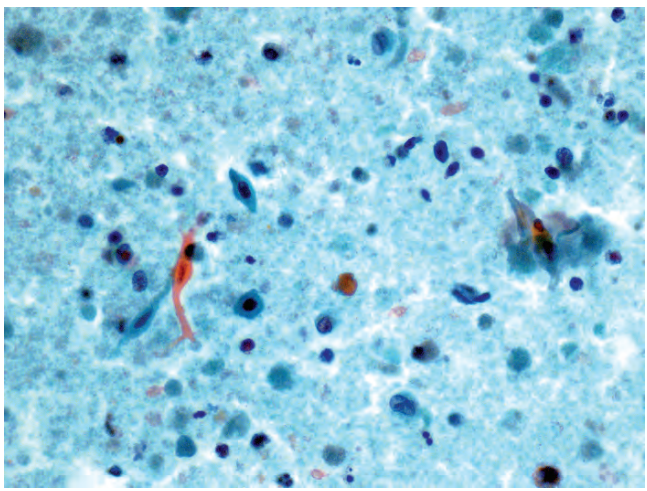


FIG. 8.7. Warthin tumor with squamous metaplastic changes. (Smear, Papanicolaou.) Degenerate squamoid cells are often seen within the background cystic debris of Warthin tumor.

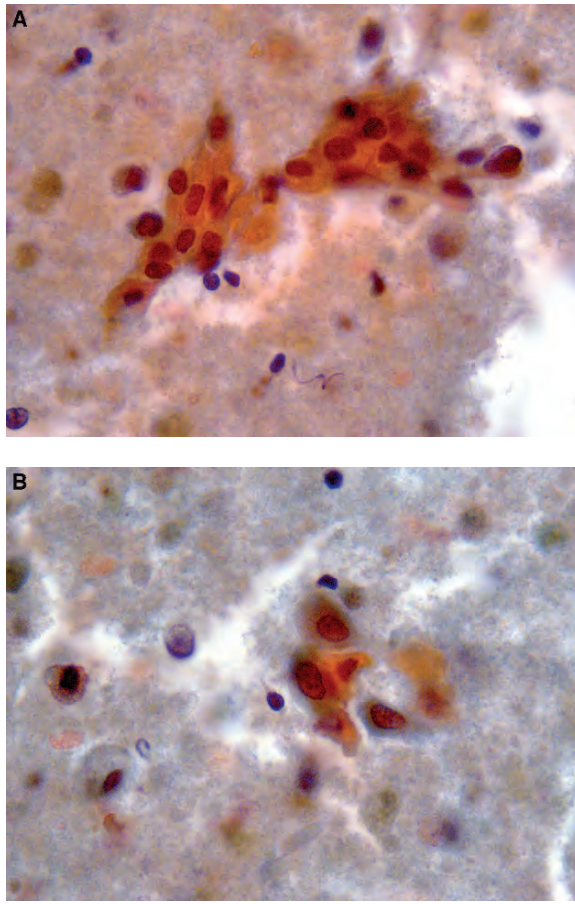


FIG. 8.8. Warthin tumor with squamous metaplastic changes. (A and B) The presence of groups of mildly atypical squamous cells can create a diagnostic problem. (Smear, Papanicolaou.)

tumor have also been reported. Mucinous metaplastic changes also consist of a spectrum from thick background mucoid material to extensive mucin-containing epithelial cells, the latter being a potential mimic of low-grade mucoepidermoid carcinoma (Fig. 8.9). As with squamous metaplasia, mucinous changes, particularly when

presenting as abundant thick background mucin in the context of other conventional features of Warthin tumor, are acceptable. However, when groups of cells with intracellular mucin are present, caution is warranted to avoid a misdiagnosis. Most of these cases will be diagnosed as "atypical," and surgical excision will be necessary to exclude mucoepidermoid carcinoma.

Squamous and mucinous metaplastic changes occur in over 30% of Warthin tumors and can be extensive.

Variants of Warthin Tumor That Can Lead to Diagnostic Difficulties

- Lymphocyte-predominant
- Oncocyte-predominant
- Squamous metaplasia
- Mucinous metaplasia

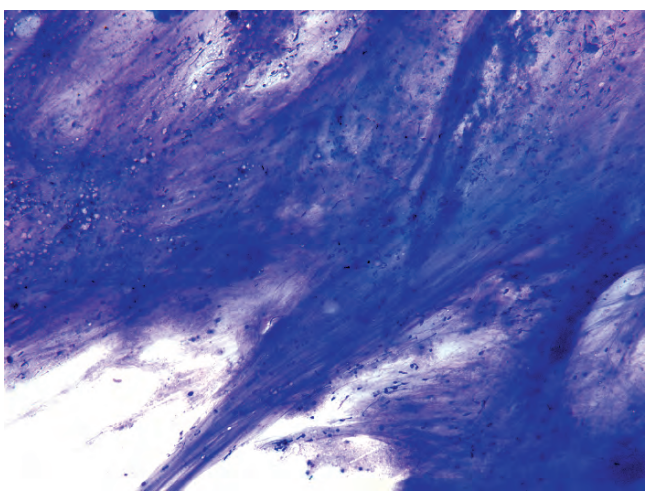


FIG. 8.9. Warthin tumor with mucinous metaplasia. Abundant thick background mucoid material is seen in a subset of Warthin tumors. (Smear, Papanicolaou.)

Acinic Cell Carcinoma

Aspirates of acinic cell carcinoma can contain any of a variety of cell types, including serous acinar, intercalated duct, vacuolated, clear, and nonspecific glandular. The hallmark cytologic and histologic feature, however, is at least focal serous acinar differentiation with its characteristic cytoplasmic zymogen granules. Fortunately, the serous acinar cell is the most common cell type encountered in aspirates of acinic cell carcinoma. In addition to various cell types, the histologic appearance of acinic cell carcinoma can include any of 4 different architectural patterns: solid, microcystic, papillary cystic, and follicular. The solid and microcystic patterns are the most common, and are seen in over 70% of acinic cell carcinomas. In contrast, the follicular pattern is the rarest, present in less than 5% of cases.

At least focal serous acinar differentitation is the hallmark of acinic cell carcinoma.

- Cell types seen in acinic cell carcinomas
  - Serous acinar
  - Intercalated duct
  - Vacuolated
  - Clear
  - Non-specific glandular
- Architectural patterns seen in acinic cell carcinomas
  - Solid
  - Microcystic
  - Papillary cystic
  - Follicular

The classic aspirate of acinic cell carcinoma is cellular and comprised of large polygonal cells with abundant delicate, vacuolated cytoplasm (Fig. 8.10). The cells are haphazardly arranged in 3-dimensional groups, sheets, and as single cells. In some cases, a capillary meshwork or even papillary formations around a fibrovascular core can be seen. The nuclei are cytologically bland, round to oval,

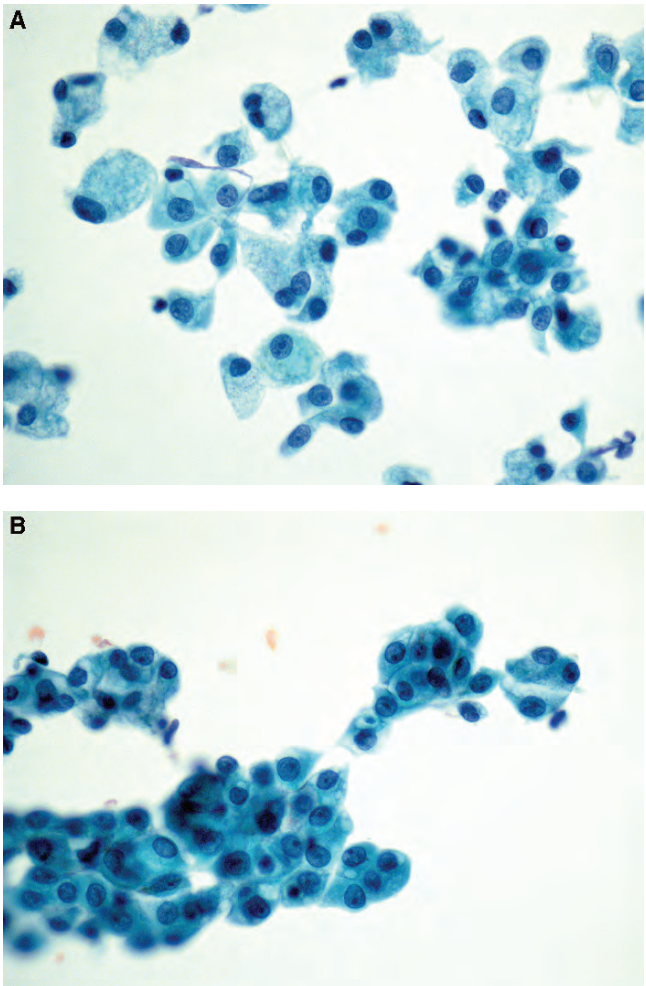


FIG. 8.10. Acinic cell carcinoma. Large polygonal cells with abundant delicate cytoplasm (A) solid subtype; (B) papillary cystic subtype. (Thin-layer preparation, Papanicolaou.)

uniform, and eccentrically placed with small distinct nucleoli. Mitoses are absent to rare. The cytoplasm is the key to the cytologic diagnosis of acinic cell carcinoma. It is abundant, vacuolated, and slightly basophilic to eosinophilic. While the vacuolated nature of

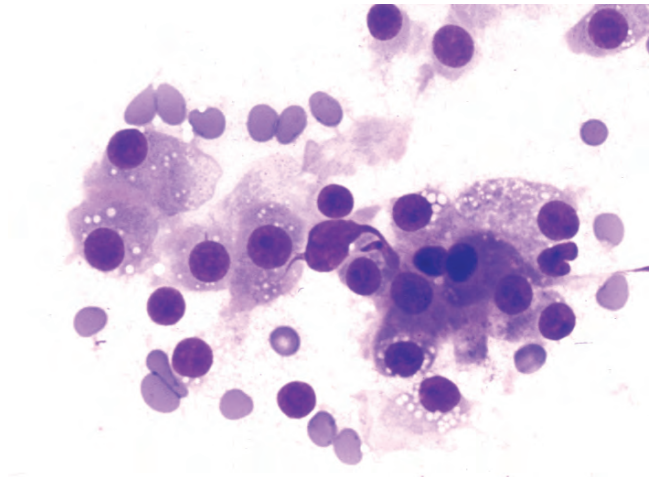


FIG. 8.11. Acinic cell carcinoma. Small cytoplasmic vacuoles are most easily seen in Diff-Quik stained preparations. (Smear, Diff-Quik.)

the cytoplasm can be appreciated using Papanicolau stains, it can be subtle, and the vacuoles are much more visible using Romanowsky stains (Fig. 8.11). Occasional small dark-staining cytoplasmic zymogen granules can often also be seen; while they are sometimes abundant, they are more often sparse and difficult to find (Fig. 8.12). For cases where a cell block is available, staining with PAS + diastase can be used to help demonstrate the presence of cytoplasmic zymogen granules. The background in aspirates of acinic cell carcinoma is clean, but may contain scattered stripped nuclei reflecting the delicate nature of the cytoplasm. Aspirates of the papillary cystic variant will yield a cystic background, and a 10% subset of acinic cell carcinomas will have a mixed lymphoid background, including lymphohistiocytic aggregates. Psammoma bodies can occasionally be found, especially in the papillary cystic variant (Fig. 8.13). Acinic cell carcinomas are also particularly prone to cystic degeneration with associated hemosiderin-laden macrophages. A rare subset of acinic cell carcinomas is dedifferentiated and exhibits high-grade nuclear features that are generally not recognizable by FNA as acinic cell carcinomas.

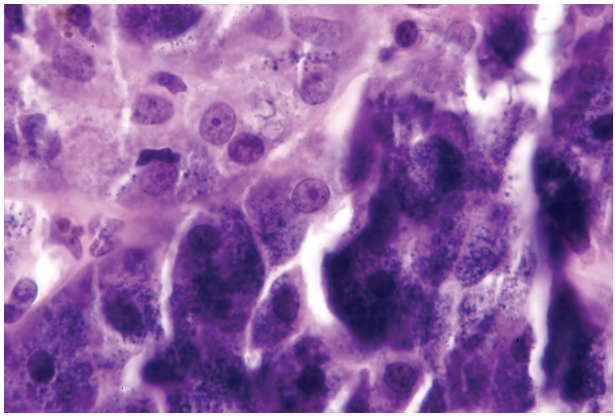


FIG. 8.12. Acinic cell carcinoma. Course basophilic cytoplasmic zymogen granules are the hallmark of serous acinar differentiation. (Cell block, H&E.)

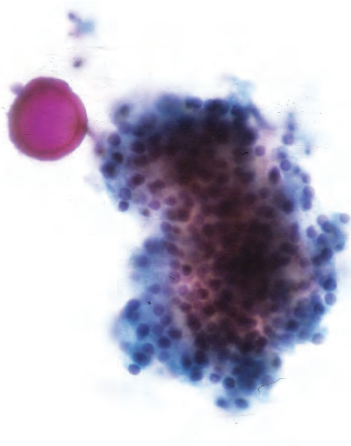


FIG. 8.13. Acinic cell carcinoma. Psammoma bodies are sometimes seen in aspirates of acinic cell carcinoma, especially the papillary cystic subtype. (Smear, Papanicolaou.)

The cytoplasmic vacuoles and zymogen granules are the key to the cytologic diagnosis of acinic cell carcinoma.

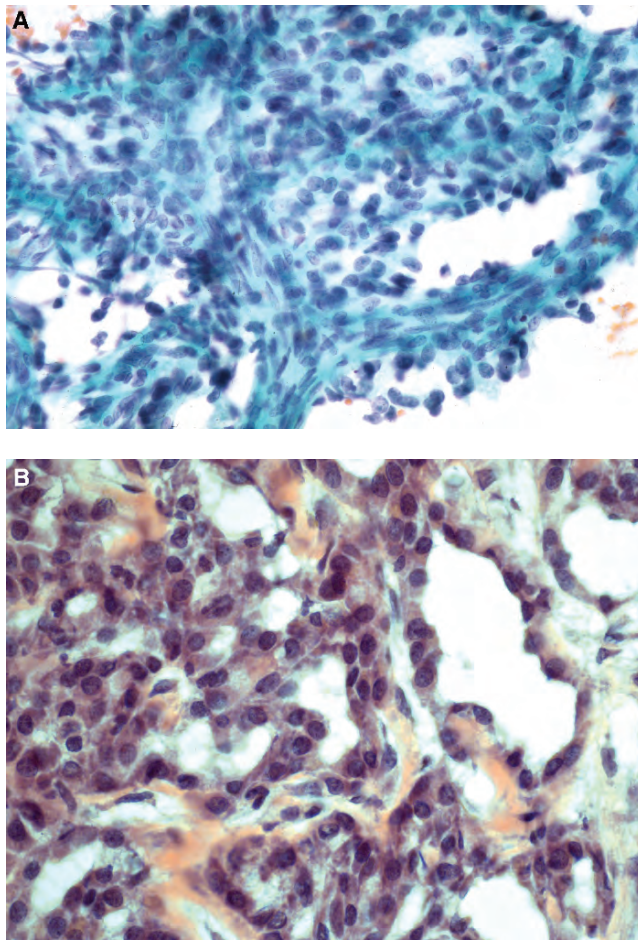


FIG. 8.14. Acinic cell carcinoma. (A and B) A small subset of cases contain a predominance of intercalated duct cells which are cuboidal and lack cytoplasmic zymogen granules. (A, Smear, Papanicolaou; B, Cell block, H&E.)

Cytologic Features of Conventional Acinic Cell Carcinoma

- Large polygonal cells
- Uniform, round eccentric nuclei
- Delicate vacuolated cytoplasm
- PAS-positive, diastase-resistant zymogen granules
- Background stripped nuclei
- Lymphocytes may be present
- Rare psammoma bodies

When acinic cell carcinomas show well-defined features of serous acinar differentiation, a definitive cytologic diagnosis is usually possible. However, a small subset of cases will contain a predominance of cells lacking serous acinar differentiation that can include intercalated duct, vacuolated, clear, and nonspecific glandular types. Of these, the intercalated duct and nonspecific glandular cells pose the greatest diagnostic challenge. While a careful search for focal serous acinar differentiation may be helpful, cases with a predominance of these cell types will usually receive a descriptive diagnosis such as "low-grade glandular neoplasm" with a note and differential diagnosis. Histologically, most cases are recognized as acinic cell carcinoma, but some of these may be classified as adenocarcainoma, NOS. In contrast to serous acinar cells, intercalated duct cells are smaller, cuboidal, have a higher N:C ratio, have centrally placed nuclei, and form cohesive groups (Fig. 8.14). The cytoplasm is more dense and eosinophilic than in serous acinar cells, and it lacks well-defined small vacuoles and zymogen granules. Aspirates of nonspecific glandular cells are similar to intercalated duct cells but more round to polygonal in shape.

Cases with a predominance of intercalated duct cells will usually receive a descriptive diagnosis such as "low-grade glandular neoplasm" with a note and differential diagnosis.

Differential Diagnosis and Pitfalls

The differential diagnosis of salivary gland tumors with oncocytic features includes oncocytoma and oncocytosis, Warthin tumor, acinic cell carcinoma, the oncocytic variant of mucoepidermoid carcinoma, oncocytic carcinoma, and metastatic renal cell carcinoma. Other very rare cystic lesions that can sometimes be considered in the differential diagnosis of oncocytic tumors are cystadenoma, cystadenocarcinoma, and sclerosing polycystic adenosis, which will be discussed in Chapter 9. Among the oncocytic salivary gland tumors, the most common recurring problem in our experience is the distinction between oncocytoma and acinic cell carcinoma (Table 8.1). At low magnification, these two entities appear similar, but at higher magnifaction and using Diff-Quik stains, acinic cells have delicate cytoplasm with small vacuoles, while oncocytomas have densely granular to waxy-appearing cytoplasm without vacuoles. For difficult cases, ancillary studies can be performed using cell block material. While they may be sparse, zymogen granules are present in the cytoplasm of acinic cell carcinomas and can be demonstrated using PAS+ diastase. In addition, phosphotungstic acid-hematoxylin (PTAH), a stain for mitochondria, shows strong positive cytoplasmic staining in oncocytomas, while acinic cell carcinomas are negative or only weakly positive.

The most common recurring problem in our experience is the distinction between oncocytoma and acinic cell carcinoma.

Differential Diagnosis of Oncocytic Salivary Gland Lesions

- Oncocytoma and oncocytosis
- Warthin tumor
- Acinic cell carcinoma
- Oncocytic variant of mucoepidermoid carcinoma
- Oncocytic carcinoma
- Pleomorphic adenoma with oncocytic features
- Metastatic renal cell carcinoma

TABLE 8.1. Comparison of selected differential diagnostic entities.

Feature	Normal Salivary Gland	Oncocytoma	Warthin Tumor	Acinic Cell Carcinoma
Cellularity	Low to moderate	Moderate to high	Variable	Moderate to high
Cells	Acinar and few ductal	Oncocytes	Oncocytes and lymphocytes	Acinar and intercalated duct
Background	Stripped nuclei	Clean	"Dirty" granular debris	Stripped nuclei
Cell Arrangement	Polarized in lobular groups	Sheets and 3-dimensional groups	Sheets and 3- dimensional groups	Haphazard in 2- and 3-dimensional groups
Cell borders	Indistinct	Well defined	Well defined	Indistinct
Atypia	Absent	Absent	Atypical metaplasia	Variable
Cytoplasm	Abundant and vacuolated	Moderate and densely granular	Moderate and densely granular	Abundant and vacuolated
PTAH Stain	Negative	Positive	Positive	Negative
PAS + Diastase	Positive	Negative	Negative	Positive
Electron Microscopy	Zymogen granules	Mitochondria	Mitochondria	Zymogen granules

As mentioned in Chapter 4, an important pitfall to avoid is confusing aspirates of normal acinar cells with those of acinic cell carcinoma. Aspirates of both normal salivary gland tissue and acinic cell carcinoma can look similar when they contain many single cells and stripped acinar cell nuclei within the background. Since individual cells of acinic cell carcinoma can be virtually indistinguishable from normal acinar cells, it is important to search instead for groups of cells. Acinic cell carcinomas contain cells that are crowded together haphazardly, lacking the polarity and ductal elements of normal salivary gland tissue.

As described previously in this chapter, the most common pitfall in the diagnosis of Warthin tumor is related to squamous and mucinous metaplastic changes, which can be extensive. Thorough sampling of the lesion and good clinicoradiologic correlation can be very helpful in avoiding a false positive diagnosis. When the metaplastic changes are present in a background of otherwise normal Warthin tumor components, a correct, definitive diagnosis of Warthin tumor can be made. However, when only metaplastic elements are found, it will not be possible to exclude carcinoma.

The rare oncocytic variant of mucoepidermoid carcinoma (see Chapter 9) is a potential pitfall in the evaluation of oncocytic salivary gland lesions (Fig. 8.15). Most cases are low-grade tumors with minimal nuclear atypia, but an infiltrative growth pattern. Aspirates are cellular and composed predominantly of bland oncocytic cells with scattered mucinous goblet cells and only rare groups of epidermoid and intermediate cells. The background is often partially cystic and mucoid. The oncocytic cytoplasm of the neoplastic cells is positive with PTAH due to the abundant cytoplasmic mitochondria. The key to avoiding misinterpretion of this carcinoma as an oncocytoma or Warthin tumor is to search carefully for the characteristic mucinous goblet cells that are usually a significant component of this tumor combined with the rare epidermoid and intermediate cells. If cell block material is available, stains to confirm intracellular mucin can be performed.

Oncocytic carcinoma is another very rare carcinoma that can pose a potential pitfall in the cytologic diagnosis of oncocytic salivary gland lesions (Fig. 8.16). This is an aggressive salivary gland carcinoma that occurs in older adults (mean age = 62.5 years), and in some cases develops from a pre-existing oncocytoma. Aspirates

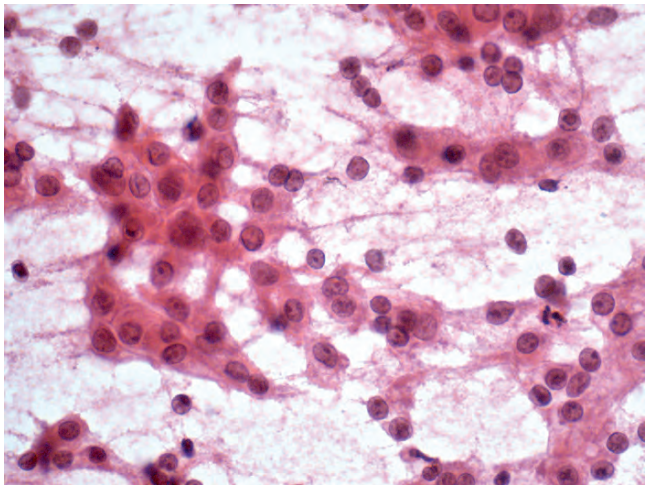


FIG. 8.15. Oncocytic variant of mucoepidermoid carcinoma. A combination of epidermoid and polygonal cells with dense granular oncocytic cytoplasm. (Smear, Papanicolaou.)

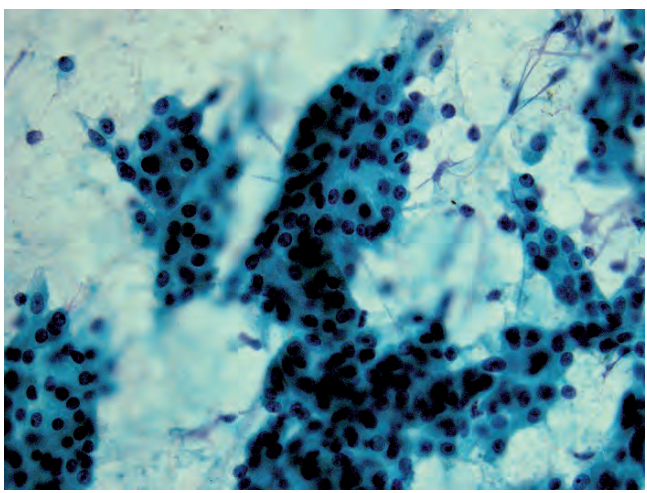


FIG. 8.16. Oncocytic carcinoma. Cells have abundant densely granular cytoplasm, and large nuclei with prominent nucleoli. (Smear, Papanicolaou.)

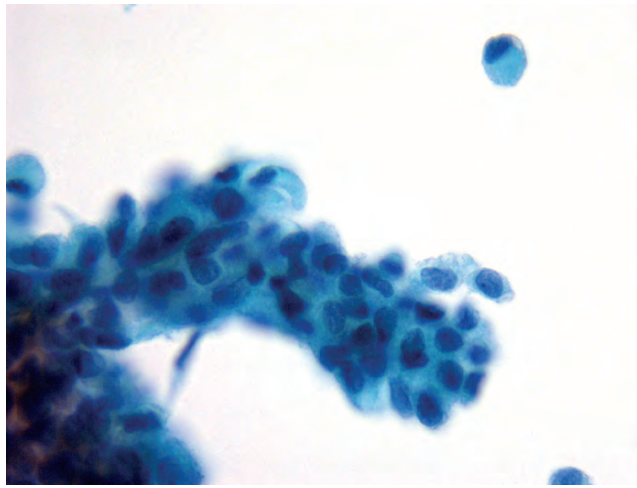


FIG. 8.17. Pleomorphic adenoma with oncocytic features. (Smear, Papanicoloau.)

of oncocytic carcinoma can easily be misinterpreted as an oncocytoma if careful attention is not given to the atypical variation in cell size and shape, and the moderate degree of nuclear pleomorphism. In addition, mitotic activity is often present. and sometimes background necrosis will be found. Tumors are infiltrative and often large - features that are usually appreciated both clinically and radiologically. Therefore, good clinicoradiologic correlation combined with careful microscopic analysis can help to avoid a potential false negative diagnosis.

Oncocytosis is a metaplastic process that occurs in some older adults. It can result in the formation of variably sized nodules of oncocytes that are nearly indistinguishable from true oncocytomas except for their lack of a true fibrous capsule. Oncocytosis can affect various salivary gland tumors; most commonly this has been reported in pleomorphic adenomas (Fig. 8.17). Aspirates of pleomorphic adenoma with oncocytic features will contain many oncocytes, singly and in groups, but thorough sampling will also usually yield characteristic fragments of metachromatic fibrillar matrix material (see Chapter 5). In addition,

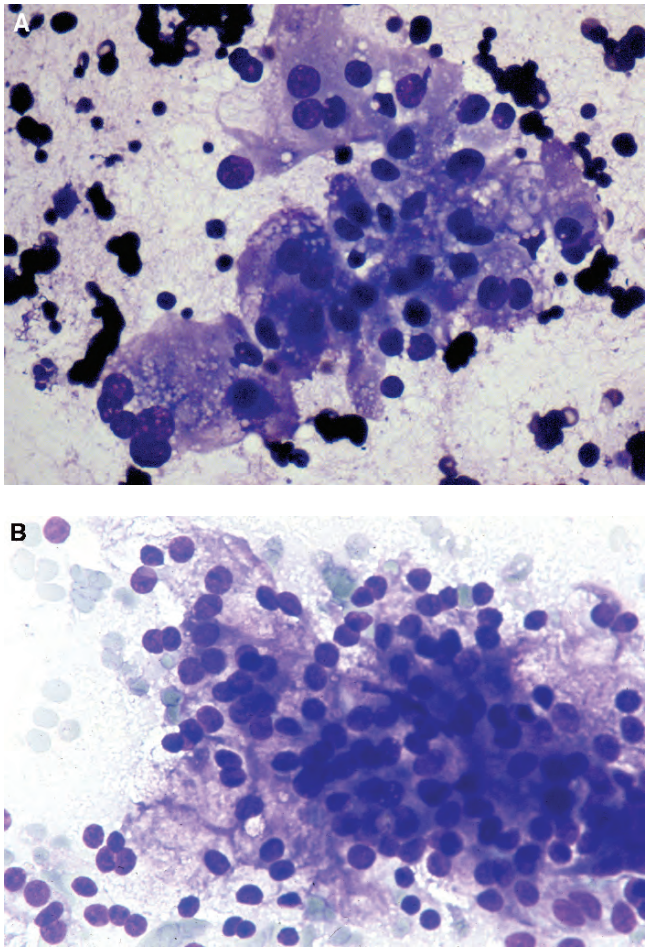


FIG. 8.18. Metastatic renal cell carcinoma (A) can appear very similar to acinic cell carcinoma (B) in salivary gland aspirates. (Smears, Diff–Quik.)

ancillary marker studies such as smooth muscle actin, calponin, and S-100 can be used to demonstrate myoepithelial differentiation, which is not seen in other oncocytic tumors discussed in this chapter.

Metastatic renal cell carcinoma is among the most common distant tumors metastasizing to the major salivary glands, where it can mimic a primary tumor, especially acinic cell carcinoma (Fig. 8.18). Both tumors are characterized by similar abundant delicate eosinophilic cytoplasm and bland nuclear features. The presence of small basophilic cytoplasmic granules favors acinic cell carcinoma. A majority, but not all, patients with metastatic renal cell carcinoma will have a clinical history to alert the cytopathologist to the potential diagnostic pitfall. Applying ancillary immunohistochemical studies using cell block material is the most reliable method for distinguishing renal cell carcinoma and acinic cell carcinoma. In contrast to acinic cell carcinoma, renal cell carcinomas are positive for CD10, renal cell carcinoma marker, and EMA, and negative for cytokeratin 7.

Ancillary Techniques

In assessing an oncocytic lesion of the salivary gland, Diff-Quik stains for identification of cytoplasmic vacuoles should be used in conjunction with standard Papanicolaou stain preparations. In addition, special studies to better define characteristics of the oncocytic cytoplasm can be applied to cell block material. PTAH stains are used for demonstrating cytoplasmic mitochondria, PAS+ diastase will stain zymogen granules, and mucicarmine can be used to demonstrate intra- and extracellular mucin. Although not always an available option, material can also be placed into glutaraldehyde fixative for evaluation by electron microscopy, which is a very sensitive method for distinguishing true oncocytes with abundant cytoplasmic mitochondria from acinar cells with secretory vacuoles and zymogen granules. Immunohistochemical stains are of limited value in the distinction of oncocytic salivary gland tumors. Anti-mitochondrial antibody can be used to help distinguish oncocytoma and acinic cell carcinoma, and a subset of acinic cell carcinomas is positive for amylase. With the exception of pleomorphic adenoma with oncocytic changes, most oncocytic salivary gland tumors, including oncocytoma, Warthin tumor, acinic cell carcinoma, oncocytic mucoepidermoid carcinoma, and oncocytic carcinoma. are immunohistochemically negative for myoepithelial markers (e.g., calponin, smooth muscle actin, S-100).

Most oncocytic salivary gland tumors are immunohistochemically negative for myoepithelial markers.

Clinical Management and Prognosis

The treatment for oncocytoma and Warthin tumor is complete surgical excision, which for parotid-based tumors will most often entail superficial parotidectomy with facial nerve preservation. Local recurrence of either tumor is uncommon. Less than 6% of Warthin tumors recur, and the majority of these are thought to be due to multifocal tumors. Rarely, malignancy can develop within a Warthin tumor, and may originate from either the epithelial (e.g., squamous cell carcinoma, mucoepidermoid carcinoma, oncocytic carcinoma) or the lymphoid component (e.g., small lymphocytic lymphoma, follicular lymphoma, MALT lymphoma). Malignant transformation of oncocytoma to oncocytic carcinoma is also very rare.

Acinic cell carcinomas are treated by complete surgical excision with disease-free resection margins. Postsurgical radiation therapy is usually reserved for cases with positive resection margins or with high-grade or undifferentiated histologic features where the overall prognosis is very poor. The average rate of recurrent acinic cell carcinoma is 33%–44%, and the risk of metastatic disease is 16%–19%. Clinical stage is considered the best predictor of outcome. The least aggressive clinical behaviors are reported for tumors of the minor salivary glands; tumors in the submandibular gland have been reported to have a worse clinical outcome than those arising in the parotid. An unusual feature of acinic cell carcinoma is that local recurrence or metastatic disease can occur decades after the initial diagnosis and resection; therefore, patients treated for acinic cell carcinoma require longterm follow-up.

Suggested Reading

Ali SZ. Acinic-cell carcinoma, papillary-cystic variant: a diagnostic dilemma in salivary gland aspiration. Diagn Cytopathol 2002;27:244–250.

Arida M, Barnes EL, Hunt JL. Molecular assessment of allelic loss in Warthin tumors. Mod Pathol 2005;18:964–968.

Ballo MS, Shin HJC, Sneige N. Sources of diagnostic error in the fine-needle aspiration diagnosis of Warthin tumor and clues to a correct diagnosis. Diagn Cytopathol 1997;17:230–234.

Brandwein MS, Huvos AG. Oncocytic tumors of major salivary glands. A study of 68 cases with follow-up of 44 patients. Am J Surg Pathol 1991;15:514–528.

Chang A, Harawi Sj. Oncocytes, oncocytosis, and oncocytic tumors. Pathol Annu 1992;27:263–304.

Flezar M, Pogacnik A. Warthin's tumor: unusual vs. common morphological findings in fine needle aspiration biopsies. Cytopathol 2002;13:232–241.

Gonzalez-Peramato P, Jimenez-Heffernan JA, Lopez-Ferrer P, Vicandi B, Viguer JM. Fine needle aspiration cytology of dedifferentiated acinic cell carcinoma of the parotid gland: a case report. Acta Cytol 2006;50:105–108.

Jahan-Parwar B, Huberman RM, Donovan DT, Schwartz MR, Ostrowski ML. Oncocytic mucoepidermoid carcinoma of the salivary glands. Am J Surg Pathol 1999;23:523–529.

Klijanienko J, Vielh P. Fine-needle sampling of salivary gland lesions V: Cytology of 22 cases of acinic cell carcinoma with histologic correlation. Diagn Cytopathol 1997;17:347–352.

Parwani AV, Ali SZ. Diagnostic accuracy and pitfalls in fine-needle aspiration interpretation of Warthin tumor. Cancer Cytopathol 2003;99:166–171.

Williamson JD, Simmons BH, El-Naggar A, Medeiros LJ. Mucoepidermoid carcinoma involving Warthin tumor: A report of five cases and review of the literature. Am J Clin Pathol 2000;114:564–570.

9
Cystic and Mucinous Lesions: Mucocele and Low-Grade Mucoepidermoid Carcinoma

Background

Cystic lesions are estimated to account for up to 8% of all salivary gland masses. They represent a wide range of salivary gland pathology from the non-neoplastic mucocele, salivary duct cyst, and sclerosing polycystic adenosis, to benign tumors such as cystadenoma and Warthin tumor, to malignant cystic tumors, including low-grade mucoepidermoid carcinoma and cystadenocarcinoma. In some cases, cystic degeneration occurs in what are ordinarily noncystic tumors such as pleomorphic adenoma and basal cell adenoma, and some tumors have cystic variants such as the papillary cystic subtype of acinic cell carcinoma. In the parotid gland, cystic lesions also include the difficult differential diagnosis of lymphoepithelial cysts, and metastatic cystic squamous cell carcinoma to an intraparotid lymph node. Of all of these entities, the most commonly encountered differential diagnostic problem is the cytologic distinction between mucocele and low-grade mucoepidermoid carcinoma. This chapter will focus foremost upon this common diagnostic problem, followed by an examination of other differential diagnostic entities.

Because aspirates of cysts typically produce hypocellular specimens consisting primarily of cyst contents, FNA has a poor track record for diagnosing cystic malignancies in any anatomic site, including the salivary glands. The general overall diagnostic accuracy of FNA for the evaluation of cystic lesions is estimated to be approximately 40%, mostly due to sample adequacy issues. This is true in the salivary

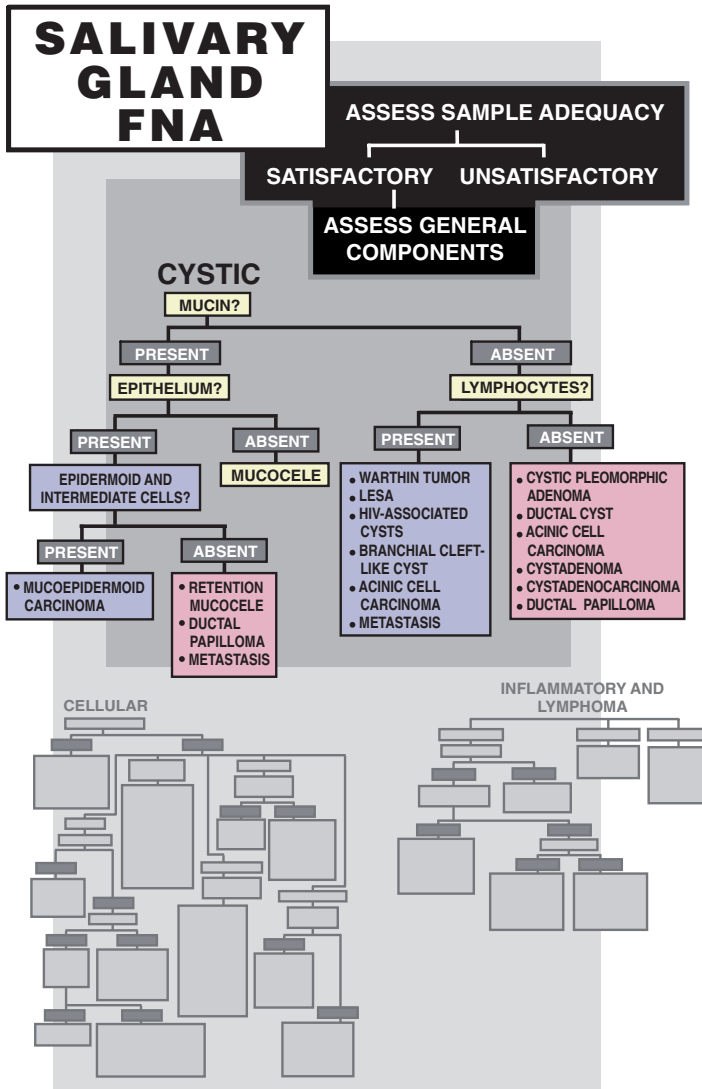


FIG. 9.1. Algorithm for cystic and mucinous salivary gland lesions.

gland as well, where one of the most common causes of a false negative FNA diagnosis is low-grade mucoepidermoid carcinoma.

Mucoceles and mucus retention cysts are the most commonly acquired non-neoplastic salivary gland lesions. Of these, mucoceles are by far the more frequent, and occur secondary to transection or obstruction of the excretory salivary duct. Causes of obstruction can include salivary duct stones (sialoliths), trauma such as biting, and scarring secondary to sialadenitis, tumors, and parasites. Most mucoceles occur in the minor salivary glands of the oral cavity, particularly in the lower lip, where they are most common in the third decade. Typically they are less than 1.0 cm in size. Because mucoceles are uncommon in the parotid and submandibular gland, caution is particularly warranted when interpreting mucin-containing aspirates from these two sites. Mucoceles are actually pseudocysts since they lack a true epithelial cyst lining. They are characterized by accumulation and extravasation of mucin within a connective tissue space. Hence, they are also referred to as "extravasation mucoceles." A clinically aggressive subtype of mucocele is the ranula, which occurs in the floor of the mouth and involves the sublingual gland. In some cases, the mucus dissects into extra-oral soft tissues of the neck and is referred to as a plunging ranula. In contrast to extravasation mucoceles, retention mucoceles are true cysts with an epithelial lining that can be cuboidal, ductal, oncocytic, or squamous. They represent accumulation of mucin within a dilated ductal space and are more common in older adults. Salivary duct cysts are similar to a mucus retention cyst but are more common in the parotid gland, where they are usually larger and have a more watery content rather than primarily mucinous.

Mucoceles and mucus retention cysts are the most common acquired non-neoplastic salivary gland lesions.

Mucoepidermoid carcinoma represents 10%–15% of salivary gland tumors. It occurs over a broad age range and is the most common major and minor salivary gland malignancy in both children and adults. The majority occur in the parotid gland, but among the minor salivary glands, the buccal mucosa and palate are the most common sites of origin. In addition, a very rare intraosseous

form of mucoepidermoid carcinoma (central mucoepidermoid carcinoma) occurs within the mandible, and another rare subtype, sclerosing mucoepidermoid carcinoma with eosinophilia, occurs most often in the thyroid gland. Patients with low-grade mucoepidermoid carcinoma have a mean age in the fifth decade and typically present with a slowly enlarging, painless mass that has been present for several months to many years. Histologically, mucoepidermoid carcinomas are divided into low-, intermediate-, and high-grade. Features used in the histologic grading include the presence and extent of a cystic component, perineural invasion, necrosis, mitotic activity, and nuclear atypia. The cytology of these tumors is variable depending upon the grade of the tumor, but low-grade forms are the most frequently encountered (see Chapter 10 for a discussion of high-grade mucoepidermoid carcinoma). Low-grade mucoepidermoid carcinoma is also the most common cause of a false negative cytologic diagnosis, in part because aspirates may yield only cyst contents. In addition, the epithelial cells are cytologically bland, and the mucin-containing cells can easily be misinterpreted as histiocytes or muciphages. When aspirating a mucinous cyst, it is important to always aspirate any residual solid mass that remains after the initial FNA. Ultrasound-guided FNA is very useful for aspirating cystic lesions, including those in the salivary glands. Because mucoepidermoid carcinomas can be extremely challenging to diagnose by FNA, when possible, material for a cell block preparation should be obtained. The latter can be quite useful since ancillary marker stains for keratins and for intracellular mucin will help to solidify the diagnosis.

Mucoepidermoid carcinoma is the most common salivary gland malignancy in children and adults.

General Diagnostic Approach

The algorithm for cystic salivary gland lesions begins by considering the presence or absence of background mucin within the cystic aspirate (Fig. 9.1). For those cysts where a mucinous background is present, the differential diagnosis includes low grade mucoepidermoid carcinoma and mucocele. The key to identifying low

grade mucoepidermoid carcinoma is the identification of an epithelial component consisting of three cell types: epidermoid, intermediate, and mucus. In contrast, extravasation mucoceles lack an epithelial component and contain histiocytes and muciphages instead. When a mucinous cyst with epithelial cells does not contain epidermoid and intermediate forms, lesions besides low-grade mucoepidermoid carcinoma such as retention mucocele, ductal papilloma, and metastasis should be considered. For nonmucinous cysts, the presence or absence of background lymphocytes is a useful feature. Warthin tumor is the classic example of a salivary gland cyst with oncocytes and background lymphocytes (see Chapter 8). A wide variety of salivary gland cysts lacking background mucin and lymphocytes can be encountered, and distinguishing among them requires careful attention to the cell types and subtle cytoarchitectural patterns.

Diagnostic Criteria

General Features of Cystic Salivary Gland Lesions

As with other anatomic sites, aspirates of salivary gland cysts yield hypocellular specimens with cyst fluid that may be mucinous or proteinaceous and contain histiocytes, inflammatory cells, and debris. Especially for tumors that are cystic due to degenerative changes (e.g., pleomorphic adenoma and acinic cell carcinoma), histiocytes may be hemosiderin-laden, but are most often foamy and sometimes are muciphages. As mentioned above, a key to the evaluation of any cyst is the presence or absence of an epithelial component. In the absence of epithelial cells, caution is warranted since the aspirate may simply not be representative of the underlying lesion, and the possibility of a false negative diagnosis exists. This is particularly true for mucinous cysts of the parotid and submandibular glands.

Caution is warranted in the evaluation of mucinous cysts, especially in the parotid and submandibular glands.

Low-Grade Mucoepidermoid Carcinoma

Aspirates of low grade mucoepidermoid carcinoma are often hypocellular (Fig. 9.2) and characterized by a combination of three epithelial cell types: epidermoid (squamoid) cells, intermediate cells, and mucus cells in a background of thick purple to blue-staining mucoid

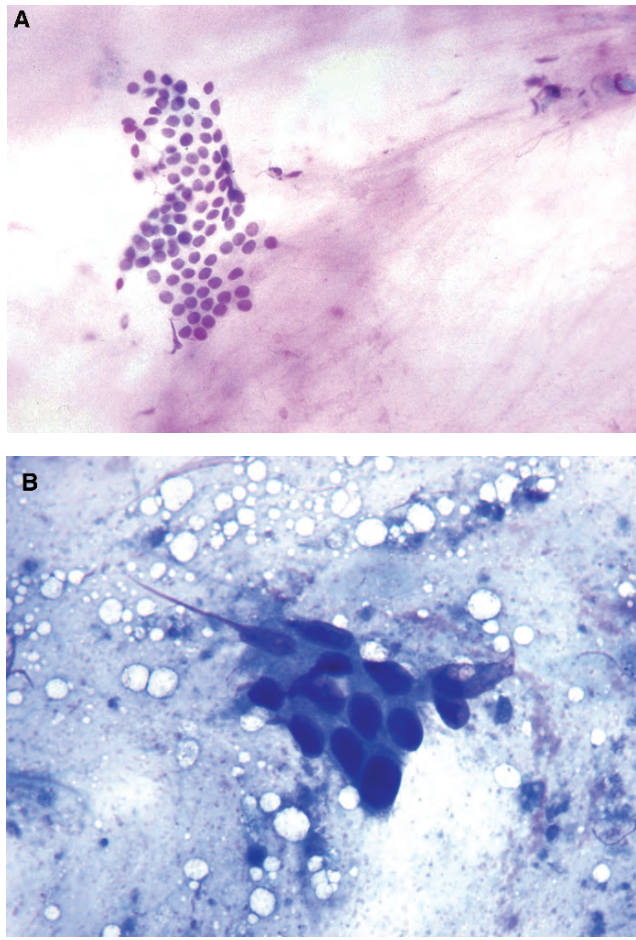


FIG. 9.2. Low-grade mucoepidermoid carcinoma. (A and B) Most aspirates are hypocellular, containing rare flat sheets of epidermoid cells in a background of abundant thick mucoid material. (A, Smear, Papanicolaou; B, Smear, Diff-Quik.)

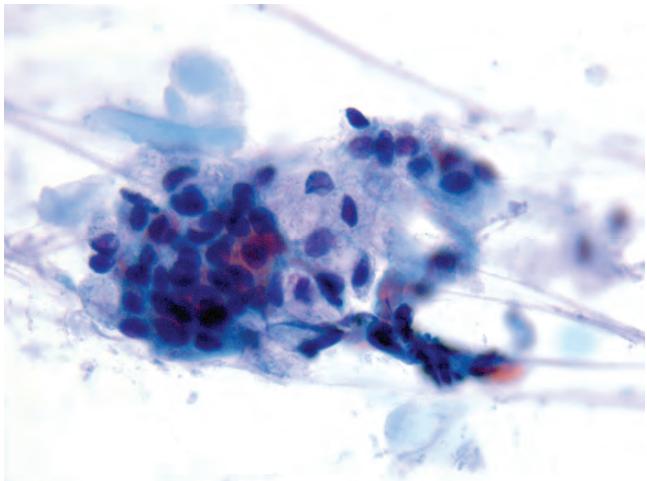


FIG. 9.3. Low-grade mucoepidermoid carcinoma. Groups with a combination of epidemoid cells, intermediate cells, and admixed mucinous cells is the most diagnostic feature. (Thin-layer preparation, Papanicolaou.)

material with cellular debris (Fig. 9.3). Background lymphocytes are seen in approximately 20% of cases, and some cases will contain hemosiderin-laden macrophages and cholesterol crystals. Other cell types that can be seen include clear, columnar, and oncocytic. In general, low-grade tumors have a much higher percentage of mucinous epithelial cells while high-grade ones have more epidermoid cells. Myoepithelial cells are not present, a feature that can be useful for distinguishing mucoepidermoid carcinomas from lesions such as pleomorphic adenoma with mucinous metaplasia.

Cytologic Features of Low-Grade Mucoepidermoid Carcinoma

- Hypocellular
- Combination of 3 epithelial cell types:
  - Epidermoid
  - Mucus
  - Intermediate
- Histiocytes and muciphages
- Thick mucoid background with cellular debris
- Background lympocytes may be present

The epidermoid cells of mucoepidermoid carcinoma occur in bland cohesive but crowded sheets with squamoid features including well-defined intercellular borders and abundant dense waxy cytoplasm (Fig. 9.4). Keratinization is not a feature of mucoepidermoid carcinomas helping to distinguish them from other cysts containing keratinizing squamous cells such as metastatic squamous cell carcinoma. Mucus cells or goblet cells have a very low N:C ratio owing to the presence of abundant delicate pale-staining mucoid cytoplasm (Fig. 9.5). The nuclei of mucus cells are eccentrically placed against the cytoplasmic membrane and indented. They can easily be mistaken for foamy histiocytes and muciphages, but the latter generally have more multivacuolated cytoplasm, and the nucleus is often more centrally placed and round. Mucus cells can form loosely cohesive groups or be present individually within a group of epidermoid cells. Intermediate cells resemble the suprabasalar cells of the epidermis or metaplastic squamous cells of the cervix. In contrast to epidermoid cells, intermediate cells have a much higher N:C ratio with darker oval nuclei (Fig. 9.6). They are columnar to polygonal and occur in flat cohesive sheets that are

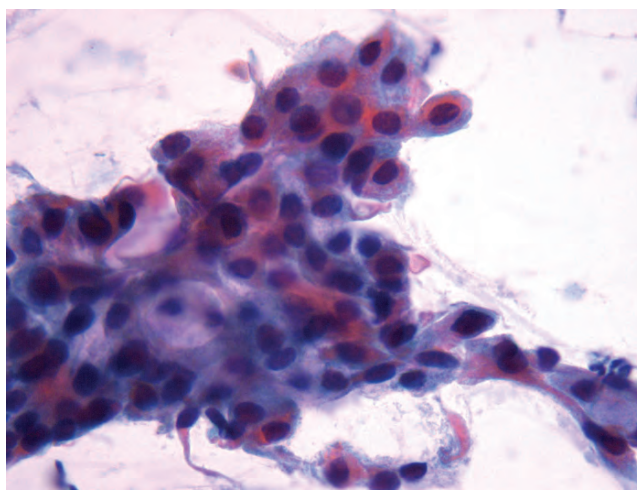


FIG. 9.4. Low-grade mucoepidermoid carcinoma. Epidermoid cells (A and B) occur in flat cohesive sheets and have abundant dense waxy cytoplasm and well defined cell borders. (Smears, Papanicolaou.)

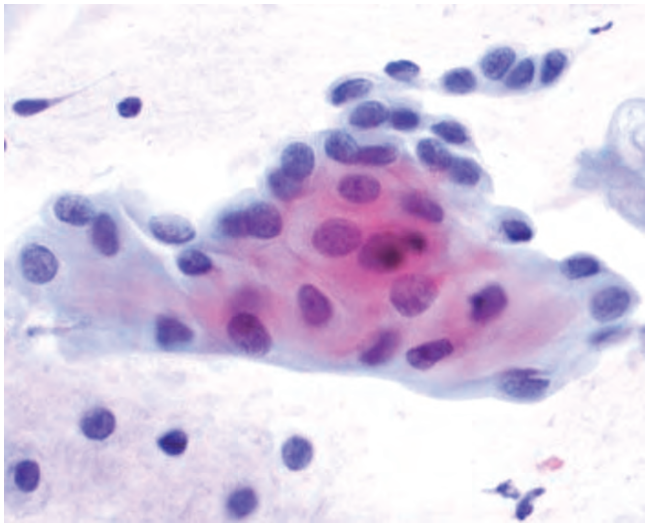


FIG. 9.4. (continued)

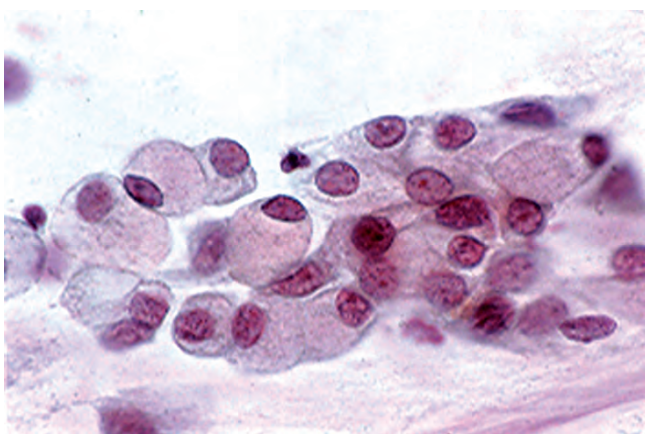


FIG. 9.5. Low-grade mucoepidermoid carcinoma. Mucus cells have abundant delicate mucinous cytoplasm with a peripherally placed and indented nucleus. (Smear, Papanicoloau.)

sometimes elongated. Nuclei of all 3 cell types are cytologically bland, exhibiting minimal nuclear pleomorphism. Nuclei are oval with evenly distributed chromatin, and only slight irregularities

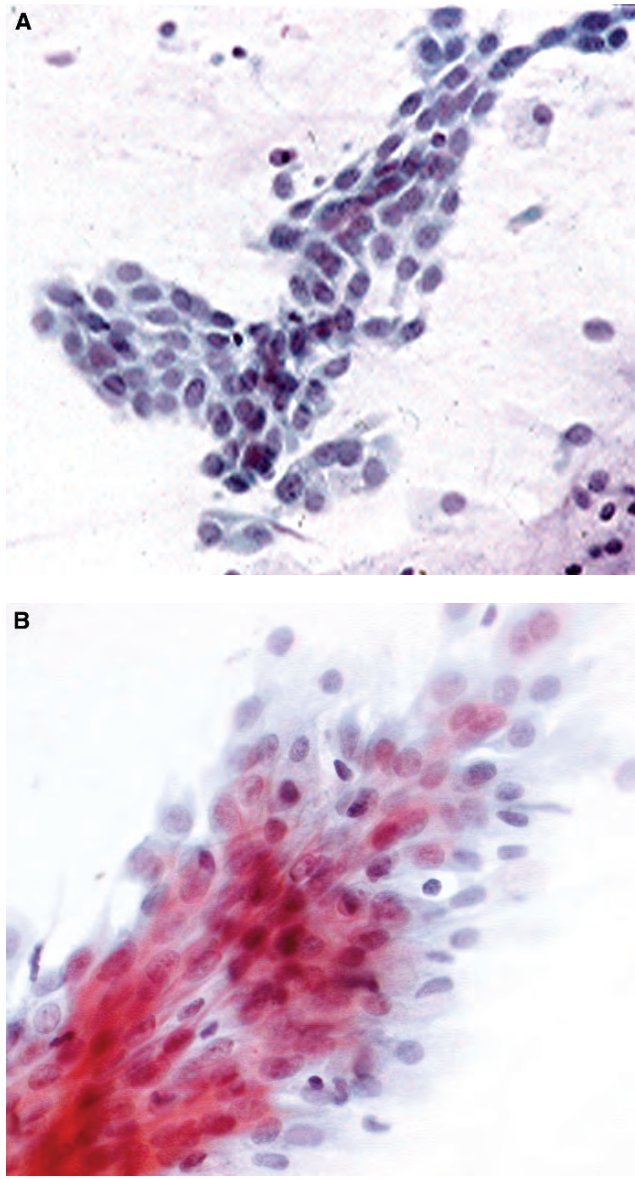


FIG. 9.6. Low-grade mucoepidermoid carcinoma. (A and B) Intermediate cells have a high N:C ratio, small amounts of dense cytoplasm, and a dark oval nucleus (Smears, Papanicolaou.)

and grooves in the nuclear membrane. Nucleoli are often present but are small and indistinct. The key to recognizing mucoepidermoid carcinoma in aspirates is to identify groups of epithelial cells containing a combination of cell types, especially the presence of a mucus cell within a group of intermediate or epidermoid cells.

There are several very rare variants of mucoepidermoid carcinoma, but the most problematic one and more commonly encountered is the oncocytic variant, which can easily be mistaken for an onco-

Keratinization is not a feature of mucoepidermoid carcinomas.

cytic neoplasm (see Chapter 8) (Fig. 9.7). The oncocytic variant of mucoepidermoid carcinoma is characterized by a predominant uniform population of epithelial cells with moderate amounts of homogenous dense granular oncocytic cytoplasm. The cytoplasm contains abundant mitochondria as in other oncocytes. Aspirates are more cellular and solid than conventional low-grade mucoepidermoid carcinomas. The cells are cytologically bland and in crowded sheets and singly. The clue to the diagnosis is the presence of rare admixed mucinous goblet cells that are best detected using special mucin stains in cell block material. The presence of background mucoid material is variable.

Cytologic Features of the Oncocytic Variant
of Mucoepidermoid Carcinoma

- Cellular aspirate
- Sheets of bland cells with densely granular oncocytic cytoplasm
- Rare admixed mucus cells
- Clean to variably mucoid background

Mucocele

Aspirates of mucoceles are hypocellular and lack an epithelial component (Fig. 9.8). They contain variable amounts of background mucoproteinaceous material, abundant foamy histiocytes and muciphages, occasional giant cells, acute and chronic inflammatory cells, cholesterol crystals, and fibrous tissue fragments. Since mucoceles sometimes occur secondary to sialolithiasis, fragments of salivary gland stones may be found. Retention cysts and ductal

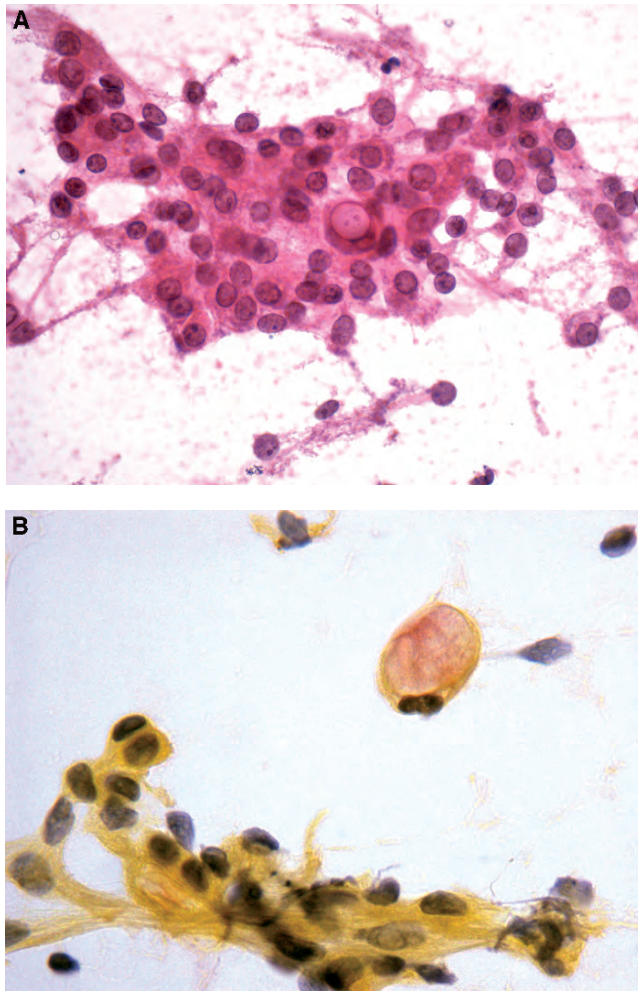


FIG. 9.7. Oncocytic variant of mucoepidermoid carcinoma. (A) Aspirates are cellular and comprised of cells with abundant densely granular oncocytic cytoplasm. (Smear, Papanicolaou); (B) Mucicarmine stain confirms the presence of intracellular mucin.

cysts are similar but contain occasional groups of epithelial cells and have a cleaner background with fewer inflammatory cells since the mucin is encapsulated; importantly, the combination

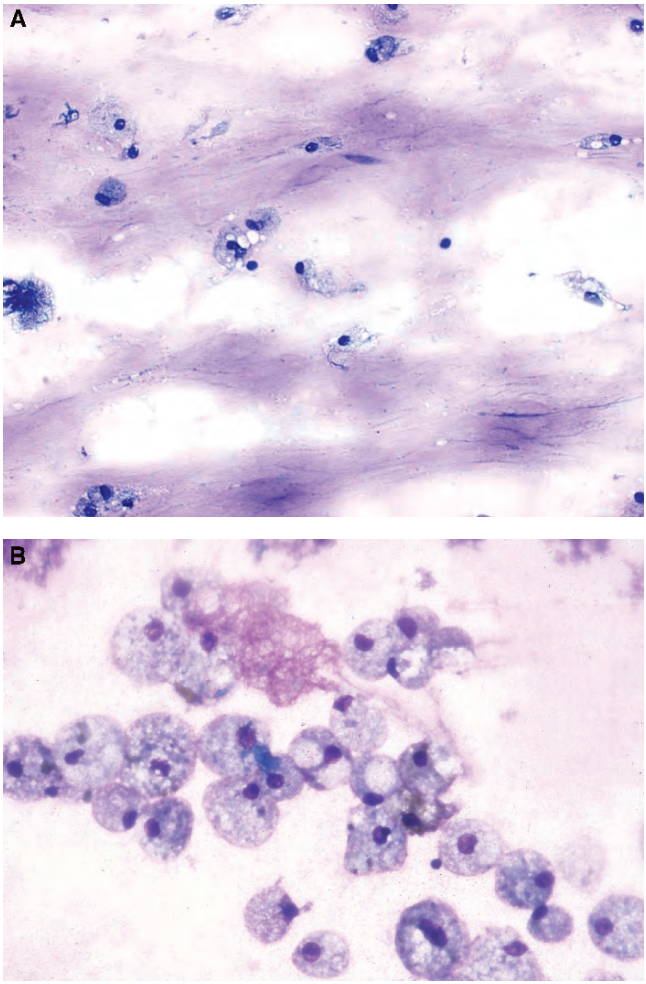


FIG. 9.8. Mucocele. (A) Hypocellular aspirate containing histiocytes, acute and chronic inflammation, and a watery mucoid background. An epithelial component is absent. (B) Abundant foamy histiocytes and muciphages. (Smears, Diff-Quik.)

of crowded epidermoid, intermediate, and mucus cells seen in mucoepidermoid carcinomas is not present. This distinction can be subtle and caution is warranted. Good clinical correlation is

recommended for all cases, and in most instances, a descriptive diagnosis should be used with a differential diagnosis that includes mucoepidermoid carcinoma, especially for cases where a residual mass is present after aspiration.

Cytologic Features of Mucoceles

- Hypocellular specimen
- Mucoproteinaceous background
- Foamy histiocytes, muciphages, and occasional giant cells
- Acute and chronic inflammation
- Cholesterol crystals
- Fibrous tissue fragments

Differential Diagnosis and Pitfalls

As alluded to above, low-grade mucoepidermoid carcinomas are distinguished from mucoceles and retention cysts by the presence of at least rare groups of epithelial cells that include epidermoid and intermediate cells with admixed mucus cells. For cases of retention cysts secondary to sialolithiasis, ciliated columnar cells are sometimes present, and their presence is a useful sign that one is not dealing with a mucoepidermoid carcinoma. The epithelial cells of mucoepidermoid carcinoma are present in sheets, but the epithelial cells are crowded together. In some cases, the distinction between mucoepidermoid carcinoma and a benign cyst can be subtle. A major pitfall is that a significant subset of mucoepidermoid carcinomas yield aspirates with only thick mucoid material admixed with histiocytes and muciphages. For this reason, the aspiration of any cystic salivary lesion containing background mucin should suggest the differential diagnosis of a low-grade mucoepidermoid carcinoma, and this is especially true for mucinous cysts in the major salivary glands. Furthermore, any residual mass present following drainage by FNA of a cystic lesion should be reaspirated. In many cases, a residual mass will need to be surgically excised to exclude the possibility of a neoplasm.

Differential Diagnosis of Low-Grade Mucoepidermoid Carcinoma

- Mucocele
- Retention cyst
- Warthin tumor with mucinous metaplasia
- Cystic pleomorphic adenoma with metaplasia
- Sclerosing polycystic adenosis
- Ductal papilloma
- Metastasis

A small subset of Warthin tumors with squamous and/or mucinous metaplasia can be difficult to distinguish from low-grade mucoepidermoid carcinoma, especially for those rare cases where the metaplastic changes are extensive (Fig. 9.9). Fortunately, most cases of Warthin tumor will contain characteristic sheets of evenly spaced oncocytes and lymphocytes in a background with granular debris, and the mucinous and/or squamous components will be

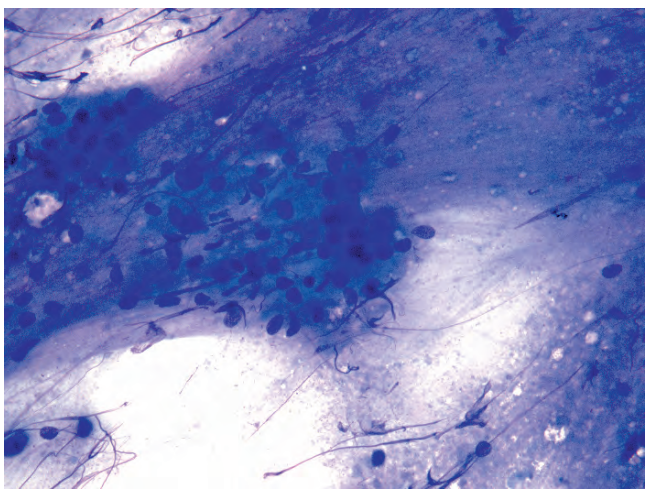


FIG. 9.9. Warthin tumor with extensive mucoid background. When abundant thick background mucin is present together with squamoid cells, it can be very difficult to exclude a low-grade mucoepidermoid carcinoma. (Smear, Diff-Quik.)

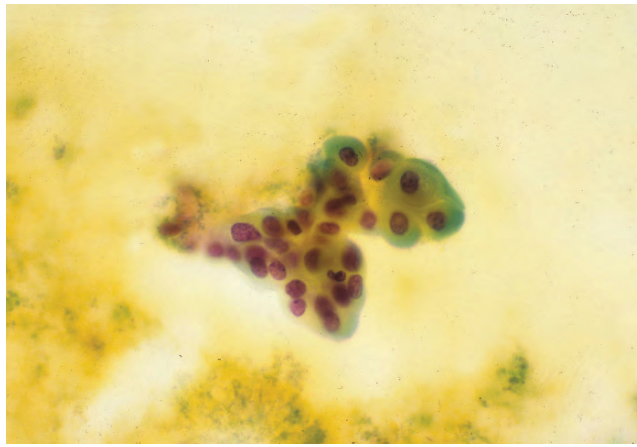


FIG. 9.10. Cystic pleomorphic adenoma with metaplastic changes. (Smear, Papanicolaou.)

minor. Pleomorphic adenomas are occasionally cystic and can exhibit squamous and mucinous metaplastic changes mimicking a low-grade mucoepidermoid carcinoma (Fig. 9.10). For well-sampled lesions, the aspirates will contain the characteristic metachromatic fibrillar matrix of pleomorphic adenoma. In addition, if material is available for immunohistochemical studies, the presence of myoepithelial cells can help to exclude a mucoepidermoid carcinoma. Most often, cystic pleomorphic adenomas with squamous metaplastic changes produce keratin pearls, and this is not a characteristic feature of mucoepidermoid carcinomas. Metastatic head and neck squamous cell carcinoma to an intraparotid lymph node can occasionally be misinterpreted as mucoepidermoid carcinoma (Fig. 9.11). This pitfall can be avoided by obtaining adequate clinical information, since it would be very unusual for a patient to develop metastatic disease to a major salivary gland from an unknown primary. In addition, cystic and metaplastic pleomorphic adenomas, as well as metastatic squamous cell carcinomas, are often at least partially keratinizing, helping to exclude a mucoepidermoid carcinoma. Ductal papillomas are a group of rare benign salivary gland tumors that occur most often in the minor salivary glands. Aspirates can be indistinguishable from mucoepidermoid carcinoma since both bland epidermoid and mucinous cells can

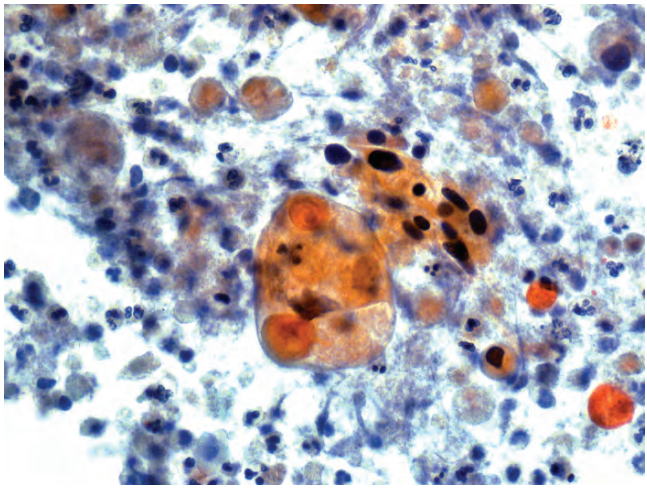


FIG. 9.11. Metastatic head and neck squamous cell carcinoma to an intraparotid lymph node. Clinical history of a head and neck squamous cell carcinoma together with the presence of keratinizing cells helps to exclude a mucoepidermoid carcinoma. (thin-layer preparation, Papanicolaou.)

be found. A papillary architectural arrangement, if present, would favor a lesion other than mucoepidermoid carcinoma.

Sclerosing polycystic adenosis is a rare and recently described cytologic mimic of low-grade mucoepidermoid carcinoma. It can also be confused with benign and low-grade oncocytic salivary gland tumors. Sclerosing polycystic adenosis is analogous to fibrocystic disease of the breast, and most indications are that it is non-neoplastic. Patients present with a very slowly enlarging, firm mass, usually in the parotid gland. Aspirates are characterized by flat sheets of evenly spaced epidermoid-appearing cells, bland ductal cells with granular oncocytic cytoplasm, occasional vacuolated cells, and scattered cells with apocrine metaplastic changes (Fig. 9.12). The background is clean and contains watery to delicate mucoproteinaceous material. The apocrine metaplastic change is the most useful cytologic feature, which together with a prolonged benign clinical history favors sclerosing polycystic adenosis over most other salivary gland tumors, including low-grade mucoepidermoid carcinoma.

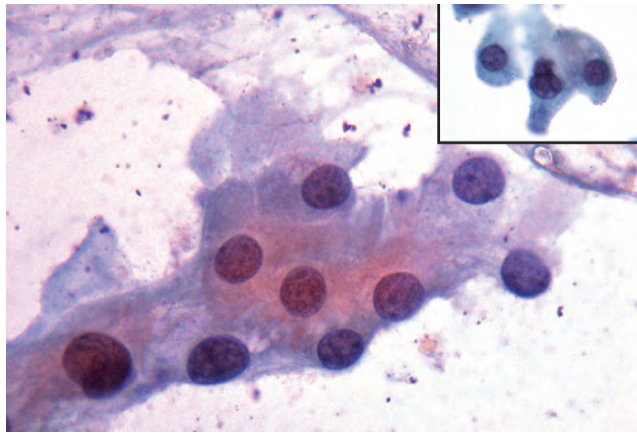


FIG. 9.12. Sclerosing polycystic adenosis. This rare reactive lesion is characterized by a combination of epidermoid cells with eosinophilic cytoplasm and focal cells with apocrine changes (inset). (Thin-layer preparation, Papanicolaou.)

Cytologic Features of Sclerosing Polycystic Adenosis

- Variably cellular aspirate
- Watery to mucoproteinaceous background
- Small sheets of evenly spaced epidermoid cells
- Bland ductal cells with oncocytic cytoplasm
- Occasional vacuolated clear cells
- Apocrine metaplastic changes

Nonmucinous Salivary Gland Cysts: Branchial Cleft Cyst Versus Metastatic Squamous Cell Carcinoma

Lymphoepithelial cysts (branchial cleft-like cysts) of the parotid gland are congenital cysts that may not become clinically apparent until adulthood. They are analogous to the branchial cleft cysts of the neck and occur in the parotid gland, intraparotid lymph nodes, and oral cavity. They are more common in men. Aspirates of lymphoepithelial cysts of the parotid gland contain turbid proteinaceous fluid, histiocytes, keratin debris, and bland mature and anucleate squamous cells (Fig. 9.13); occasional benign-appearing glandular

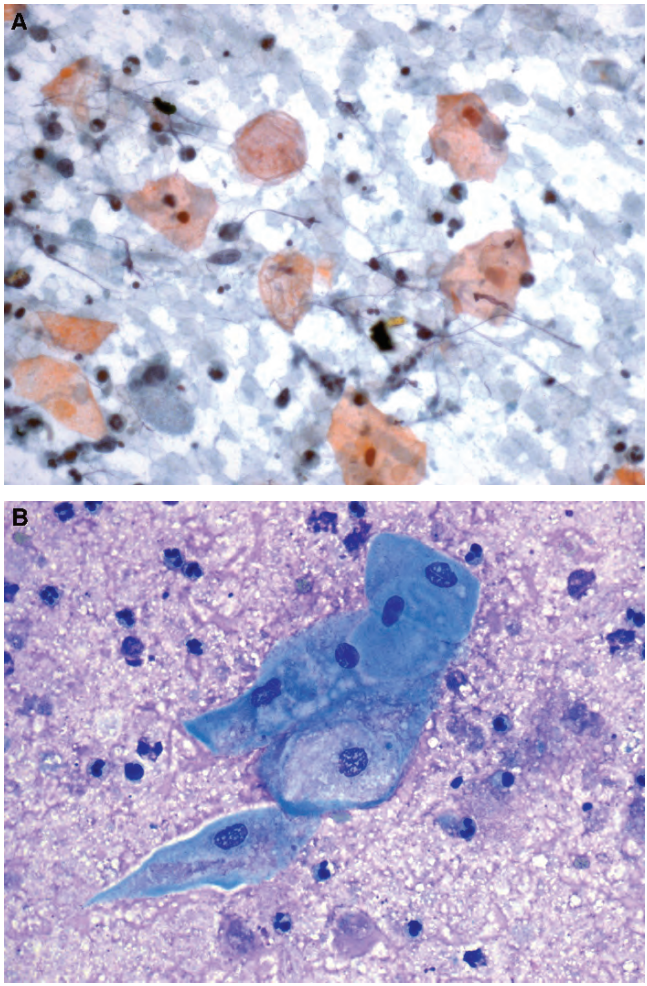


FIG. 9.13. Lymphoepithelial cyst. (A and B) Aspirates contain abundant mature and anucleate squamous cells, often with a lymphoid background. (A, Smear, Papanicolaou; B, Smear, Diff-Quik.)

cells that may be mucin-containing or ciliated can sometimes also be seen. Variable numbers of background lymphocytes are usually present and can include lymphohistiocytic aggregates. In a young patient without a history of squamous cell carcinoma, the findings

are compatible with a benign lymphoepithelial cyst. However, in patients over 30 or without adequate clinical information, it may be impossible to distinguish a lymphoepithelial cyst from a very well-differentiated metastatic squamous cell carcinoma of the head and neck. The problem becomes even more challenging for aspirates of lymphoepithelial cysts that contain reactive squamous atypia (Fig. 9.14) secondary to acute inflammation. A descriptive diagnosis is given for such cases. In most instances of metastatic well-differentiated squamous cell carcinoma to an intraparotid lymph node, at least occasional squamous cells will show an increased N/C ratio with hyperchromatic irregular nuclei. Cystic LESA and HIV-associated lymphoepithelial cysts can be indistinguishable from lymphoepithelial cysts in salivary gland aspirates, but a good clinical history will usually reveal evidence of Sjögren's syndrome or HIV infection, respectively, resolving the differential diagnosis. Both contain lymphoepithelial lesions that are not found in simple lymphoepithelial cysts. Another benign salivary gland cyst is polycystic disease of the parotid gland, which yields hypocellular aspirates comprised of watery fluid and few bland low cuboidal

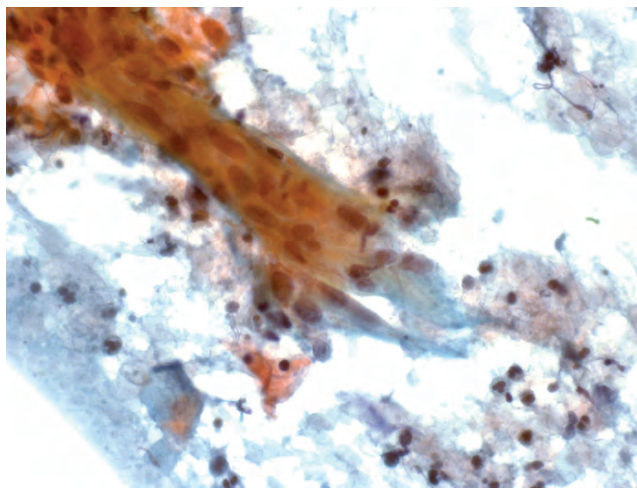


FIG. 9.14. Lymphoepithelial cyst with reactive atypia. Occasionally, atypical squamous cells will be present that can mimic metastatic squamous cell carcinoma. (Smear, Papanicolaou.)

cells. Clinical history is the key, since patients are female with bilateral parotid gland involvement and a characteristic multicystic radiologic appearance.

Cytologic Features of Lymphoepithelial Cysts

- Proteinaceous background
- Mature-appearing squamous cells and anucleate squames
- Variable presence of bland mucinous ductal cells and ciliated cells
- Background lymphocytes and lymphohistiocytic aggregates
- Variable acute inflammation and reactive atypia

Other Nonmucinous Salivary Gland Cysts

Cystadenomas are rare benign multicystic salivary gland tumors that occur slightly more commonly in the minor salivary glands. Aspirates are usually hypocellular and contain a background of cyst debris with eosinophilic material and sometimes psammoma bodies. Cells are cuboidal to columnar and cytologically bland (Fig. 9.15).

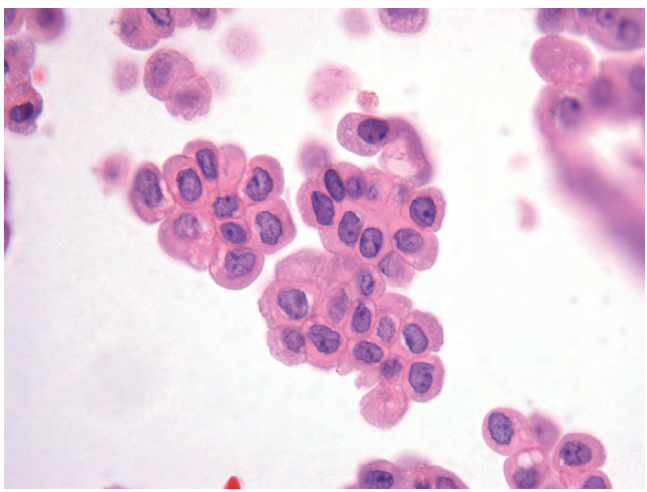


FIG. 9.15. Cystadenoma. Aspirates are hypocellular and contain bland oncocytic cuboidal to columnar cells in small groups and papillary clusters (Smear, Papanicolaou.)

The cytoplasm is most often oncocytic, but some cases can contain mucus or even epidermoid cells resembling low-grade mucoepidermoid carcinoma. Making things even more challenging is its malignant counterpart, cystadenocarcinoma, which is nearly indistinguishable from cystadenoma in aspirates. Cystadenoma and cystadenocarcinoma are distinguished by histologic evidence of invasion rather than by cytologic atypia. Therefore, while a benign cystic lesion would be favored for aspirates of cystadenoma, the cytologic findings are not specific and a descriptive signout with differential diagnosis will be used. The diffierential diagnosis includes low-grade mucoepidermoid carcinoma (especially if mucus cells are present), the papillocystic subtype of acinic cell carcinoma, cystic oncocytoma, ductal papilloma, and cystadenocarcinoma.

Ancillary Techniques

Cystic lesions of the salivary gland, particularly the differential diagnosis of mucocele and low-grade mucoepidermoid carcinoma, can present a diagnostic challenge. For difficult cases, cell block material can be used to perform immunohistochemical stains for keratin and CD68 (i.e., epithelial-containing mucus cells versus muciphages/histiocytes), as well as histochemical stains for mucin. The finding of keratin-positive groups of epidermoid cells and mucus cells in the appropriate context favors a mucoepidermoid carcinoma. For the differential diagnosis of cystic pleomorphic adenoma and mucoepidermoid carcinoma, immunohistochemical stains for myoepithelial cells (e.g., calponin, smooth muscle actin, keratin, S-100) can be used; myoepithelial differentiation is not a feature of mucoepidermoid carcinoma.

Clinical Management and Prognosis

Mucoceles are treated by local excision, including removal of the salivary gland tissue supplying the mucin. The latter is very important, since failure to adequately excise the mucocele can result in recurrent disease, including fistula formation. Because over one-third of cystic salivary gland lesions are neoplastic, surgical excision is usually performed to exclude malignant disease. In contrast to

their high-grade counterparts, low-grade mucoepidermoid carcinomas have an overall excellent clinical prognosis, with a greater than 95% survival rate, although submandibular gland tumors follow a less predictable clinical course. Prognosis is correlated with several independent factors, including site of lesion, histologic grade, clinical stage, and disease-free surgical resection margins. Clinical management of mucoepidermoid carcinomas consists primarily of surgical excision with facial nerve preservation for parotid gland primaries. Dissection of the submandibular triangle is usually done for submandibular lesions. A modified neck dissection is reserved for those patients where a positive lymph node is detected either by FNA or at surgery by frozen section. Postoperative irradiation is used for patients with positive surgical margins, or for patients with metastatic disease or high-grade forms.

Suggested Reading

Brandwein MS, Ivanov K, Wallace DI, et al. Mucoepidermoid carcinoma: a clinicopathologic study of 80 patients with special reference to histological grading. Am J Surg Pathol 2001;25:835–845.

Cheuk W, Chan JKC. Advances in salivary gland pathology. Histopathology 2007;51:1–20.

Droese M. Cytological diagnosis of sialadenosis, sialadenitis, and parotid cysts by fine-needle aspiration biopsy. Adv Oto-Rhino-Layng 1981;26:49–96.

Edit D, Pilch BZ, Osgood R, Faquin WC. Fine-needle aspiration biopsy in sclerosing polycystic adenosis. Diagn Cytopathol 2007;35-444–447.

Goode RK, Auclair PL, Ellis GL. Mucoepidermoid carcinoma of the major salivary glands: clinical and histopathologic analysis of 234 cases with evaluation of grading criteria. Cancer 1998;82:1217.

Jahan-Parwar B, Huberman RM, Donovan DT, Schwartz MR, Ostrowski ML. Oncocytic mucoepidermoid carcinoma of the salivary glands. Am J Surg Pathol 1999;23:523–529.

Klijanienko J, Vielh P. Fine-needle sampling of salivary gland lesions. IV. Review of 50 cases of mucoepidermoid carcinoma with histologic correlation. Diagn Cytopathol 1997;17:92–98.

Layfield LJ, Gopez EV. Cystic lesions of the salivary glands: cytologic features in fine-needle aspiration biopsies. Diagn Cytopathol 2002;27:197–204.

10
High-Grade Salivary Gland Tumors: Salivary Duct Carcinoma, High-Grade Mucoepidermoid Carcinoma, and Carcinoma *ex* Pleomorphic Adenoma

Background

High-grade salivary gland tumors are extremely important to recognize in FNA samples. In contrast to benign and low-grade salivary gland neoplasms, which are managed conservatively, the diagnosis of a high-grade salivary gland tumor will usually result in an aggressive clinical response that can include radical surgical resection, nerve sacrifice, lymph node dissection, and chemoradiation therapy. Among the more common high-grade salivary gland cancers that will be discussed in this chapter are salivary duct carcinoma, high-grade mucoepidermoid carcinoma, and carcinoma *ex* pleomorphic adenoma. With regard to impact on clinical management, the primary goal is to diagnose the tumor as a "high-grade carcinoma," rather than focusing on the specific subtype of high-grade carcinoma. As will be discussed in more detail, this is fortunate, since there is significant cytologic overlap between the 3 major high-grade salivary gland tumors. Other even rarer high-grade salivary gland malignancies that can be encountered include primary squamous cell carcinoma, small cell carcinoma, undifferentiated carcinoma, and certain metastatic cancers.

In contrast to benign and low-grade salivary gland neoplasms, which are managed conservatively, the diagnosis of a high-grade salivary gland tumor will usually result in an aggressive clinical response.

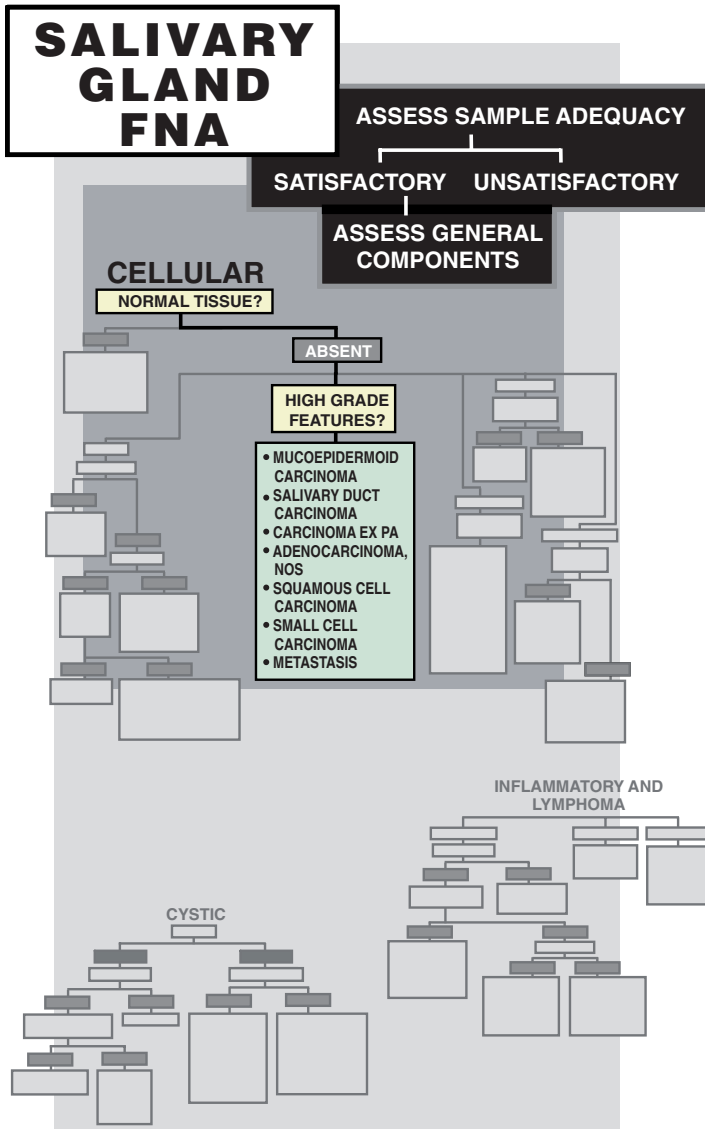


FIG. 10.1. Algorithm for high-grade salivary gland tumors.

Salivary duct carcinoma is a high-grade salivary gland malignancy, accounting for approximately 9% of all malignant salivary gland tumors; over 85% of cases occur in the parotid gland. Up to three-fourths of patients are men, and the peak incidence is in the sixth to seventh decade. Patients usually present with a rapidly enlarging mass, and approximately 25% of patients exhibit symptoms related to facial nerve involvement (e.g., pain and facial paralysis). Histologically, salivary duct carcinoma is often compared with high-grade comedo-type ductal carcinoma of the breast. In addition to its de novo presentation, salivary duct carcinoma is also the most common form of malignancy in carcinoma *ex* pleomorphic adenomas.

Clinicopathologic Features of Salivary Duct Carcinoma

- 9% of all salivary gland malignancies
- 85% occur in the parotid gland
- 80% of cases in men
- Peak incidence in sixth decade

Mucoepidermoid carcinoma represents 10%–15% of salivary gland tumors, and it occurs over a broad age range. It consists of three grades – low, intermediate, and high, although some institutions prefer a two-tiered grading system which may be more practical for cytologic samples. In terms of its clinical behavior, the high-grade form differs markedly from low and intermediate grades. While mucoepidermoid carcinoma in general is the most common salivary gland carcinoma in both children and adults, high-grade mucoepidermoid carcinomas are much less common than the low- and intermediate-grade forms. In addition, in contrast to its low-grade counterpart (see Chapter 9), high-grade mucoepidermoid carcinomas are predominantly solid tumors that present with rapid enlargement often with associated pain, facial paralysis, and surface ulceration.

Carcinoma *ex* pleomorphic adenomas (aka malignant mixed tumors) are uncommon salivary gland tumors representing 5%–12% of salivary gland malignancies. They result from malignant transformation of the benign counterpart, pleomorphic adenoma. There are actually 3 different types of malignant pleomorphic adenoma: carcinoma *ex* pleomorphic adenoma, carcinosarcoma *ex* pleomorphic adenoma, and metastatisizing pleomorphic adenoma. The

latter 2 are quite rare entities, and we will focus upon carcinoma *ex* pleomorphic adenomas. It is estimated that the rate of malignant transformation of pleomorphic adenomas is 5%–7%, and in some reports, the rate of malignant transformation after 15 years is as high as 9.5%. Aside from the associated high morbidity rate, this risk of malignant transformation explains part of the problem faced by patients with recurrent pleomorphic adenomas secondary to incomplete surgical excision. Patients with carcinoma *ex* pleomorphic adenoma are characteristically in the sixth or seventh decade, and present with rapid enlargement of a previous longstanding parotid mass. Most cases occur in the parotid gland, but can also be seen in the submandibular and minor salivary glands, especially the palate. For patients with recurrent pleomorphic adenoma, FNA is often used to monitor for malignant transformation. The diagnostic accuracy of FNA in the identification of a malignant component in pleomorphic adenoma is estimated at 70%–90%. In our experience, a majority of cases are recognizable as malignant, but the benign pleomorphic adenoma component is scant or absent from the aspirate due to sampling issues. Both invasive and noninvasive histologic forms of carcinoma *ex* pleomorphic adenoma are recognized, but they cannot be distinguished by FNA.

Clinicopathologic Features of Carcinoma *Ex* Pleomorphic Adenoma

- 5%–12% of all salivary gland malignancies
- 5%–7% rate of malignant transformation of pleomorphic adenoma
- Salivary duct carcinoma is the most common malignant component
- Parotid gland is the most common site
- Sixth to seventh decade
- Rapid enlargement of a longstanding mass

General Diagnostic Approach

The algorithm for high-grade salivary gland carcinomas is probably the easiest to apply since it depends primarily upon identification of high-grade nuclear features such as nuclear pleomorphism, coarse chromatin, mitotic activity, and necrosis (Fig. 10.1). Aspirates

are typically cellular and include large 3-dimensional groups of malignant cells. Sometimes a background tumor diathesis will be present. Distinction between the various subtypes of high-grade salivary gland malignancies can be difficult due to overlapping cytologic features. Among the most important distinctions to make in the algorithm is to exclude the possibility of metastatic disease, especially in patients with a history of nonsalivary gland cancer.

Diagnostic Criteria

Salivary Duct Carcinoma

Aspirates of salivary duct carcinoma are cellular and consist of large cohesive 3-dimensional clusters of glandular epithelial cells with marked nuclear atypia (Figs. 10.2–10.3). The groups of cells form sheets, papillae, and cribriform clusters. Cells are polygonal with abundant vacuolated to finely granular eosinophilic cytoplasm. In most cases of salivary duct carcinoma, the cytoplasmic vacuoles are negative for mucin. In a subset of salivary duct carcinomas, oncocytic metaplasia is extensively present. Focal squamoid differentiation can also be seen. The nuclei are oval, pleomorphic, and hyperchromatic (Fig. 10.4). A prominent centrally placed nucleolus is characteristically seen. Mitotic activity, atypical mitoses, and background necrotic debris are both easily identified, the latter often as a prominent tumor diathesis. Occasionally, psammoma bodies can be identified. Mucin-rich, micropapillary, and sarcomatoid variants have been described but are rare.

Cytologic Features of Salivary Duct Carcinoma

- Cellular aspirate
- Cribriform, papillary, and sheetlike groups
- Polygonal cells with abundant vacuolated or granular cytoplasm
- Enlarged, pleomorphic and hyperchromatic nuclei
- Prominent central macronucleolus
- Mitoses including atypical forms
- Background necrosis

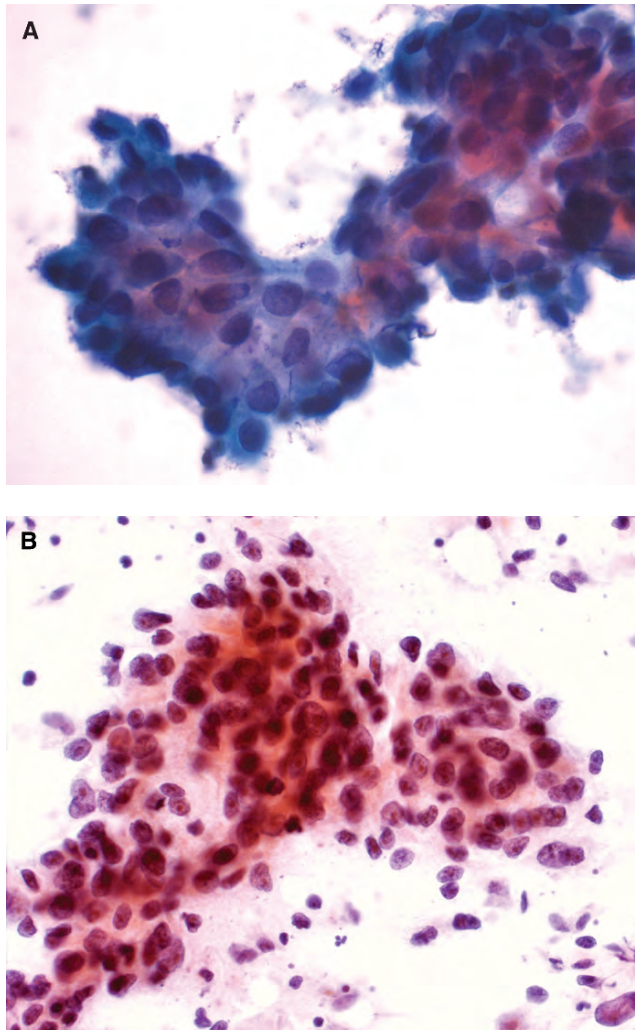


FIG. 10.2. Salivary duct carcinoma. (A and B) Cellular aspirates containing crowded 3-dimensional groups of glandular cells with large hyperchromatic nuclei and prominent nucleoli. (Smears, Papanicoloau.)

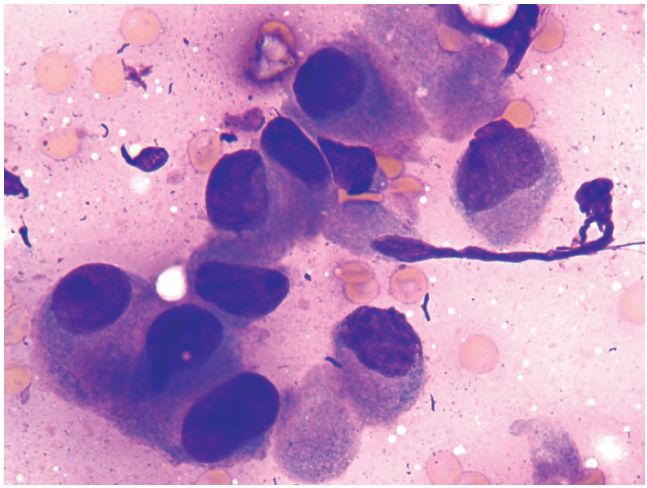


FIG. 10.3. Salivary duct carcinoma. Cells have moderate amounts of vacuolated cytoplasm and large pleomorphic nuclei. (Smear, Diff-Quik.)

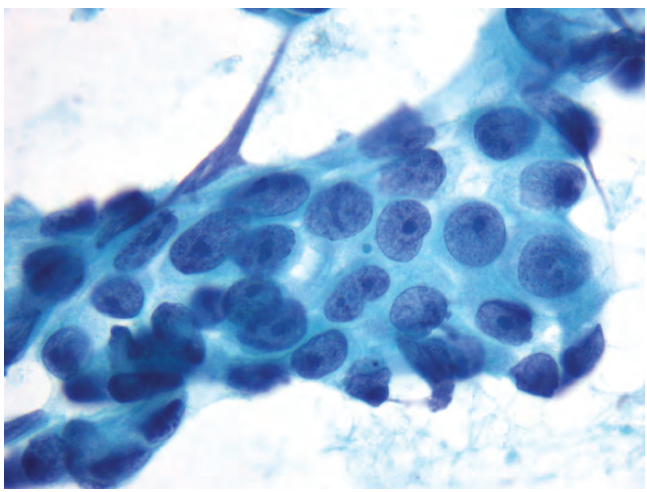


FIG. 10.4. Salivary duct carcinoma. The nuclei are oval, enlarged, and pleomorphic with a prominent central red nucleolus. (Smear, Papanicoloau.)

High-Grade Mucoepidermoid Carcinoma

High-grade mucoepidermoid carcinomas yield cellular aspirates comprised of crowded 3-dimensional groups and sheets of cells with squamoid features (Figs. 10.5–10.6). Cells are polygonal and have moderate amounts of dense waxy cytoplasm; cell borders are usually well defined. Some cases may also contain a subset of intermediate type cells which have a higher N:C ratio. Nuclei are large, hyperchromatic, and oval, often with a prominent nucleolus, and mitoses are readily identified. Careful inspection will sometimes reveal at least rare goblet cells containing cytoplasmic mucin. If a cell block is available, a mucicarmine stain can be used to verify the presence of intracellular mucin. The background contains scattered single cells and in some cases is mucoid. The rare oncocytic variant of mucoepidermoid carcinoma is characterized by cells with abundant eosinophilic granular cytoplasm in sheets (Fig. 10.7). Nuclei are less pleomorphic than conventional high-grade mucoepidermoid carcinomas and may exhibit nuclear grooves and pseudoinclusions. Importantly, scattered mucin-containing cells can be identified, helping to distinguish this rare variant from other oncocytic salivary gland tumors.

Cytologic Features of High-Grade Mucoepidermoid Carcinoma

- Cellular aspirate
- Squamoid cells in crowded 3-dimensional groups and sheets
- Well-defined cell borders
- Enlarged hyperchromatic nuclei with prominent nucleolus
- Occasional goblet cells containing intracellular mucin
- Mitotic activity

Carcinoma ex Pleomorphic Adenoma

Carcinoma *ex* pleomorphic adenomas are characterized by aspirates containing a high-grade malignant component in a background of focal metachromatic, fibrillary matrix with embedded myoepithelial cells characteristic of conventional pleomorphic adenoma (Figs. 10.8–10.9). The malignant component is most often salivary duct carcinoma as described above; however, any of the various forms of salivary gland carcinoma (e.g., mucoepidermoid

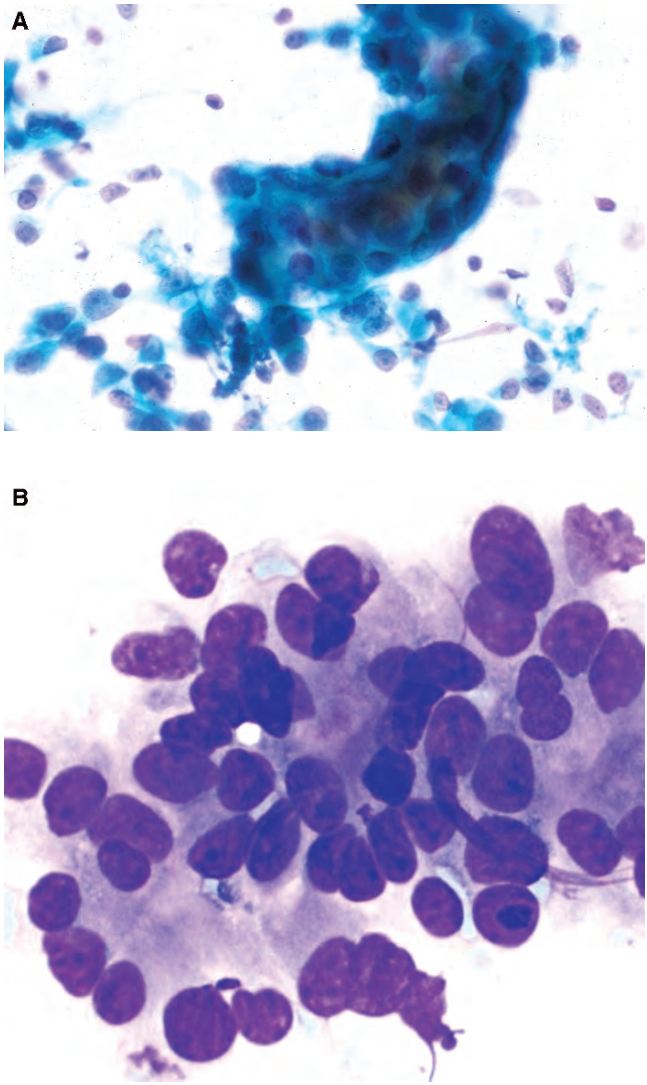


FIG. 10.5. High-grade mucoepidermoid carcinoma. (A and B) Cells have squamoid features with large atypical nuclei. (A, Smear, Papanicoloau; B, Smear, Diff-Quik.)

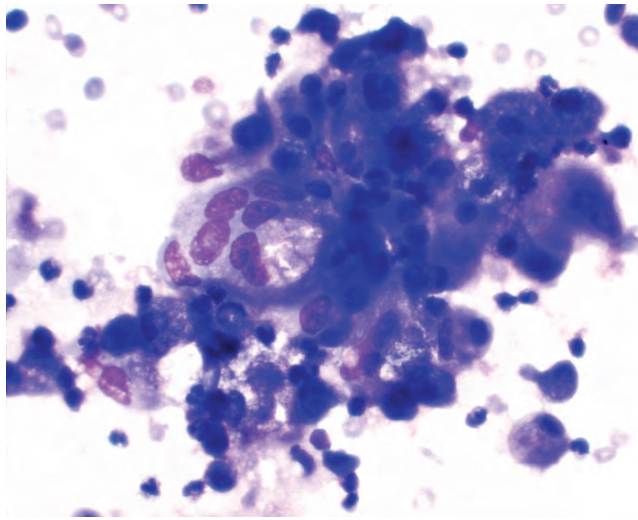


FIG. 10.6. High-grade mucoepidermoid carcinoma. Cells with abundant squamoid to vacuolated cytoplasm, and large nuclei with a prominent nucleolus. (Smear, Diff-Quik.)

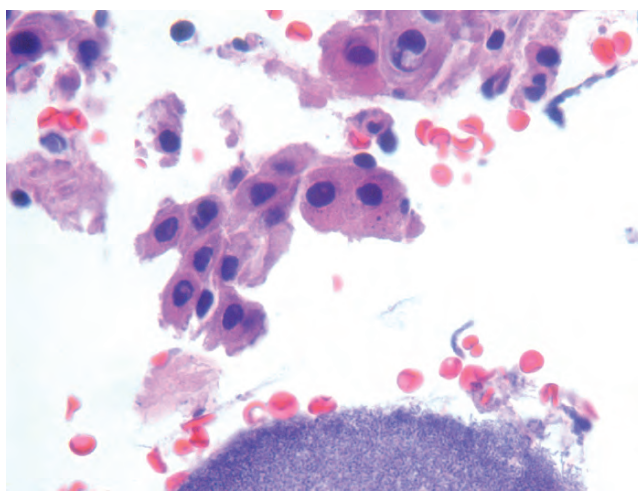


FIG. 10.7. Oncocytic variant of mucoepidermoid carcinoma. Cells have abundant dense granular cytoplasm. (Cell block, H&E).

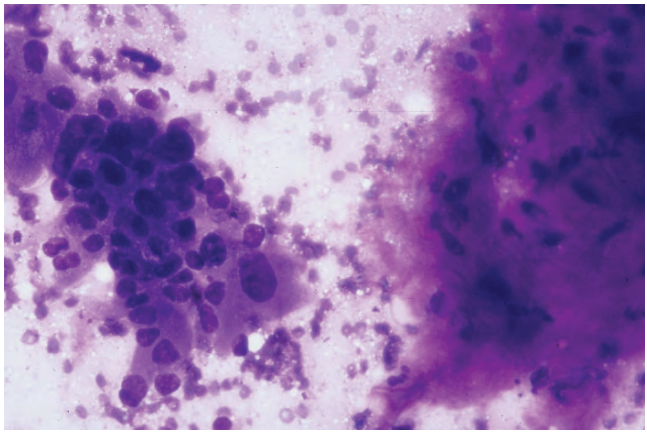


FIG. 10.8. Carcinoma *ex* pleomorphic adenoma. Group of high-grade carcinoma cells opposite the metachromatic matrix material of pleomorphic adenoma (Smear, Diff-Quik.)

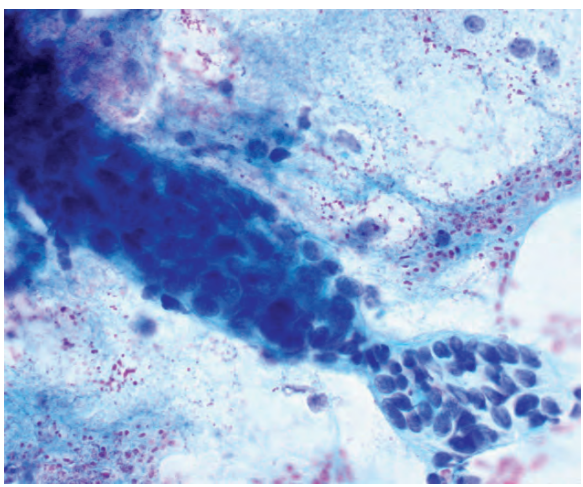


FIG. 10.9. Carcinoma *ex* pleomorphic adenoma. Crowded 3-dimensional group of high-grade carcinoma in a background with pale-staining matrix material (Smear, Papanicoloau.)

carcinoma, adenoid cystic carcinoma, acinic cell carcinoma) can be seen. In some cases, the malignant component will dominate the aspirate so that signs of a precursor pleomorphic adenoma will not be present. Such specimens are indistinguishable from aspirates of salivary duct carcinoma.

Cytologic Features of Carcinoma *Ex* Pleomorphic Adenoma

- Cellular aspirate
- High-grade carcinoma, usually salivary duct carcinoma
- Metachromatic fibrillar matrix with embedded myoepithelial cells

Differential Diagnosis and Pitfalls

The differential diagnosis of high-grade salivary gland tumors includes entities with malignant cytologic atypia. The three most common among these are salivary duct carcinoma, high-grade mucoepidermoid carcinoma, and carcinoma *ex* pleomorphic adenoma. The cytologic features of these 3 carcinomas overlap significantly, and in many cases, it may be extremely difficult if not impossible to definitively distinguish among them by FNA. From the practical standpoint of clinical management, the distinction is not critical since current treatment protocols tend to be similar. This may change in the not so distant future as we learn more about molecular differences between some of these tumors, but hopefully, such differences would also provide us with molecular signatures that could be applied through ancillary studies to improve our diagnostic abilities.

Differential Diagnosis of High-Grade Salivary Gland Tumors

- Salivary duct carcinoma
- High-grade mucoepidermoid carcinoma
- Carcinoma *ex* pleomorphic adenoma
- Primary squamous cell carcinoma
- Small cell carcinoma
- Undifferentiated carcinoma
- Large B-cell lymphoma
- Metastasis

Despite overlapping cytologic features, there are certain findings that can be used to favor one carcinoma over another when considering a differential diagnosis of a high-grade malignancy. Salivary duct carcinomas and high-grade mucoepidermoid carcinomas have similar appearances, especially in view of the squamoid features that can occasionally be seen in salivary duct carcinoma. Although a rare mucinous variant of salivary duct carcinoma exists, the finding of goblet cells containing intracellular mucin strongly favors high-grade mucoepidermoid carcinoma over salivary duct carcinoma. On the other hand, the presence of abundant background necrotic debris, and either papillary or cribriform cytomorphologic patterns, favor salivary duct carcinoma. For carcinoma *ex* pleomorphic adenoma, the key to recognizing it in aspirates is finding cytologic evidence of conventional pleomorphic adenoma, namely the characteristic metachromatic fibrillar matrix with embedded myoepithelial cells. When this material is absent, it will not be possible to distinguish carcinoma *ex* pleomorphic adenoma from other high-grade carcinomas based upon microscopic findings alone.

High-grade mucoepidermoid carcinomas have a predominance of epidermoid cells that raise the challenging differential diagnosis of either metastatic or primary squamous cell carcinoma. Most cases of salivary gland squamous cell carcinoma will represent metastatic disease from head and neck squamous cell carcinoma, particularly cutaneous squamous cell carcinomas metastatic to intraparotid lymph nodes (Fig. 10.10). Clinical history is important when the diagnosis of primary squamous cell carcinoma is considered. Primary squamous cell carcinoma is rare and accounts for less than 1% of all salivary gland tumors. It occurs most often as a high-stage malignancy in the parotid gland of elderly men. When it is nonkeratinizing, the distinction between squamous cell carcinoma and high-grade mucoepidermoid is particularly difficult. When present, keratinization is helpful since it is not a characteristic of high-grade mucoepidermoid carcinomas (Note: remember that keratinization can be seen in certain benign salivary gland tumors such as pleomorphic adenoma.) In addition to keratinization, the finding of goblet cells and demonstration of intracellular mucin favors the diagnosis of mucoepidermoid carcinoma over squamous cell carcinoma, especially when present in combination with both epidermoid and intermediate cells. Another rare entity,

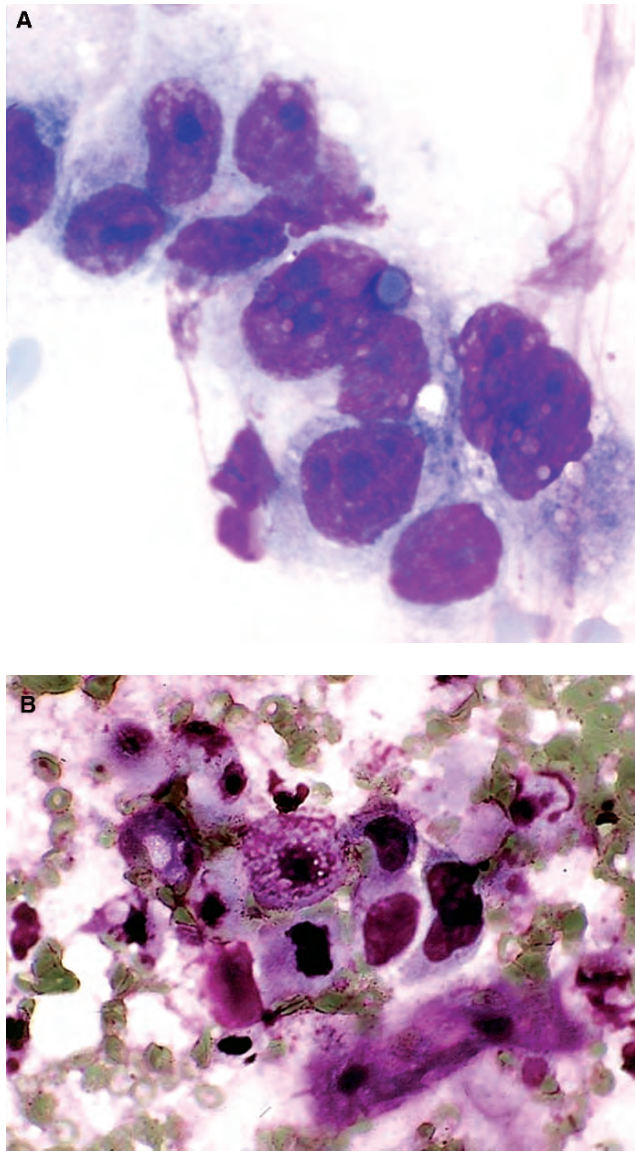


FIG. 10.10. Primary salivary gland squamous cell carcinoma. (A and B) Aspirates are cellular and comprised of nonkeratinizing squamous cells with high-grade nuclear features. (Smears, Diff-Quik.)

adenosquamous carcinoma, cannot be distinguished from the more common high-grade mucoepidermoid carcinoma by FNA since the former differs primarily by the presence of an in situ mucosal component. Similarly, large cell undifferentiated carcinoma of the salivary gland is a diagnosis of exclusion that requires histologic evaluation for classification – it is a rare high-grade malignancy defined by its large pleomorphic cells with abundant cytoplasm and absence of features of other salivary gland tumor types.

Among the most important entities to exclude from the differential diagnosis of high-grade carcinomas is metastatic disease since this can impact the patient's clinical workup and management. This differential diagnosis can be problematic based upon cytomorphologic criteria alone. For example, aspirates of metastatic high-grade ductal carcinoma of the breast look virtually identical to salivary duct carcinoma. As in other anatomic sites sampled by FNA where metastatic disease is a consideration, the key to diagnosing a metastasis depends upon having appropriate clinical information. For any patient with a history of a nonsalivary gland malignancy, consideration should be given to the possibility that the carcinoma represents a metastasis, and appropriate directed ancillary studies should be performed. Among the most useful things to do whenever metastatic disease enters into the differential diagnosis, is direct head-to-head comparison between the microscopic features of the aspirated tumor and those of the patient's original primary cancer.

Among the most important entities to exclude from the differential diagnosis of high-grade carcinomas is metastatic disease.

Primary Small Cell Carcinoma

Primary small cell carcinoma of the salivary gland is highly aggressive and is morphologically similar to small cell carcinomas from other sites. It represents approximately 2% of salivary gland malignancies and occurs primarily in the parotid gland. The overall survival rate is cited as 40%–50%. Aspirates are cellular and characterized by single cells and loose cell clusters (Fig. 10.11).

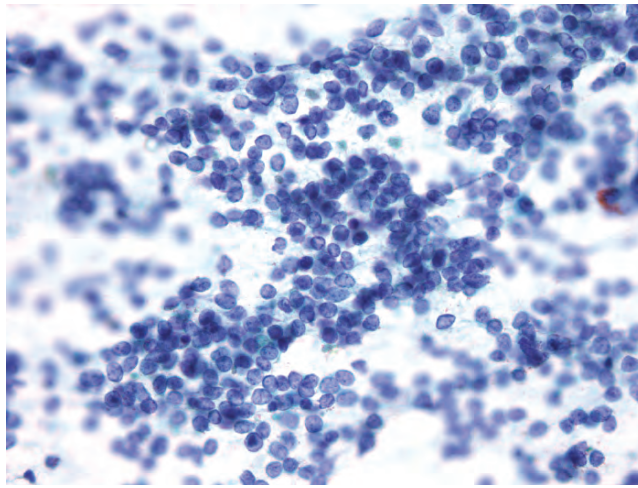


FIG. 10.11. Primary small cell carcinoma of the parotid gland. Cells are dispersed and have a high N:C ratio, focal nuclear molding, and background necrosis. (Smears, Papanicoloau.)

Individual cells are 2 to 3 times larger than a mature lymphocyte and have scant cytoplasm. The nuclei are oval to round with dispersed granular chromatin, moderate pleomorphism, and indistinct to absent nucleoli. Focal nuclear molding is usually apparent, as are frequent mitoses, apoptosis, and background necrosis. The differential diagnosis includes other small round blue cell tumors, and in the salivary gland, one should consider entities such as metastatic small cell carcinoma, metastatic melanoma, and lymphoma. Immunohistochemical studies are useful since small cell carcinoma is keratin-positive and usually reacts with one or more neuroendocrine markers: NSE, synaptophysin, chromogranin, CD56, and CD57. The keratin immunoreactivity shows a characteristic paranuclear dotlike pattern and helps to exclude large cell lymphoma and melanoma. Small cell carcinoma is negative for CD45, S-100, HMB-45, and MART-1. In contrast to small cell carcinoma of the lung, 75% of primary salivary gland small cell carcinomas are immunohistochemically positive for keratin 20. Thus, in a majority of cases, the overall cytomorphologic and immunohistochemical features are analogous to Merkel cell carcinoma.

Cytologic Features of Primary Small Cell Carcinoma

- Dispersed cells and loose clusters
- High N:C ratio
- Round to oval nuclei with dispersed granular chromatin and indistinct nucleoli
- Nuclear molding
- Frequent mitoses and apoptosis
- Background necrosis
- 75% are keratin 20+

In addition to small cell carcinoma as a subset of undifferentiated carcinomas, other undifferentiated carcinomas that can be encountered include large cell undifferentiated carcinoma and lymphoepithelial carcinoma. Some undifferentiated carcinomas arise as transformations from a better differentiated tumor to a higher grade or dedifferentiated form. Such dedifferentiated carcinomas of the salivary gland are rare. This process is uncommon but has been observed most frequently in acinic cell carcinomas and is now recognized to occur in many different tumor types. The prognosis for patients with dedifferentiated carcinoma is very poor. Aspirates of dedifferentiated carcinomas are cellular and contain highly malignant-appearing cells with pleomorphic nuclei, high N:C ratio, and frequent mitoses (Fig. 10.12).

Ancillary Techniques

Among the high-grade salivary gland carcinomas, special staining for intracellular mucin is probably among the most useful ancillary studies that can be performed on cell block material. As discussed, the presence of scattered cells containing intracellular mucin strongly favors a diagnosis of mucoepidermoid carcinoma over either salivary duct carcinoma or squamous cell carcinoma. Both mucoepidermoid carcinoma and salivary duct carcinoma (but not carcinoma *ex* pleomorphic adenoma) are immunohistochemically negative for myoepithelial markers. If cell block material is available for ancillary studies, the immunoprofile for salivary duct carcinomas can also be useful in distinguishing it from other high-grade carcinomas. Its profile includes reactivity for CEA, EMA,

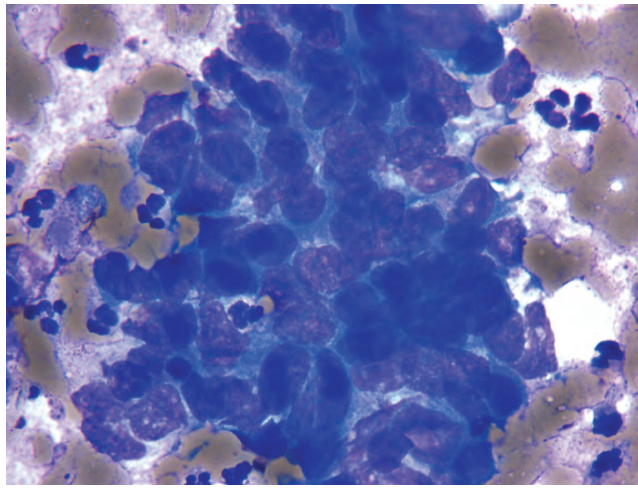


FIG. 10.12. Dedifferentiated acinic cell carcinoma. The aspirate is comprised of highly malignant-appearing undifferentiated cells. (Smear, Diff-Quik.)

androgen receptor, and HER-2/neu. Interestingly, in view of its cytomorphologic resemblance to breast cancer, it also often shows reactivity for the breast marker GCDFP-15, but it is negative for estrogen and progesterone receptors; it is also variably positive for PSA and prostatic acid phosphatase.

Clinical Management and Prognosis

The clinical management for high-grade salivary gland carcinomas includes radical surgical resection, neck dissection, and postoperative adjuvant radiotherapy particularly for widely invasive tumors. The use of postoperative radiotherapy has been shown to improve locoregional control. Thus, given the therapeutic consequences, accurate FNA diagnosis is important. Most of the high-grade carcinomas, and especially salivary duct carcinoma, can metastasize to distant sites, including lungs, bones, liver, and brain. For salivary duct carcinoma, the 10-year survival rate is estimated at 35%, with most patients dying within 4 years. Among carcinoma *ex*

pleomorphic adenomas, both invasive and noninvasive carcinomas are recognized histologically. The noninvasive type is analogous to squamous cell carcinoma in situ, and lacks metastatic potential. For invasive tumors, the prognosis depends upon the type of carcinoma present, but they are generally very aggressive with a less than 50% 10-year survival rate. For high-grade mucoepidermoid carcinomas, the survival rate approaches 55%; however, tumors of the submandibular gland have an overall poorer prognosis.

Suggested Reading

Boahene DK, Olsen KD, Lewis JE, et al. Mucoepidermoid carcinoma of the parotid gland: the Mayo Clinic experience. Arch Otolaryngol Head Neck Surg 2004;130:849–856.

Etges A, Pinto DS, Kowalski LP et al. Salivary duct carcinoma: immunohistochemical profile of an aggressive salivary gland tumor. J Clin Pathol 2003;56:914–918.

Fowler MH, Fowler J, Ducatman B, Barnes L, Hunt JL. Malignant mixed tumors of the salivary gland: a study of loss of heterozygosity in tumor suppressor genes. Mod Pathol 2006;19:350–355.

Henke AC, Cooley ML, Hughes JH, Timmerman TG. Fine-needle aspiration cytology of small-cell carcinoma of the parotid. Diagn Cytopathol 2001;25:126–129.

Henley JD, Geary WA, Jackson CL, Wu CD, Gnepp DR. Dedifferentiated acinic cell carcinoma of the parotid gland: a distinct rarely described entity. Hum Pathol 1997;28:869–873.

Hocwald E, Korkmaz H, Yoo GH, et al. Prognostic factors in major salivary gland tumors. Laryngoscope 2001;111:1434.

Nagao T, Gaffey TA, Olsen KD, Serizawa H, Lewis JE. Small cell carcinoma of the major salivary glands: clinicopathologic study with emphasis on cytokeratin 20 immunoreactivity and clinical outcome. Am J Surg Pathol 2004;28:762–770.

Nasser SM, Faquin WC, Dayal Y. Expression of androgen, estrogen, and progesterone receptors in salivary gland tumors: frequent expression of androgen receptor in a subset of malignant salivary gland tumors. Am J Clin Pathol 2003;119:801–806.

Nigam S, Kumar N, Jain S. Cytomorphologic spectrum of carcinoma *ex* pleomorphic adenoma. Acta Cytol 2004;48:309–314.

Skalova A, Starek I, Vanecek T, et al. Expression of HER-2/neu gene and protein in salivary duct carcinomas of parotid gland as revealed by fluorescence in-situ hybridization and immunohistochemistry. Histopathol 2003;42:348–356.

11
Spindle Cell Tumors: Spindled Myoepithelioma, Myoepithelial-Predominant Pleomorphic Adenoma, and Schwannoma

Background

As in other anatomic sites, FNA of spindle cell lesions of the salivary glands is a very challenging area of cytopathology. This is due in part to the wide range of benign and malignant neoplastic tumors as well as reactive lesions with a spindle cell cytomorphology that may be encountered. In addition to the variety of lesions, salivary gland tumors with a predominant spindle cell pattern are rare, accounting for less than 3% of salivary gland aspirates. The most common spindle cell lesions of the salivary gland are myoepithelial-predominant pleomorphic adenoma, myoepithelioma, and schwannoma, which will be the focus of this chapter. Other rare, but important, spindle cell lesions in the differential diagnosis, including nodular fasciitis, solitary fibrous tumor, myoepithelial carcinoma, spindle cell carcinoma, and melanoma, will also be discussed.

The most common spindle cell lesions of the salivary gland are myoepithelial-predominant pleomorphic adenoma, myoepithelioma, and schwannoma.

The World Health Organization recognized myoepitheliomas as a distinct entity in 1991. They are rare benign neoplasms accounting for approximately 1.5% of all salivary gland tumors. Myoepitheliomas are a recurring theme, since they are considered in several differential diagnostic categories as a result of their various cell morphologies,

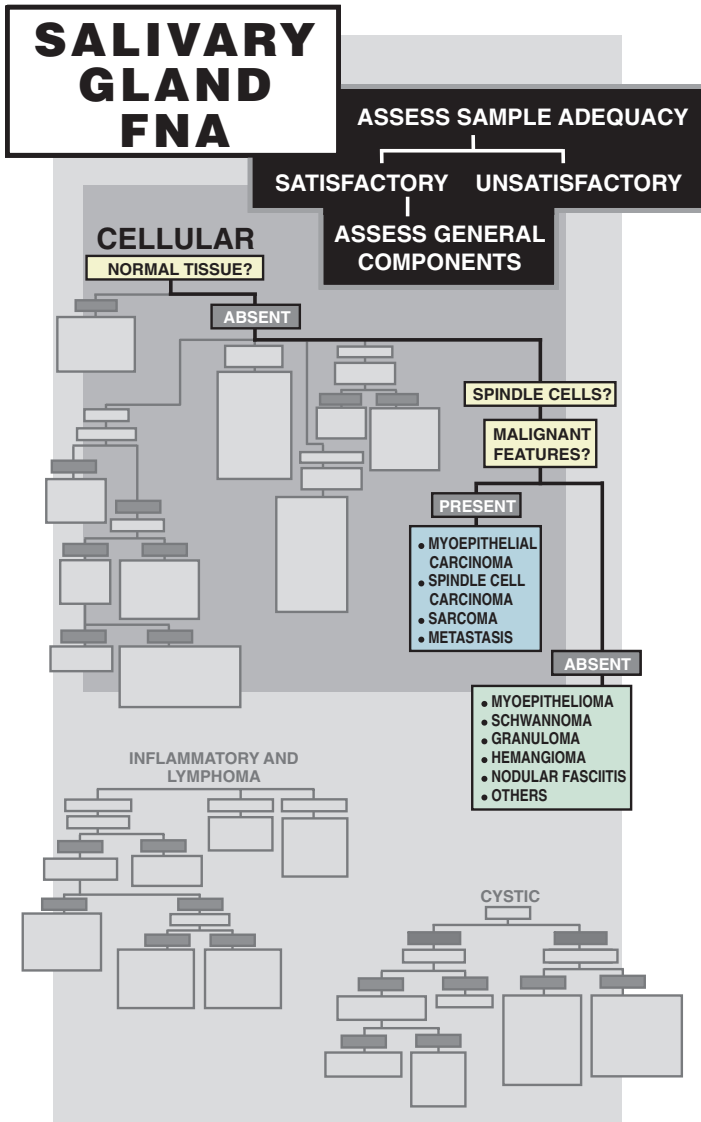


FIG. 11.1. Algorithmic approach to spindle cell tumors of the salivary gland.

including spindled, plasmacytoid, epithelioid and polygonal, stellate, and clear. Thus, the spindled cell form of myoepithelioma is just one subset that presents a particular diagnostic challenge. Most of the myoepithelial-predominant tumors detected in both fine needle aspirates and histologic specimens are actually myoepithelial cell-rich pleomorphic adenomas rather than true myoepitheliomas (see Chapter 6). The distinction, however, is mainly academic since the clinicopathologic features and management of both tumors are very similar. Myoepitheliomas are distinguished histologically from pleomorphic adenomas by the absence (or near absence) of ducts and lack of a chondroid stromal component. Few tumors when well sampled will meet these stringent histologic criteria. Myoepitheliomas can be found in both major and minor salivary glands, with the parotid gland and palate being the most common sites. They occur with equal frequency in both genders, and patients have an average age at presentation of 44 years. As with pleomorphic adenomas, myoepitheliomas can undergo malignant transformation to myoepithelial carcinoma or to various forms of carcinoma *ex* myoepithelioma.

Myoepitheliomas are distinguished from pleomorphic adenomas by the absence of ducts and lack of a chondriod stromal component.

Clinicopathologic Features of Myoepitheliomas

- 1.5% of all salivary gland tumors
- Absence of ducts and chondroid stroma
- Most common sites: parotid gland and palate
- Cytomorphologic patterns:
  - Spindled
  - Plasmacytoid (hyaline)
  - Epithelioid
  - Stellate
  - Clear
- Equal gender predilection
- Average age at presentation: 44 years

Schwannomas (neurilemmomas) are among the more common spindle cell tumors of the head and neck, where approximately 40% of cases occur. When a salivary gland is involved, it is usually the parotid gland and associated facial nerve or, less often, the submandibular gland. Schwannomas were first described by Verocay in 1910 and are the most common form of peripheral nerve sheath tumor. They are solitary, benign, encapsulated tumors originating from the Schwann cell of peripheral, autonomic, and cranial nerves. There are four major forms of schwannoma that differ in their clinicopathologic features: conventional, cellular, melanotic, and plexiform. Their clinical presentation is similar to that of pleomorphic adenoma – a gradually enlarging, often painless mass, although in some cases, facial weakness, pain, or paralysis can be seen. Schwannomas occur equally in both genders with the majority presenting in adults during the fourth decade of life. Most occur within subcutaneous tissues. A subset of cases, vestibular schwannomas, can be associated with neurofibromatosis type 2 (NF2). Aspirates of schwannoma are often reported as painful. Based upon reports from the literature, FNA has a high accuracy (approximately 89%) for diagnosing schwannoma because of its characteristic cytomorphologic and immunocytochemical pattern, but these tumors can also be very difficult to distinguish from certain cellular pleomorphic adenomas and other spindle cell tumors of the head and neck, especially if material for ancillary studies is not available.

Clinicopathologic Features of Schwannomas

- 40% occur in the head and neck
- Neuroectodermal origin
- Most common peripheral nerve sheath tumor
- Solitary, benign, and encapsulated
- Four forms:
    - Conventional
    - Cellular
    - Melanotic
    - Plexiform
- Equal gender predilection
- Fourth decade
- Subset associated with NF2

General Diagnostic Approach

Salivary gland aspirates exhibiting a predominant spindle cell pattern raise a broad differential diagnosis that includes reactive lesions, benign neoplasms, and malignancies (Fig. 11.1). Because of the diagnostic challenge posed by aspirates of spindle cell lesions, detailed clinicoradiologic information and thorough sampling are very important. A key decision point in the evaluation of a spindle cell lesion is whether malignant cytologic features such as nuclear pleomorphism, hyperchromasia, atypical mitoses, and necrosis are present. While certain cytomorphologic characteristics such as nuclear shape, character of the cytoplasm, cytoarchitecture of groups, and presence of extracellular stromal elements are useful for making a definitive classification, many of the spindle cell lesions have remarkably similar and overlapping microscopic features. For this reason, material for ancillary studies is crucial for narrowing the differential diagnosis.

Diagnostic Criteria

Spindled Myoepithelioma and Myoepithelial-Predominant Pleomorphic Adenoma

Spindled myoepitheliomas and myoepithelial-predominant spindled pleomorphic adenomas yield cellular aspirates comprised of cytologically bland myoepithelial cells arranged haphazardly in cohesive groups or as isolated spindle cells (Figs. 11.2–11.4). Most cases lack appreciable background stromal material, but a subset will contain small amounts of metachromatic stroma which can be fibrillary, myxoid, or hyaline. The stromal material is cytologically identical to that seen in aspirates of pleomorphic adenoma, but it lacks a chondroid component. The cells of myoepithelioma have various cytomorphologic forms, including plasmacytoid, epithelioid, clear, stellate, and spindled, with the plasmacytoid form being the most common (see also Chapter 6). These different cytomorphologic forms can be seen in combination, but more often, one will predominate, and when that form is spindled, it can create a diagnostic challenge. The myoepithelial cells in the spindle

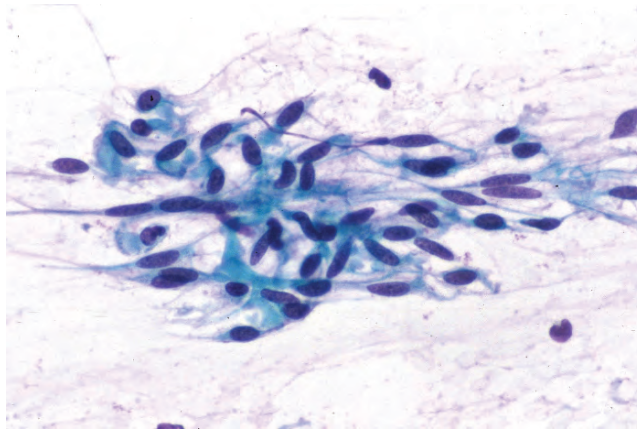


FIG. 11.2. Spindled myoepithelioma. Cluster of haphazardly arranged spindled myoepithelial cells. (Smear, Papanicolaou.)

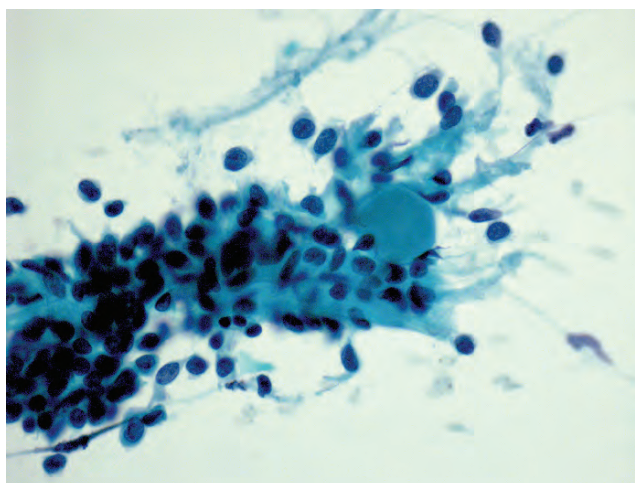


FIG. 11.3. Spindled myoepithelioma. Focal dense hyaline stroma is present inbetween the spindled cells (Smear, Papanicolaou.)

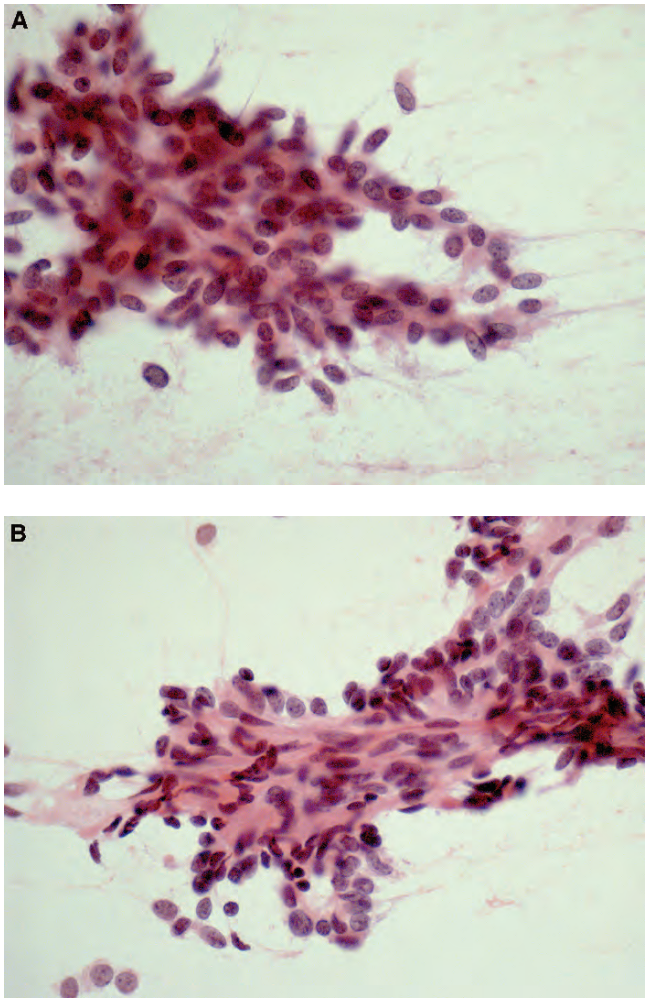


FIG. 11.4. Spindled myoepithelioma. (A and B) Cells have elongate nuclei with rounded ends, evenly dispersed chromatin, and scant pale cytoplasm (Smear, Papanicolaou.)

cell type are elongate with uniform ovoid to fusiform nuclei that have rounded ends. The chromatin is evenly dispersed, nucleoli are indistinct, and mitotic activity is absent or rare. In Papanicolaou

stained preparations, subtle nuclear grooves can often be found. The cytoplasm is scant and pale, and lacks the bipolar cytoplasmic processes seen in myofibroblastic lesions such as nodular fasciitis (Fig. 11.4).

Cytologic Features of Spindled Myoepithelioma

- Moderately cellular
- Cohesive groups and single cells
- Uniform ovoid to elongate nuclei with rounded ends
- Even chromatin, nuclear grooves, and indistinct nucleoli
- Small amounts of pale cytoplasm
- Scant to absent myxoid, fibrillar, or hyaline stroma

Schwannoma

Aspirates of schwannoma are often hypocellular, bordering on nondiagnostic to variably cellular, consisting of cohesive groups of spindled to rounded cells within a fibromyxoid connective tissue stroma (Figs. 11.5–11.6). Individual groups tend to be large with irregular frayed borders. Tissue fragments often appear fibrillar owing to the many fine cytoplasmic processes. Some groups can

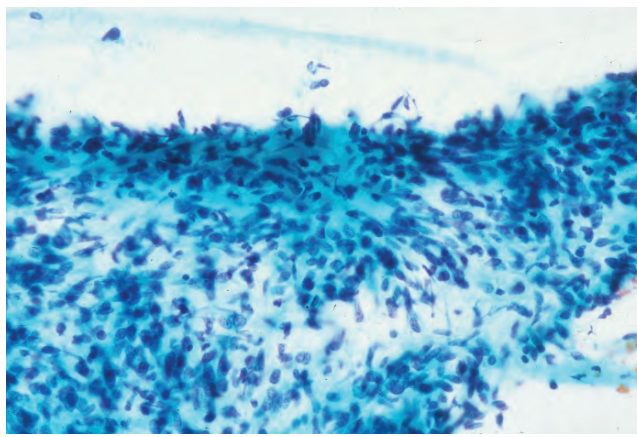


FIG. 11.5. Schwannoma. Spindled cells within a fibromyxoid stroma with subtle palisading. (Smear, Papanicolaou.)

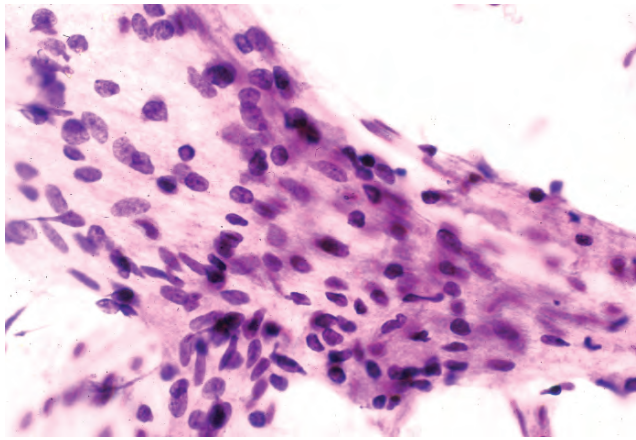


FIG. 11.6. Schwannoma. Large fragment of spindle cells within a loose fibrillar stroma with frayed edges. (Smear, modified H&E.)

also exhibit a whorling pattern reminiscent of a squamous morule. Nuclei are plump to elongate and wavy with pointed ends, sometimes imparting a fishhook shape. The chromatin is evenly distributed, nucleoli are inconspicuous, and intranuclear inclusions are sometimes present. Overall, the nuclei are cytologically bland, although the less common schwannomas with "ancient change" include scattered hyperchromatic nuclei with smudged chromatin and pleomorphism, but mitotic activity is rare to absent. The cytoplasm of individual cells is pale, delicate, and poorly defined. The background is clean and contains scattered single cells, isolated stripped fishhook-shaped nuclei, and occasional mast cells. An important cytologic clue to the diagnosis of schwannoma is the presence of subtle nuclear palisading within hypocellular fibrillar stromal fragments. This feature is representative of the characteristic Verocay bodies (Fig. 11.7).

An important cytologic clue to the diagnosis of schwannoma is the presence of subtle nuclear palisading within hypocellular fibrillar stromal fragments (Verocay body).

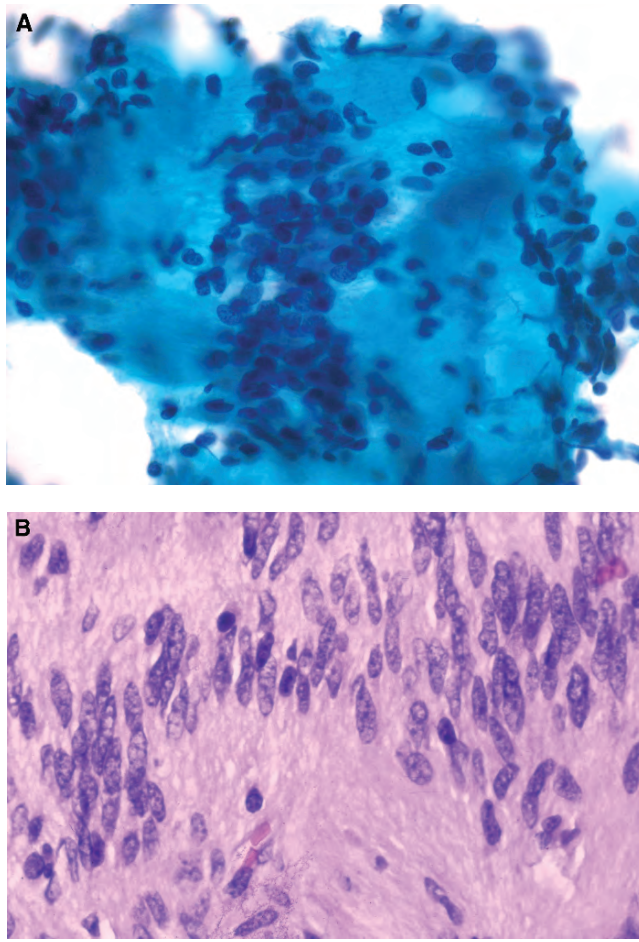


FIG. 11.7. Schwannoma. (A and B) Cells have elongate wavy nuclei with pointed ends forming a palisaded arrangement known as a Verocay body. (A, Smear, Papanicolaou; B, Cell block, H&E.)

Cytologic Features of Schwannoma

- Variably cellular
- Large fibrillar or myxoid groups with frayed edges
- Elongate wavy or fishhook-shaped nuclei with pointed ends

- Even chromatin, indistinct nucleoli, intranuclear inclusions
- Mitoses are rare to absent
- Poorly defined pale cytoplasm
- Focal fibrillar fragments with nuclear palisading (Verocay body)

Differential Diagnosis and Pitfalls

The most common diagnostic pitfall in the evaluation of spindle cell aspirates of the salivary gland, particularly the parotid gland, is the distinction between cellular pleomorphic adenoma (or myoepithelioma) and schwannoma. In contrast to schwannomas that have long wavy nuclei tapering to a pointed end, the nuclei of myoepithelial cells have rounded ends and are less elongate. The stromal component is not a useful distinguishing cytologic feature (in fact it is a pitfall), since in both entities it is metachromatic and myxoid to fibrillar. While cellular pleomorphic adenomas can very rarely exhibit nuclear palisading, the presence of focal nuclear palisading associated with fibrillar areas strongly favors a diagnosis of schwannoma. Aspirates of neurofibroma closely resemble that of schwannoma; however, they are unencapsulated lesions, exhibit variation in nuclear size and shape with some hyperchromatic forms, lack palisading, and immunohistochemical stains usually demonstrate the presence of scattered neurofilament-positive axons (Table 11.1). Confirmatory immunohistochemical marker studies using cell block material are recommended in the evaluation of any spindle cell aspirate. Schwannomas are strongly positive for S-100 and vimentin, and variably immunoreactive with GFAP. They are immunohistochemically negative for keratin, smooth muscle markers, and neurofilament. In contrast, myoepitheliomas are immunoreactive with keratins, smooth muscle actin, p63 and calponin, and weakly reactive for S-100 and GFAP. The immunoprofile of myoepithelial cells is unique among spindle cell lesions.

The diagnosis of spindle cell lesions is challenging even in biopsy material. Therefore, caution is warranted in evaluating aspirates of these lesions. Many of the benign spindle cell lesions of the head and neck are comprised of fibroblasts and myofibroblasts with similar cytomorphology. Clinical information including the exact tissue site, size, rate of growth, and age of the patient can be extremely useful in differentiating among these lesions. Adequate material for ancillary

TABLE 11.1. Features of selected benign head and neck spindle cell lesions.

Benign Spindle Cell Lesion	Key Cytologic Features	Key Immuno-markers:	Ker-atin	S-100	Calponin	SM Actin	Neuro-fila-ment	Vimen-tin	Desmin	EMA	CD99	HMB-45	Leu-7
Spindled Myoepithelioma & Myoepithelial-Predominant Pleomorphic Adenoma	Oval to elongate nuclei with rounded ends; Metachromatic myxoid stroma		+	+	+	+	−	+	±	+	−	−	−
Schwannoma	Wavy or fishhook nuclei with taper-ing pointed ends; Palisading in fibromyxoid areas		−	++	−	−	−	+	−	−	−	−	−
Neurofibroma	Heterogeneous pattern of oval and wavy nuclei; Absence of palisading Fibro-myxoid stroma		−	+	−	−	+	+	−	−	−	−	+
Leiomyoma	Fascicles of spindle cells; Cigar-shaped nuclei; Abundant eosinophilic cyto-plasm; Background stripped nuclei		−	−	+	+	−	+	+	−	±	−	−

TABLE 11.1. (continued)

Benign Spindle Cell Lesion	Key Cytologic Features	Key Immuno-markers:	Ker-atin	S-100	Calponin	SM Actin	Neuro-fila-ment	Vimen-tin	Desmin	EMA	CD99	HMB-45	Leu-7
Nodular Fasciitis	Spindled and stellate myofibroblasts; Plump elongate and oval nuclei; Wispy bipolar cytoplas-mic processes; Loose groups with collagen; Mitoses may be present; Myxoid and bloody background		−	−	+	+	−	+	−	−	−	−	−
Fibromatosis	Fibroblasts and myofibroblasts; Plump elongate and oval nuclei		−	−	+	+	−	+	±	−	−	−	−
Granuloma	Bland reniform nuclei; Vacuolated cytoplasm		−	−	−	−	−	+	−	−	−	−	−
Solitary Fibrous Tumor	Hypercellular groups; Monotonous tapered spindle cells; Dense ropy collagen; Absence of myxoid stroma		−	−	±	−	−	+	−	−	+	−	±

studies, preferably as a paraffin-embedded cell block, is recommended. Because an FNA containing bland spindle cells can be quite difficult, especially when material for ancillary studies is limited, a note recommending close clinical follow-up and re-biopsy is suggested should the lesion fail to resolve or should it show evidence of growth.

Clinical information including the exact tissue site, size, rate of growth, and age of the patient can be extremely useful in sorting out the nature of the lesion.

While much less common than spindled pleomorphic adenoma, myoepithelioma, and schwannoma, several other benign spindle cell lesions should be considered in the differential diagnosis of spindle cell salivary gland aspirates (Table 11.1). A select group of them will be considered here, and as described above, detailed clinical information in addition to ancillary marker studies are necessary for accurate interpretation. A common pitfall in the differential diagnosis is granulomatous inflammation. Epithelioid histiocytes, either in the setting of a reactive (foreign body or infectious) condition, or sarcoidosis, can mimic a spindle cell tumor. The cells form loose clusters and have bland reniform nuclei and moderate amounts of vacuolated cytoplasm (Fig. 11.8). Birefringence can be used to look for foreign material. In infectious conditions, the background can be necrotic or suppurative. Immunohistochemical stains are positive for CD68 but negative for other markers; special stains for organisms and/or microbiologic studies are indicated.

Nodular fasciitis is traditionally considered a reactive myofibroblastic lesion, usually in young adults. that in some cases is associated with previous trauma. The diagnosis is usually strongly suggested by the clinical history of a rapidly enlarging, often tender, subcutaneous nodule. FNA simply serves to confirm the clinical suspicion. It is important for the clinician to be aware that nodular fasciitis is a self-limiting condition and that recurrence is uncommon. Aspirates of nodular fasciitis are characterized by loosely cohesive groups of spindle-shaped and stellate myofibroblasts admixed with collagen. Cells have plump oval to elongate nuclei, dispersed chromatin, small distinct nucleoli, and wispy, tapering bipolar cytoplasmic processes (Fig. 11.9).

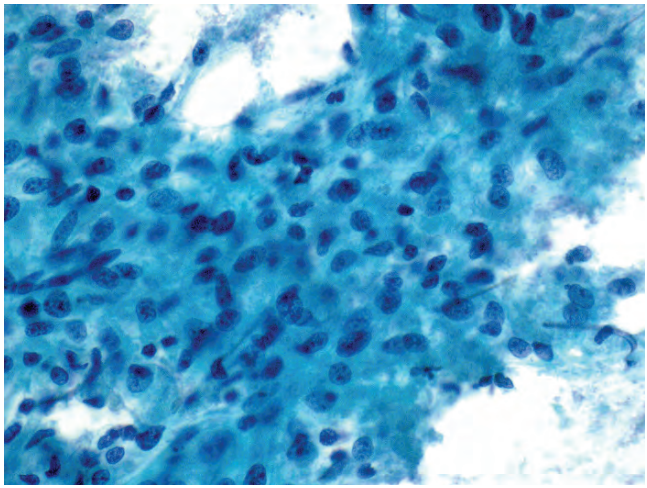


FIG. 11.8. Granuloma associated with sarcoidosis. Cells with a spindled appearance are haphazardly arranged in loose groups and have bland reniform nuclei with moderate amounts of vacuolated cytoplasm. (Smear, Papanicolaou.)

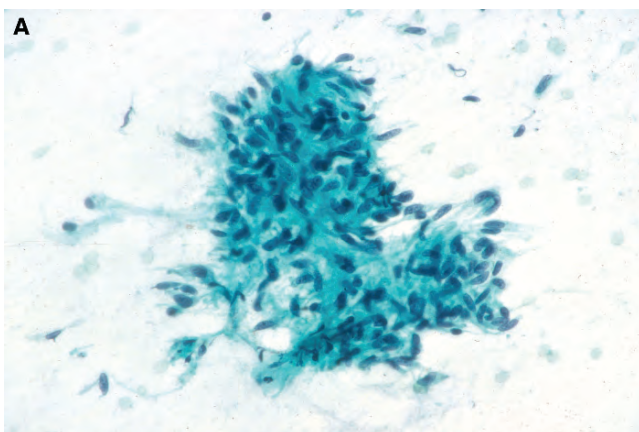


FIG. 11.9. Nodular fasciitis. (A and B) Collagenized groups of myofibroblasts with elongate bland nuclei, and wispy bipolar cytoplasmic processes. (Smears, Papanicolaou.)

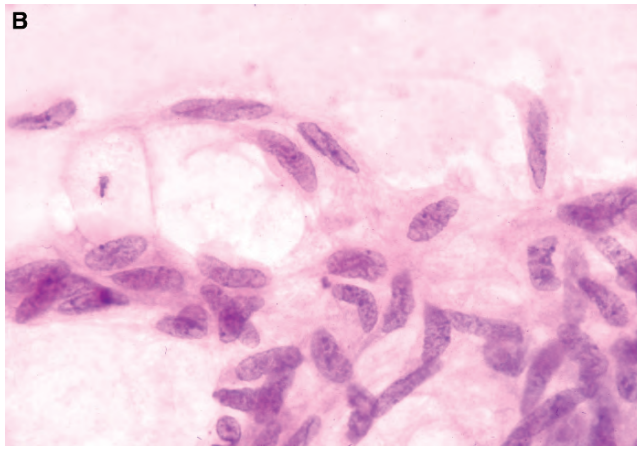


FIG. 11.9. (continued)

The background contains scattered single cells and is often metachromatic, myxoid, and bloody with admixed acute and chronic inflammatory cells. Immunohistochemically, nodular fasciitis is distinguished from myoepithelial-rich tumors and from schwannoma by its reactivity for smooth muscle actin and lack of reactivity for S-100 and keratin.

In contrast to many of the other benign spindle cell lesions, fibromatosis often presents as a poorly circumscribed mass that is sometimes partially fixed to surrounding tissues. Like nodular fasciitis, its clinical features suggest the diagnosis. Approximately 10% of cases occur in the head and neck, often in young individuals. Aspirates consist of uniform polygonal to elongate plump fibroblasts admixed within myxoid and/or collagenous tissue (Fig. 11.10). As opposed to fibrosarcoma, with its mitotically active and at least mildly atypical nuclei, the nuclei in fibromatosis are cytologically bland and lack mitotic activity. Immunohistochemically, the fibroblasts and myofibroblasts comprising the lesion are positive for vimentin, smooth muscle actin, and sometimes desmin. Solitary fibrous tumor is another rare neoplasm that occurs in the head and neck, and occasionally involves the parotid gland. It affects adults in the fourth to seventh decades. FNA of solitary fibrous tumor yields hypercellular cohesive groups of haphazardly

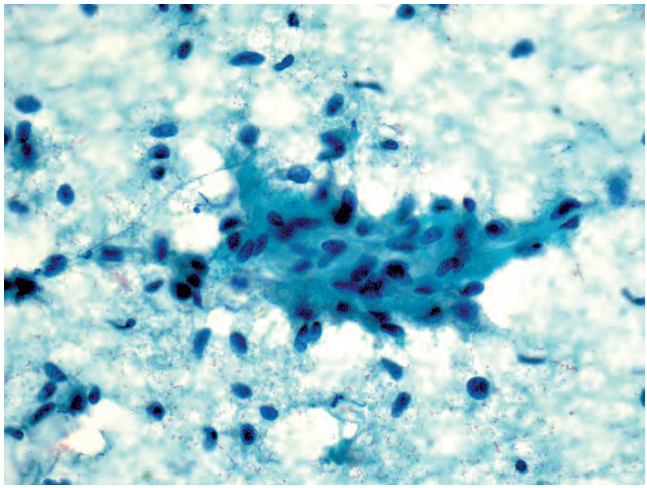


FIG. 11.10. Fibromatosis. Cytologically bland spindled cells in groups with collagenized stroma. (Smear, Papanicolaou.)

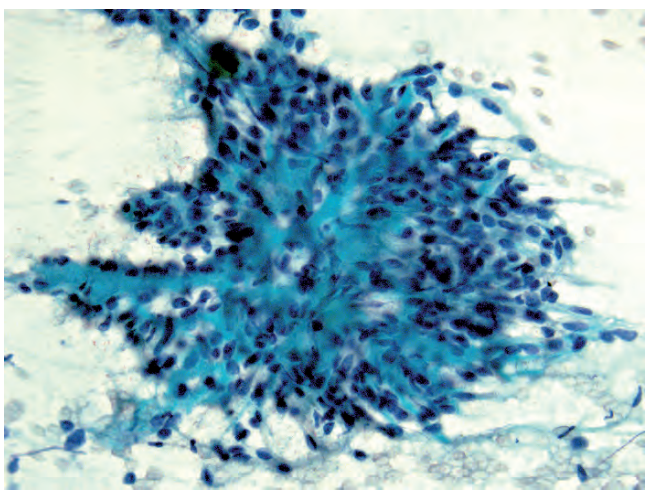


FIG. 11.11. Solitary fibrous tumor. Hypercellular groups of monotonous spindled cells and dense collagen. (Smear, H&E.)

arranged monotonous tapered spindle cells. The background contains scattered single cells and lacks myxoid matrix and inflammatory cells (Fig. 11.11). Fragments of dense ropy collagen, when observed, are characteristic of this tumor. Immunohistochemically, solitary fibrous tumor is distinguished by its reactivity with the nonspecific endothelial marker CD34 and absence of reactivity with S-100 and keratin. Leiomyomas are benign smooth muscle tumors that can occasionally be seen in the subcutaneous tissues of the head and neck and oral cavity. FNA of leiomyomas is often painful. Aspirates exhibit loose fascicular groups of spindle cells with moderate amounts of eosinophilic cytoplasm, and elongate, blunt-ended ("cigar-shaped") nuclei that lack atypia and mitotic activity (Fig. 11.12). The background often contains stripped nuclei. The characteristic immunoprofile of this tumor includes actin and desmin.

In the evaluation of spindle cell aspirates, the greatest impact upon clinical management will depend in part upon distinguishing between benign and malignant. Overall, caution is warranted in the evaluation of spindle cell tumors since malignant features can be subtle. When adequate material is obtained, a definitive diagnosis

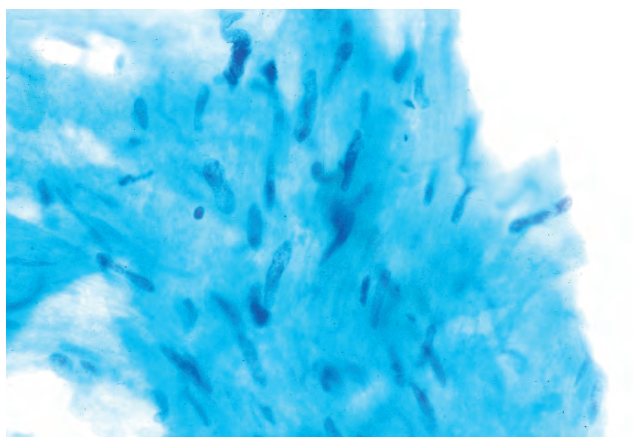


FIG. 11.12. Leiomyoma. Spindle cells have blunt-end "cigar-shaped" nuclei and moderate amounts of eosinophilic cytoplasm. (Smear, Papanicolaou.)

of the most common tumors such as myoepithelial-predominant pleomorphic adenoma, myoepithelioma, and schwannoma can usually be made. Similarly, for metastatic spindle cell tumors, the diagnosis will generally be straightforward. For other spindle cell lesions, however, a descriptive diagnosis will often be used, although it will be possible in most cases to classify the tumor as benign versus malignant. General cytologic features suggestive of a malignant spindle cell tumor include hypercellularity, nuclear pleomorphism, hyperchromasia and irregular chromatin distribution, high mitotic index, atypical mitoses, prominent nucleoli, and background necrosis and hemorrhage. Especially when multiple features are present the likelihood of malignancy is high.

Cytologic Features of Malignant Spindle Cell Tumors

- Hypercellularity
- Nuclear pleomorphism
- Hyperchromasia
- Irregular chromatin distribution
- Prominent nucleoli
- High mitotic index and apoptosis
- Atypical mitoses
- Background necrosis
- Hemorrhage

The most common primary malignant spindle cell tumor of the salivary gland that must be distinguished from other benign myoepithelial-rich tumors is myoepithelial carcinoma (Table 11.2). Myoepithelial carcinoma is a rare salivary gland malignancy, representing less than 2% of all salivary gland carcinomas. Most cases occur in the parotid gland, but the submandibular and minor salivary glands can also be involved. Over half of cases develop within pre-existing pleomorphic adenomas or myoepitheliomas, especially recurrent ones. Aspirates are cellular and comprised of myoepithelial cells that can display any of the various cytomorphologic forms (e.g., spindled, plasmacytoid, epithelioid, stellate, clear) already described for myoepithelioma (Fig. 11.13). A subset of these is predominantly spindled and can resemble sarcoma. A stromal component is sometimes present and can be myxoid or hyaline. The degree of atypia ranges from mild to obviously malignant. Myoepithelial carcinoma

TABLE 11.2. Features of selected malignant head and neck spindle cell lesions.

Malignant Spindle Cell Lesion	Key Cytologic Features	Key Immuno-markers:	Keratin	S-100	Cal-ponin	SM Actin	Neuro-filament	Vimentin	Desmin	EMA	CD99	HMB-45	Leu-7
Myoepithelial carcinoma	Variable atypia; Distinct nucleoli; Background myxoid or hyaline stroma; Necrosis; High mitotic activity		+	+	+	+	−	+	±	+	−	−	−
Spindle cell carcinoma	High-grade nuclear features; Focal squamoid changes; Dense fibrous stroma		+	−	−	±	−	+	−	±	−	−	−
Melanoma	Many single cells; High-grade nuclear features; Distinct nucleoli; Multinucleation; Intranuclear inclusions; Melanin pigment		−	+	−	−	−	+	−	−	−	+	−

TABLE 11.2. (continued).

Malignant Spindle Cell Lesion	Key Cytologic Features	Key Immuno-markers:	Keratin	S-100	Cal-ponin	SM Actin	Neuro-filament	Vimentin	Desmin	EMA	CD99	HMB-45	Leu-7
MPNST	Markedly hypercellular groups; Comma-shaped nuclei; Hyperchromasia; Distinct nucleoli; Mitotically active; Background necrosis		−	+	−	−	−	+	−	−	+	−	+
Synovial sarcoma	Hypercellular; Uniform spindle cells; Mild atypia; Hyperchromasia; Focal glandular or papillary structures; Mitotically active		+	±	±	−	±	+	−	+	+	−	±

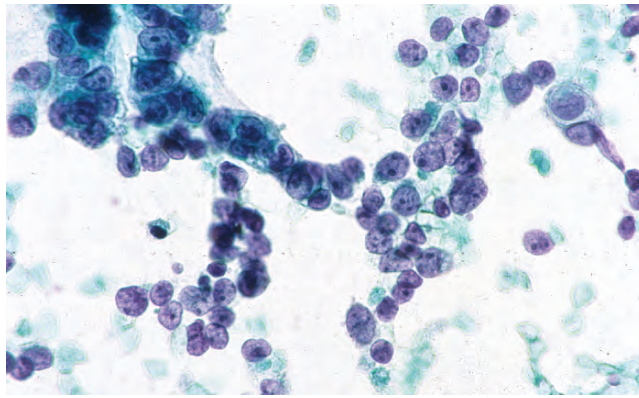


FIG. 11.13. Myoepithelial carcinoma. Plump spindled to plasmacytoid-shaped cells with distinct nucleoli in a loose stroma. (Smear, Papanicolaou.)

is distinguished from its benign counterpart, myoepithelioma, by the presence of a combination of cytologic features, including nuclear pleomorphism, coarse chromatin, prominent nucleoli, background necrosis, and high mitotic activity. In our experience, the finding of distinct nucleoli is among the most commonly present and easily recognized features suggesting the diagnosis.

Among the other malignant spindle cell neoplasms that can be encountered, especially in the parotid gland region, are certain metastatic head and neck tumors, including spindle cell carcinoma and spindled malignant melanoma. In both cases, the patient will usually have a prior history, making the cytologic workup and diagnosis easier. Spindle cell carcinoma (aka sarcomatoid carcinoma) is more common in elderly males, and is particularly seen secondary to radiation therapy. Aspirates are variably cellular and comprised of spindle cells with high-grade nuclear features that resemble those of true high-grade sarcomas such as malignant fibrous histiocytoma (Fig. 11.14). The cells are mitotically active and are usually present in loosely cohesive groups surrounded by a dense fibrous stroma. Immunohistochemical markers can be deceptive since up to 50% of cases will be negative for most cytokeratins and for p63. Therefore, a panel of cytokeratins is recommended to increase the detection of focal keratin positivity.

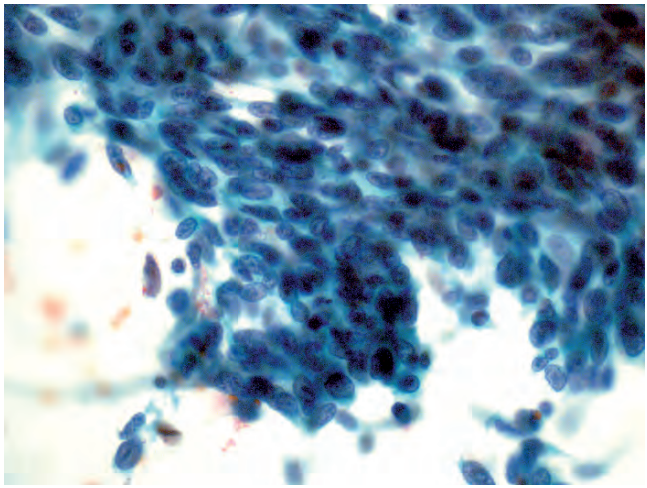


FIG. 11.14. Spindle cell carcinoma. High-grade spindled cells are haphazardly arranged within a fibrous stroma. (Smear, Papanicolaou.)

In addition to being vimentin-positive, some spindle cell carcinomas will express mesenchymal markers, including smooth muscle actin and desmin. Another metastatic tumor, malignant melanoma, can sometimes assume a predominantly spindled cytomorphology (Fig. 11.15). While the high-grade nuclear features resemble those of other malignant spindle cell tumors, malignant melanomas typically exhibit prominent nucleoli, intranuclear pseudoinclusions, and cellular dissociation. An important finding in approximately half of cases is the presence of fine brown melanin pigment granules (Fig. 11.15B) that can be seen within tumor cells as well as within histiocytes.

Sarcomas involving the salivary glands are extremely rare, and the range of different subtypes of sarcoma that can affect the salivary glands either as primary tumors or more commonly by direct extension from surrounding head and neck sites is quite broad – only two will be covered here. As with other spindle cell tumors, ample cytologic material for ancillary studies is essential if there is any hope of making a definitive primary diagnosis. A malignant spindle cell tumor that is important to differentiate from schwannoma

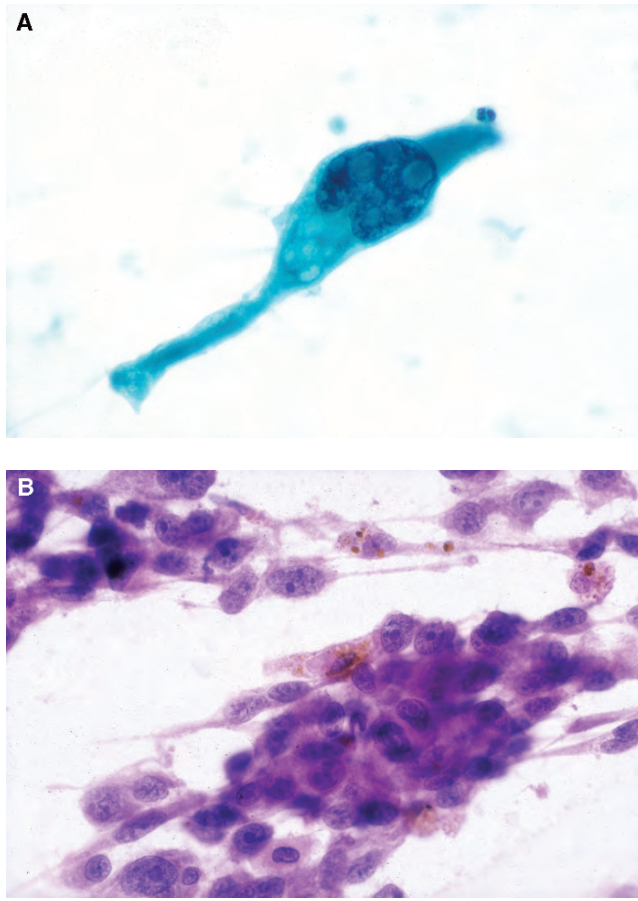


FIG. 11.15. Spindled malignant melanoma. (A) Individual malignant spindled melanoma cell with pleomorphic nucleus and intranuclear inclusions. (B) Loosely cohesive groups of spindled cells with focal finely granular melanin pigment. (Smears, Papanicolaou.)

is malignant peripheral nerve sheath tumor (MPNST). It can occur either sporadically or in association with neurofibromatosis (NF-1), the latter MPNST's being derived from neurofibromas. The microscopic spectrum of MPNST may be more varied than any other soft tissue tumor. Aspirates contain markedly hypercellular fascicular

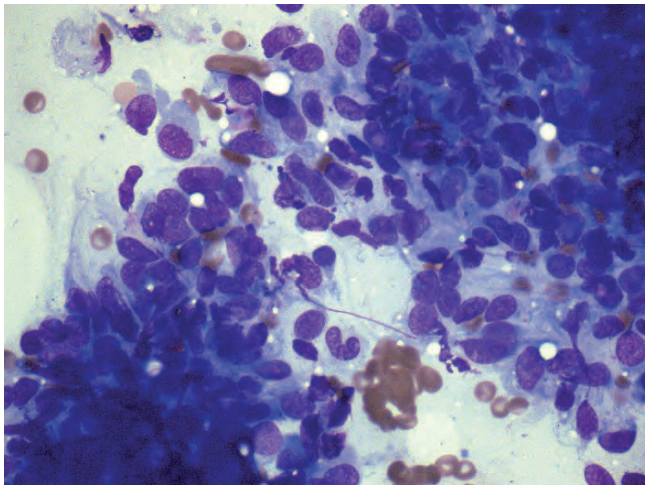


FIG. 11.16. Malignant peripheral nerve sheath tumor. Aspirates contain hypercellular groups of plump spindled cells with hyperchromatic nuclei and clumped chromatin. (Smear, Diff-Quik.)

groups of spindle cells in a background of many scattered single cells, stripped nuclei, and sometimes necrosis (Fig. 11.16). Nuclei are hyperchromatic and range from elongate and comma-shaped, to plump and oval. Cytoplasm is delicate and fibrillar, sometimes with eccentric placement of the nucleus. Occasional large bizarre multinucleate cells can be seen. Both low- and high-grade forms exist, so nuclear atypia is variable, but high-grade forms are more common. In contrast to schwannomas, even low-grade MPNST's show nuclear hyperchromasia, distinct nucleoli, and mitoses. In addition, nuclear palisading is uncommon in MPNST. Immunohistochemical studies also help distinguish MPNST from schwannoma since the former is only weakly and focally positive for S-100. Other positive immunomarkers include vimentin, and leu-7. Immunochemistry and ultrastructural studies are often needed to distinguish high-grade forms of MPNST from other high-grade spindle cell tumors.

Synovial sarcoma is a second example of a sarcoma that can be be a pitfall in the diagnosis of spindle cell tumors. Up to 10% of

synovial sarcomas occur in the head and neck region, where they can involve the salivary glands, usually indirectly. Despite its name, synovial sarcoma is not derived from synovial tissue. It is a mesenchymal malignancy which exhibits variable epithelial differentiation and is genetically identifiable by its characteristic translocation t(X;18), which can be detected using paraffin-embedded cell block material. Patients are usually young adults between the second and fourth decades. Aspirates of synovial sarcoma are hypercellular and consist of a distinctively uniform population of spindle cells (Fig. 11.17). Overall, synovial sarcomas are less pleomorphic than other malignant spindle cell tumors that have been discussed, including MPNST. Nuclei are only mildly atypical. They range from plump and oval to elongate, and the chromatin is granular, and moderately hyperchromatic. Mitoses are easily identified. The cytoplasm is scant, pale, and delicate with tapering ends, and admixed calcifications can sometimes be seen. For biphasic forms of synovial sarcoma, the epithelial component can be subtle, but in well-sampled biphasic cases distinct glandular, sheetlike, or even papillary structures can be identified; monophasic synovial

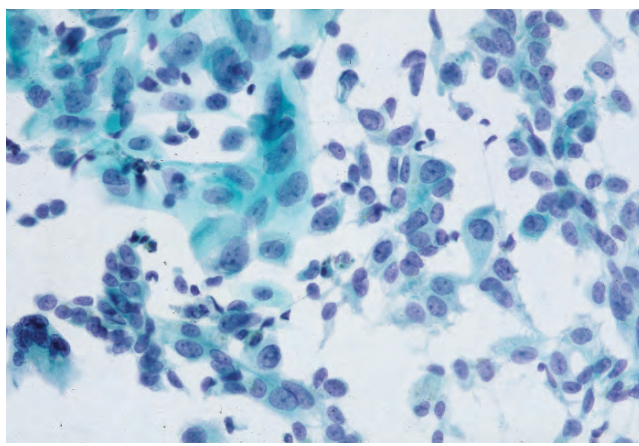


FIG. 11.17. Biphasic synovial sarcoma. Hypercellular aspirate of uniform spindle cells with mild atypia and focal glandular formation. (Smear, Papanicolaou.)

sarcomas lack microscopic features of epithelial differentiation. The immunoprofile of synovial sarcoma includes reactivity with cytokeratins, EMA, CD99, and variable reactivity with S-100 and leu-7. Synovial sarcomas lack the palisading and elongate wavy nuclei of schwannoma, and the immunoprofile is distinct from the strong and diffuse S-100 positivity of schwannoma. Because of its bland cytology and potential dual immunoreactivity for keratin and S-100, it can be misinterpreted as a myoepithelial-predominant pleomorphic adenoma or as a myoepithelioma; however, synovial sarcomas are negative for smooth muscle actin and calponin. Similarly, because of its keratin positivity, care should be taken to distinguish synovial sarcoma from spindle cell carcinoma, which is more pleomorphic and typically arises in patients with a history of treated squamous cell carcinoma.

Ancillary Techniques

More than for any other category of salivary gland tumors, ancillary marker studies, particularly immunohistochemistry, are nearly essential for the evaluation of spindle cell lesions. When possible, a paraffin-embedded cell block should be prepared. Immunocytochemistry can also be performed on cytospins, thin-layer preparations, and even on unstained or destained smears. However, these results may not be as reliable as on paraffin-embedded formalin fixed material. As outlined in Tables 11.1 and 11.2, a panel of immunohistochemical stains that includes basic markers such as S-100, keratin, smooth muscle actin, and calponin is a good starting point that can be further refined, depending upon cytomorphologic characteristics of the aspirated lesion. This approach will work for the most common differential diagnosis of myoepithelial-predominant pleomorphic adenoma, myoepithelioma, and schwannoma. When the differential diagnosis is broadened to include a wider range of benign and malignant spindle cell tumors, then aspirated material should ideally be divided for cell block, cytogenetic studies, and electron microscopy. In our experience, a dedicated FNA pass placed directly into glutaraldehyde fixative for electron microscopy can be extremely useful in the workup of a spindle cell lesion. We have encountered many difficult spindle

cell lesions where immunohistochemistry was not definitive, but ultrastructural features were either diagnostic or at least helped to focus the diagnosis. For selected cases such as synovial sarcoma, where FISH analysis can provide diagnostic information, the paraffin-embedded cell block material can be used.

Probabaly more than for any other category of salivary gland tumors, ancillary marker studies are nearly essential for the evaluation of spindle cell lesions.

Clinical Management and Prognosis

For clinical management purposes, it is usually not critical to differentiate between many of the benign spindle cell tumors that can involve the salivary glands and that have overlapping cytomorphologic features. The clinical management of most will be the same – conservative surgical excision with negative resection margins to prevent local recurrence. Both myoepithelial-predominant pleomorphic adenoma and myoepithelioma are managed in this way. Unfortunately, recurrences can be associated with significant morbidity. They are usually due to positive surgical margins or "spilling" during surgery and are seen in 3%–7% of cases. Even more problematic is the risk of malignant transformation that is seen more often in recurrent cases, and is estimated to represent 5%–10% of cases overall. For schwannoma, the treatment involves total resection of the tumor with sparing of the parent nerve when it can be identified. Recurrences are infrequent, and malignant transformation of schwannoma is considered very rare. Metastatic spindle cell tumors to the salivary glands are usually treated surgically to achieve locoregional control of the disease, and often adjunctive radiotherapy is used, but recurrence rates are high. The clinical management of high-grade sarcomas involving the salivary glands is complex and depends in part upon the specific subtype of tumor and nature of the salivary gland involvement. In general, for primary sarcomas of the salivary gland, treatment will consist of radical resection with negative surgical margins and postoperative radiotherapy. Local

recurrences for primary salivary gland sarcomas range from 40% to more than 60%, and mortality is as high as 64%.

Suggested Reading

Auclair PL, Langloss JM, Weiss SW et al. Sarcomas and sarcomatoid neoplasms of the major salivary gland regions: a clinicopathologic and immunohistochemical study of 67 cases and review of the literature. Cancer 1986;58:1305.

Chhieng DC, Cohen J-M, Cangiarella JF. Fine-needle aspiration of spindle cell and mesenchymal lesions of the salivary glands. Diagn Cytopathol 2000;23:253–259.

Darvishian F, Lin O. Myoepithelial cell-rich neoplasms: cytologic features and benign and malignant lesions. Cancer Cytopathol 2004;102:355–361.

Gerhard R, Fregnani ER, Falzoni R, Siqueira SAC, Vargas PA. Cytologic features of solitary fibrous tumor of the parotid gland: a case report. Acta Cytol 2004;48:402–406.

Kapila K, Mathur S, Verma K. Schwannomas: a pitfall in the diagnosis of pleomorphic adenomas on fine-needle aspiration cytology. Diagn Cytopathol 2002;27:53–59.

Klijianienko J, Caillaud J-M, Lagace R. Cytohistologic correlations in schwannomas (neurilemmomas), including «ancient,» cellular, and epithelioid variants. Diagn Cytopathol 2006;34:517–522.

Kumar PV, Sobhani SA, Ahmed M, Hashemi SB, Eghtadari F, Hamidi SA. Myoepithelioma of the salivary glands: fine needle aspiration biopsy findings. Acta Cytol 2004;48:302–308.

Saad RS, Takei H, Lipscomb J, Ruiz B. Nodular fasciitis of parotid region: a pitfall in the diagnosis of pleomorphic adenomas on fine-needle aspiration cytology. Diagn Cytopathol 2005;33:191–194.

Scheithauer BW, Woodruff JM, Erlandson RA. Atlas of tumor pathology: tumors of the peripheral nervous system. Washington DC: Armed Forces Institute of Pathology, 1997.

Wakely PE, Kneisl JS. Soft tissue aspiration cytopathology: diagnostic accuracy and limitations. Cancer Cytopathol 2000;90:292–298.

12
Clear Cell Neoplasms and Secondary Tumors: Epithelial-Myoepithelial Carcinoma

Background

Salivary gland tumors with clear cell features include a broad range of entities (Fig. 12.1) several of which have already been discussed in other contexts within this book. Epithelial-myoepithelial carcinoma will be the focus of this chapter; it is a rare primary salivary gland tumor that is characterized cytologically by a biphasic pattern of numerous large clear myoepithelial cells admixed with ductal cells. The differential diagnosis of clear cell neoplasms also includes other salivary gland tumors that are comprised predominantly of clear cells such as clear cell carcinoma, sebaceous lymphadenoma, and lipoma, as well as salivary gland tumors that have clear cell variants such as myoepithelioma, myoepithelial carcinoma, oncocytoma, mucoepidermoid carcinoma, and acinic cell carcinoma. Finally, certain metastatic tumors such as renal cell carcinoma and sebaceous carcinoma can also appear clear in FNA samples.

Epithelial-myoepithelial carcinoma is an unusual tumor that represents approximately 1% of all salivary gland neoplasms. Most epithelial-myoepithelial carcinomas are low grade, although a small subset is intermediate to high grade. Between 60% and 80% of epithelial-myoepithelial carcinomas occur in the parotid gland, where they are locally aggressive, but they can also be seen in the palate and other minor salivary gland sites of the oral cavity and upper respiratory tract. In minor salivary gland sites, epithelial-myoepithelial carcinoma can present as an ulcerative nodular lesion. It was first described as a distinct entity by

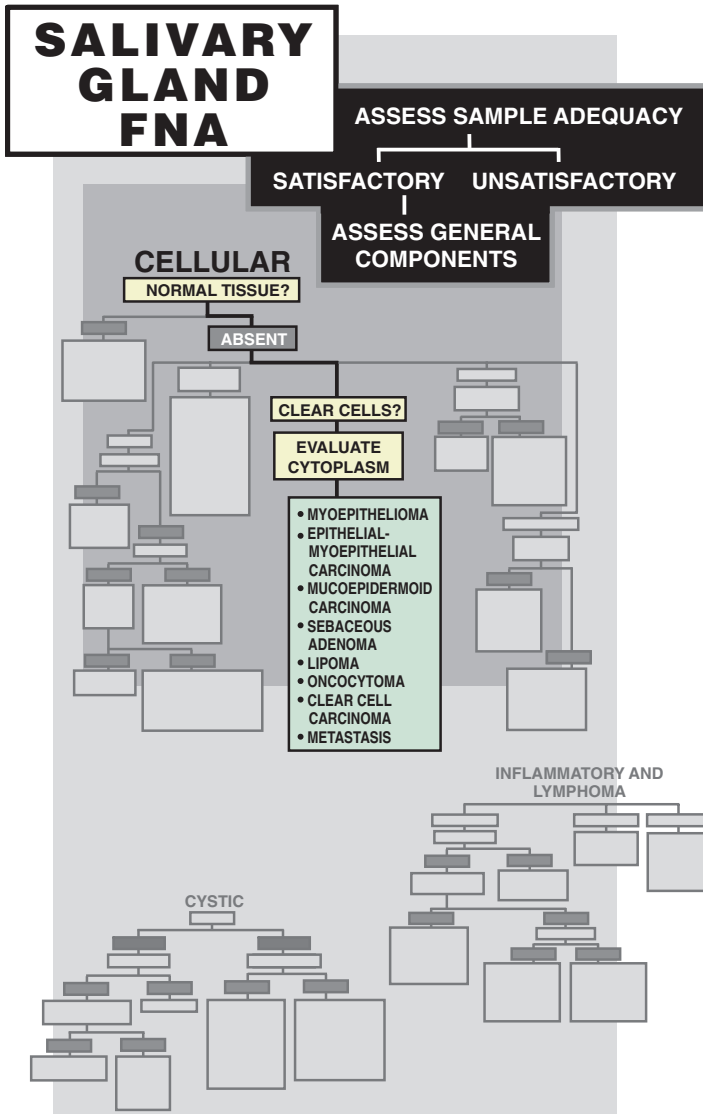


FIG. 12.1. Algorithmic approach to clear cell salivary gland tumors.

Donath in 1972. Epithelial-myoepithelial carcinoma is a tumor of adults, and it occurs in middle-age to elderly individuals. The average age at presentation is 62 years, and in most studies there is an approximately 2:1 female-to-male ratio. In the parotid gland, epithelial-myoepithelial carcinoma typically presents as a well-defined, painless, slowly enlarging mass that in some cases has been present for many years. Despite its relatively bland cytologic appearance, most cases are reported as "malignant" or "suspicious for malignancy" by FNA.

Clinical Features of Epithelial-Myoepithelial Carcinoma

- 1% of all salivary gland malignancies
- Low to intermediate grade
- 60%–80% in the parotid gland
- 62 years, average age at presentation
- 2:1 female-to-male ratio
- Presents as a slowly enlarging, well-defined, painless mass

Secondary tumors of the salivary gland represent approximately 5% of all salivary gland malignancies. Most cases occur in elderly males, with a peak incidence in the seventh to eighth decades. The most common salivary gland affected is the parotid gland, due in large part to the presence of 3 groups of parotid-associated lymph nodes occurring within the parotid fascia, parotid parenchyma, and preauricular region. These 3 groups of parotid lymph nodes drain the external ear, scalp, ipsilateral forehead, and cheek. Lymph nodes in the region of the submandibular gland drain sites in the oral cavity and oropharynx, and thus are most often affected by metastatic disease from these sites. The most common metastatic tumors to the salivary gland are head and neck cutaneous tumors, especially squamous cell carcinoma and malignant melanoma. Basal cell carcinoma can also involve the major salivary glands, most often by direct extension from the overlying dermis. Other metastatic tumors include renal cell, lung (especially small cell carcinoma), and breast carcinoma, as well as nasopharyngeal, colorectal, thyroid, and prostate cancer. As with other lymph nodes within the head and neck, those associated with the parotid gland can also become involved by secondary lymphoproliferative disorders. Interestingly, over 80% of secondary tumors in the parotid

gland are of head and neck origin, while 85% of submandibular gland secondary tumors arise from distant sites. As expected, FNA is highly sensitive and specific for the diagnosis of secondary tumors of the salivary glands.

Clinical Features of Secondary Tumors of the Salivary Glands

- 5% of all salivary gland malignancies
- Males in seventh to eighth decade
- Parotid gland
- Most common cutaneous primaries and metastases from regional and distant sites:
  - Squamous cell carcinoma
  - Malignant melanoma
  - Basal cell carcinoma
  - Sebaceous carcinoma
  - Lung carcinoma (especially small cell carcinoma)
  - Renal cell carcinoma
  - Breast carcinoma
  - Lymphoma
  - Thyroid carcinoma

General Diagnostic Approach

A salivary gland aspirate comprised of a predominance of cells with clear cytoplasm should be evaluated with caution since the differential diagnosis is broad. Clinicoradiologic information regarding the anatomic site, tumor size, and rate of growth of the lesion, as well as any associated clinical findings, can be helpful for limiting the differential diagnosis. Careful assessment of the cytomorphologic features will often be able to distinguish benign clear cell tumors from malignant ones. This is among the more critical aspects of cytologic evaluation due to its impact on clinical management. In the evaluation of an aspirate containing clear cells, material should be obtained, if possible, for a cell block for application of ancillary marker studies. In particular, studies directed at analyzing the cytoplasm for glycogen, mitochondria, mucin, or lipid can be very useful. Any clinical history of a primary malignant neoplasm outside of the salivary glands or an

unusual microscopic appearance that is distinct from other primary salivary gland tumors should prompt consideration of a secondary salivary gland neoplasm. The majority of secondary salivary gland tumors will involve salivary gland-associated lymph nodes, and will exhibit high-grade cytologic features, including marked nuclear atypia, mitotic activity, and necrosis.

Diagnostic Criteria

Epithelial-Myoepithelial Carcinoma

Aspirates of epithelial-myoepithelial carcinoma are cellular, and the key to the diagnosis is adequate sampling so that the biphasic pattern of the lesion can be appreciated. Cytologically, epithelial-myoepithelial carcinoma consists of a predominant population of polygonal myoepithelial cells that have abundant, clear, glycogen-rich cytoplasm that is very delicate (Figs. 12.2–12.5). The clear cells form loosely cohesive sheets and spheres. Occasionally, spherical groups of cells will be outlined by a thin peripheral band

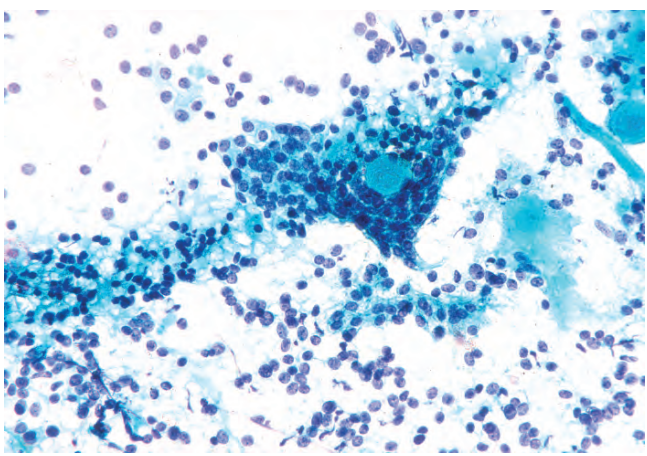


FIG. 12.2. Epithelial-myoepithelial carcinoma. At low-magnification, occasional sheets of cells are present surrounded by numerous background strippped nuclei and focal acellular material. (Smear, Papanicolaou.)

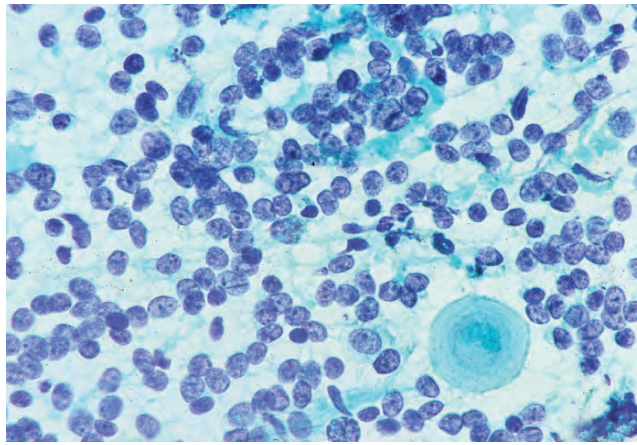


FIG. 12.3. Epithelial-myoepithelial carcinoma. The cells are predominantly myoepithelial and have abundant delicate clear cytoplasm. Many stripped nuclei are also present as well as focal concentrically laminated acellular material. (Smear, Papanicolaou.)

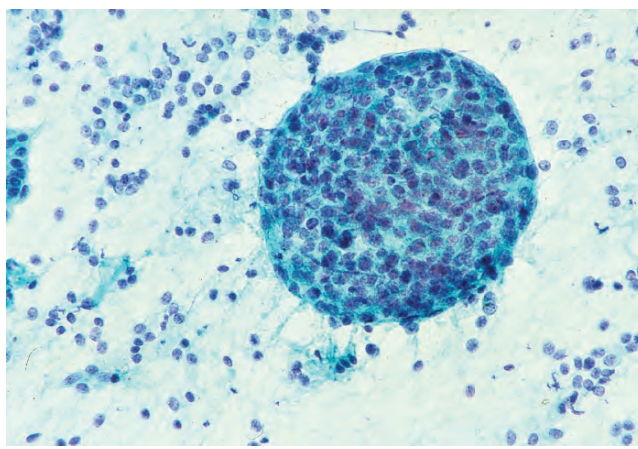


FIG. 12.4. Epithelial-myoepithelial carcinoma. Occasional cohesive spheres of cells are present. (Smear, Papanicolaou.)

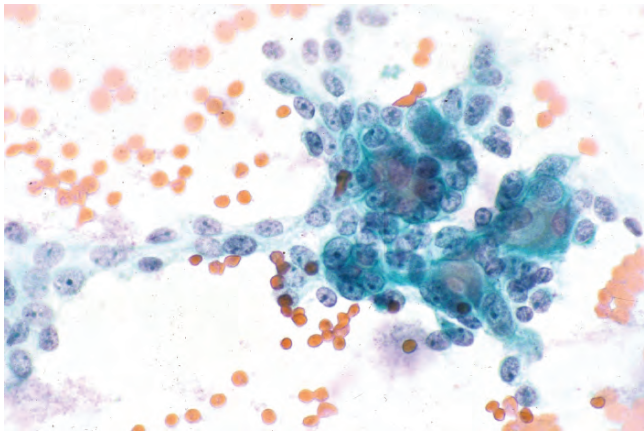


FIG. 12.5. Epithelial-myoepithelial carcinoma. The presence of occasional groups of biphasic cells consisting of central low-cuboidal ductal cells surrounded by clear myoepithelial cells is characteristic. (Smear, Papanicolaou.)

of basement membrane-like material. Nuclear atypia is mild, with most cells having uniform, enlarged oval nuclei, dispersed chromatin, and small distinct nucleoli. A second minor population of intercalated duct-type epithelial cells is also present. These cells are cuboidal, with a high N:C ratio, large round to oval nuclei, distinct nucleolus, and scant dense finely granular cytoplasm. Mitotic activity, apoptosis, and necrosis are uncommon. A small to moderate amount of secreted acellular, concentrically laminated eosinophilic material is present between groups of cells and forms spheres within the background. The background also contains many stripped myoepithelial nuclei as well as single myoepithelial cells. Fragments of fibrous tissue or dense hyalinized stroma can sometimes be seen. In some aspirates of epithelial-myoepithelial carcinoma, the clear myoepithelial cells predominate almost to the exclusion of the ductal cells, and it is this subset that is the most challenging to diagnose. If material is available for making a cell block, immunohistochemical stains for cytokeratins and myoepithelial markers can be performed which will highlight the characteristic biphasic nature of the tumor (Fig. 12.6).

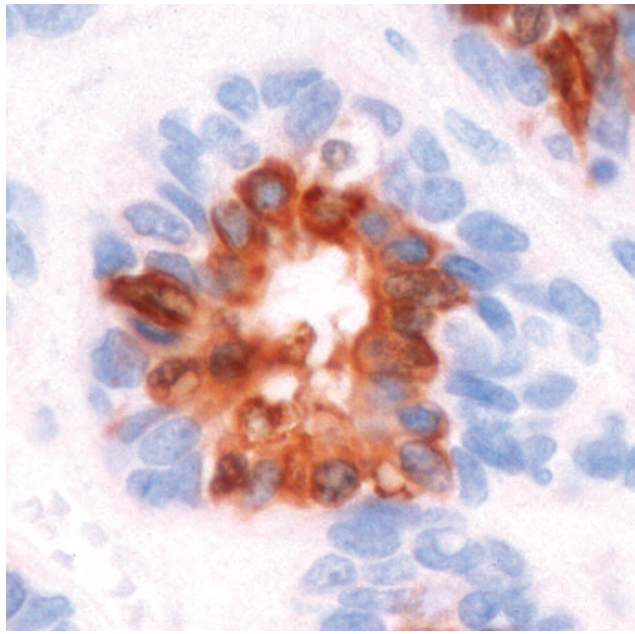


FIG. 12.6. Epithelial-myoepithelial carcinoma. The biphasic pattern is highlighted using an immunohistochemical stain for keratin. (Cell block, immunohistochemical stain.)

The key to the diagnosis of epithelial-myoepithelial carcinoma is identification of the biphasic pattern of the lesion.

Cytologic Features of Epithelial-Myoepithelial Carcinoma

- Hypercellular
- Sheets and 3-dimensional groups of biphasic cells:
  - Predominant population of large clear myoepithelial cells
  - Minor population of ductal cells with scant cytoplasm
- Mild nuclear atypia
- Oval nucleus with open chromatin and distinct nucleolus (myoepithelial cells)
- Spheres of secreted acellular laminated material

- Background stripped myoepithelial cell nuclei
- Biphasic nature of lesion highlighted using immunostains for low molecular weight keratin or myoepithelial markers

Metastatic Disease

As with aspirates from other anatomic sites, the possibility of a metastatic tumor should be considered whenever the patient has a history of nonsalivary gland cancer. This is especially true when the cytologic features of the malignant cells do not match those of other common primary salivary gland neoplasms. The metastases will usually retain many of the cytologic features of the primary tumor, and because of this, one of the best ways to confirm a secondary tumor is direct microscopic comparison with the cytologic features of the primary. Aspirates of secondary tumors are usually hypercellular and comprised of cells with malignant-appearing nuclear features. Since a majority of secondary salivary gland tumors involve parotid gland-associated lymph nodes, there will often, but not always, be a mixed population of lymphocytes, tingible body macrophages, and lymphohistiocytic aggregates in the background. In addition, a tumor diathesis can sometimes be seen.

One of the best ways to confirm a secondary tumor is direct microscopic comparison with the cytologic features seen in the primary.

General Cytologic Features of Metastatic Disease

- Hypercellular
- Malignant nuclear features
- Background tumor diathesis
- Admixed lymphocytes and lymphohistiocytic aggregates (lymph node involvement)

The most common secondary tumor of the salivary gland is metastatic head and neck squamous cell carcinoma usually, of cutaneous origin (Fig. 12.7). In many cases, the tumor is comprised of keratinizing cells with dark angulated nuclei, although high-grade

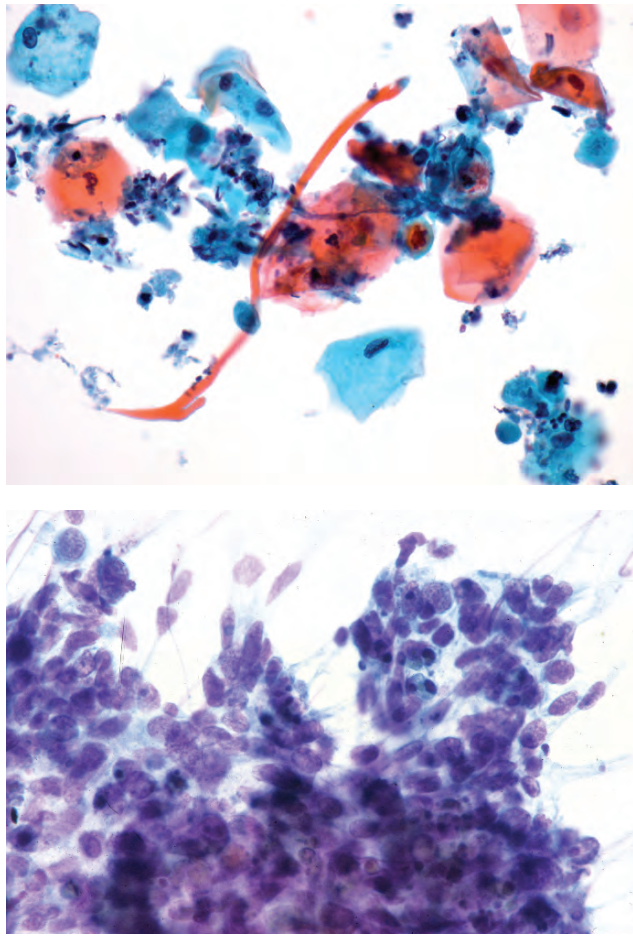


FIG. 12.7. Metastatic squamous cell carcinoma. (A) Well-differentiated keratinizing squamous cells with hyperchromatic nuclei; (B) Crowded group of non-keratinizing squamous cell carcinoma with marked atypia and basaloid features. (Smears, Papanicolaou.)

or basaloid forms, especially from noncutaneous sites, can be nonkeratinizing. In addition, metastatic squamous cell carcinomas will often be cystic (see Chapter 9). While the presence of squamous cells raises the possibility of mucoepidermoid carcinoma,

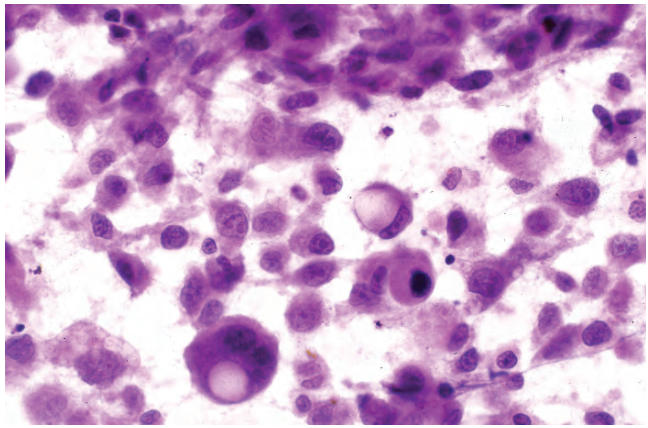


FIG. 12.8. Metastatic malignant melanoma. A dispersed population of single cells with distinct nucleoli and intranuclear inclusions is characteristic. (Smear, modified H&E.)

keratinization is not a feature of the latter and intracellular mucin would be absent. Malignant melanoma can have a very wide range of cytologic appearances, but most aspirates will consist of a dispersed, single cell pattern of pleomorphic cells (Fig. 12.8). This pattern would be unusual for a primary salivary gland tumor. Prominent nucleoli and intranuclear pseudoinclusions are typically encountered in melanomas, and a very helpful feature when present is the finely granular melanin pigment that can be found within the cytoplasm of the malignant cells and within histiocytes (Fig. 12.9). Immunohistochemical stains for S-100, HMB-45, and Mart-1 can be used to confirm the diagnosis. Basal cell carcinoma can involve the salivary gland through direct extension from an overlying cutaneous site (Fig. 12.10). In the absence of clinical history, which is crucial to avoiding this pitfall, cutaneous basal cell carcinoma can be difficult to distinguish from primary basal cell tumors and from the solid form of adenoid cystic carcinoma (See Chapter 7). Basal cell carcinomas often exhibit peripheral palisading not present in adenoid cystic carcinoma and lack the characteristic peripheral bands and intercellular droplets of matrix, and the two populations of cells seen in primary basal cell tumors. Metastatic renal cell carcinoma can present a significant diagnostic challenge in salivary

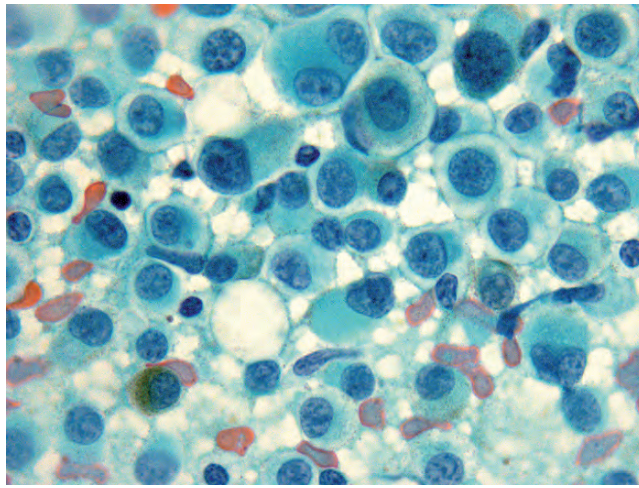


FIG. 12.9. Metastatic malignant melanoma. The malignant cells are pleomorphic and contain small fine granules of melanin pigment within the cytoplasm of tumor cells. (Smear, Papanicolaou.)

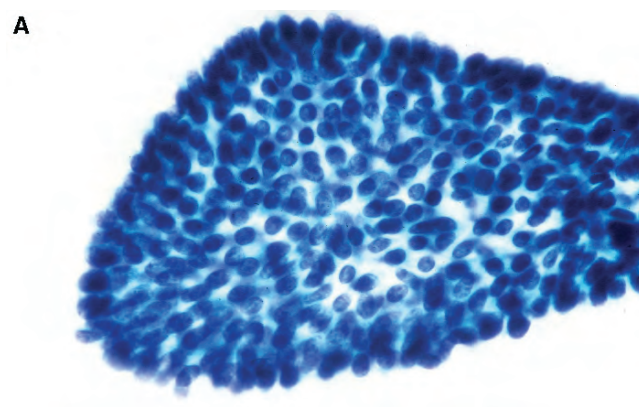


FIG. 12.10. Cutaneous basal cell carcinoma. (A and B) The cytoarchitecture and nuclear features overlap with those of primary basal tumors of the salivary gland. (Smears, Papanicolaou.)

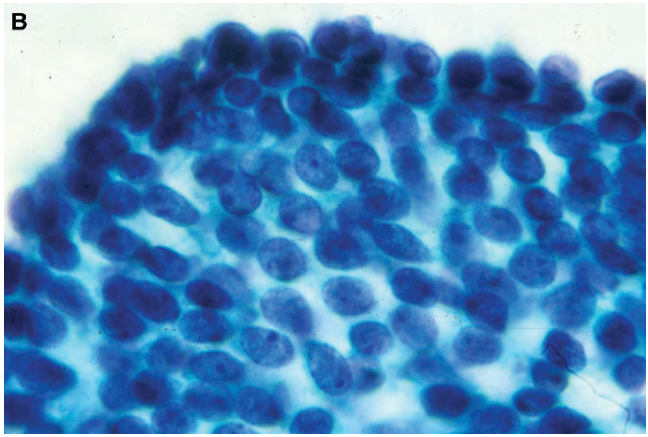


FIG. 12.10. (continued)

gland aspirates since it can mimic a primary salivary gland tumor, particularly those with oncocytic or clear cell features (Fig. 12.11). Aspirates of metastatic renal cell carcinoma are characterized by cells with abundant granular and vacuolated cytoplasm forming loosely cohesive groups. The nuclei are round and eccentrically placed, and nucleoli are prominent. Immunohistochemical studies using CD10, renal cell carcinoma marker, and high molecular weight cytokeratin can be used to confirm the diagnosis; a colloidal iron stain should be applied for oncocytic cases where the diagnosis of chromophobe renal cell carcinoma is suspected.

Differential Diagnosis and Pitfalls

The spherical groups and acellular proteinaceous material forming large extracellular globules in epithelial-myoepithelial carcinoma raise the possibility of adenoid cystic carcinoma (Fig. 12.12). However, in contrast to adenoid cystic carcinoma, which is primarily a basaloid tumor (high N:C ratio and dark angular nuclei), epithelial-myoepithelial carcinoma contains a predominance of large clear myoepithelial cells with oval vesicular nuclei. The secretory material of epithelial-myoepithelial carcinoma is laminated, a feature not

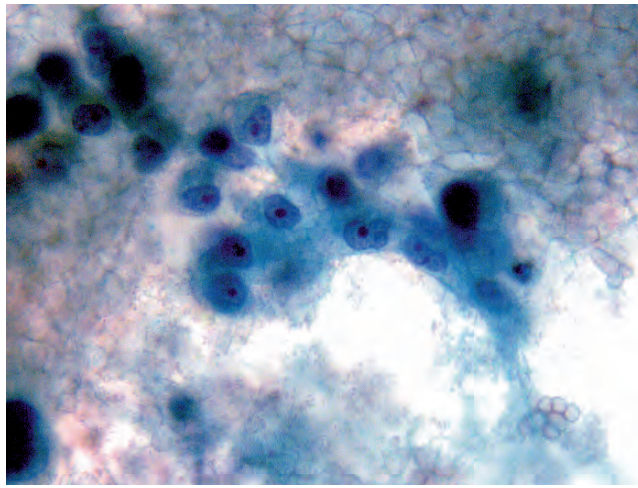


FIG. 12.11. Metastatic renal cell carcinoma to the salivary gland. Cells have abundant delicate vacuolated cytoplasm and an eccentric nucleus with distinct nucleolus; they can mimic other primary oncocytic and clear cell tumors of the salivary gland. (Smear, H&E.)

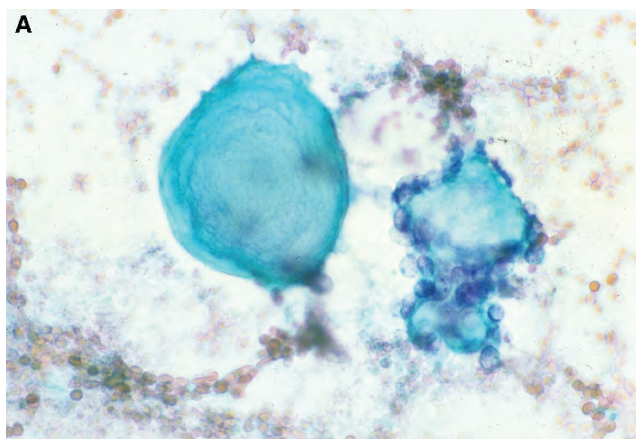


FIG. 12.12. The extracellular secretions of (A) epithelial-myoepithelial carcinoma can mimic features of (B) adenoid cystic carcinoma. (A, Smear, Papanicolaou; B,smear, Diff-Quik.)

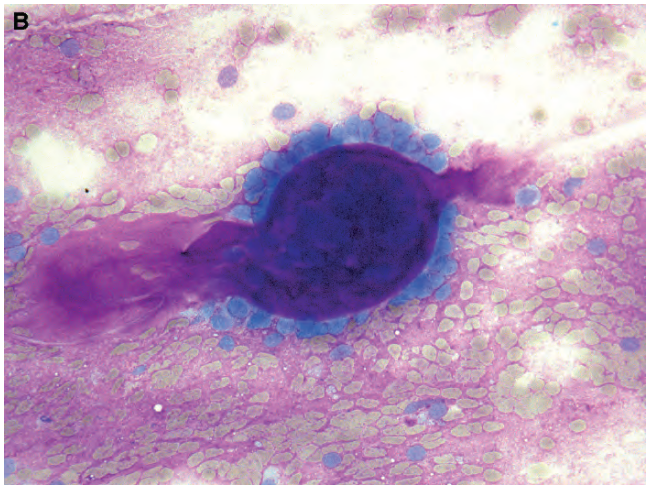


FIG. 12.12. (continued)

seen in the matrix material of adenoid cystic carcinoma. Since the predominant cell-type in epithelial-myoepithelial carcinoma is the myoepithelial cell, it is reasonable that myoepithelioma and myoepithelial carcinoma should also be considered in the differential diagnosis. Most importantly in distinguishing these, myoepitheliomas lack the biphasic pattern of cells seen in epithelial-myoepithelial carcinoma. Also, the neoplastic cells of epithelial-myoepithelial carcinoma are much larger due to their abundant glycogen-rich cytoplasm. In addition to myoepithelial carcinoma lacking a biphasic pattern, its cells are smaller and more atypical than those of epithelial-myoepithelial carcinoma. Clear cell carcinoma of the salivary gland (aka hyalinizing clear cell carcinoma) is is a low-grade malignancy that occurs primarily in the minor salivary glands of the oral cavity, especially the palate. While the cells show minimal atypia and contain pale glycogen-rich cytoplasm similar to epithelial-myoepithelial carcinoma, they lack evidence of either ductal or myoepithelial differentiation (Fig. 12.13). The latter is best confirmed using immunohistochemical stains on cell block material.

The differential diagnosis of clear cell tumors of the salivary gland is broad and includes both primary and secondary tumors. Distinction between the various clear cell tumors is challenging

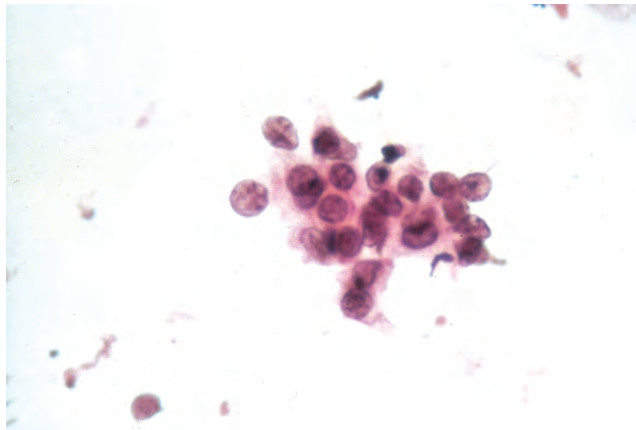


FIG. 12.13. Clear cell carcinoma. The cells form crowded groups and have oval bland nuclei and pale delicate cytoplasm. This is a diagnosis of exclusion. (Thin-layer preparation, Papanicolaou.)

and depends upon assessing the presence of and degree of nuclear atypia (e.g., myoepithelial carcinoma), detection of a secondary cell population admixed with the clear cells (e.g., mucoepidermoid carcinoma and epithelial-myoepithelial carcinoma), and a careful search for areas of the tumor without the clear cell features (e.g., acinic cell carcinoma). In the cytologic evaluation of clear cell tumors, a useful strategy is to define the nature of the clear cytoplasm, which can be due to any of a number of different causes, including mitochondrial condensation, glycogen, mucin, or fat. Useful ancillary studies include cell block material for histochemical and immunohistochemical tests and electron microscopy. In a subset of cases, oncocytomas can appear predominantly clear secondary to mitochondrial condensation, and occasionally low-grade mucoepidermoid carcinomas will contain mostly clear cells. The latter is due to abundant cytoplasmic glycogen, but small amounts of mucin can also be detected. Acinic cell carcinoma is another example of a primary salivary gland tumor that is not traditionally clear, but in approximately 6% of cases can contain clear cells. Sebaceous adenoma and sebaceous lymphadenoma (Fig. 12.14) are very rare benign salivary gland tumors containing cytologi-

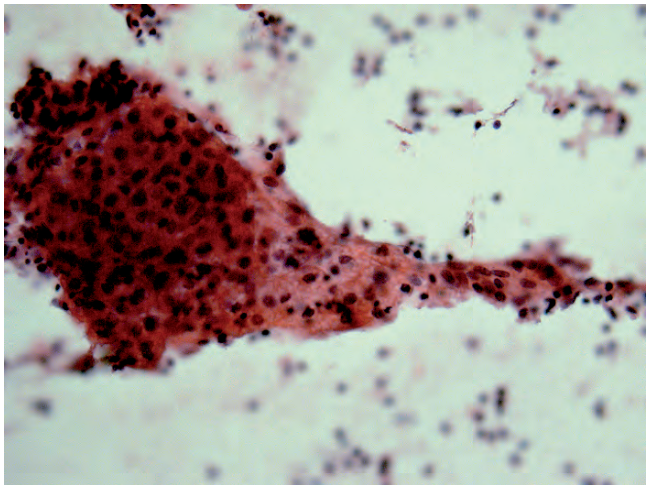


FIG. 12.14. Sebaceous lymphadenoma. The cells are present in a sheetlike arrangement of cytologically bland cells with abundant vacuolated cytoplasm and scattered background lymphocytes. (Smear, modified H&E.)

cally bland, vacuolated clear cells with fat. Similarly, both primary and metastatic sebaceous carcinomas are comprised of clear cells containing cytoplasmic lipid (Fig. 12.15A); air-dried preparations can be used to perform an oil red-o stain to demonstrate cytoplasmic lipid (Fig. 12.15B). The tumor is rare, and is considered an intermediate- to high-grade malignancy.

In the cytologic evaluation of clear cell tumors, a useful strategy is to define the nature of the clear cytoplasm.

Differential Diagnosis of Clear Cell Tumors

- Epithelial-myoepithelial carcinoma
- Myoepithelioma
- Myoepithelial carcinoma
- Oncocytoma
- Mucoepidermoid carcinoma
- Acinic cell carcinoma

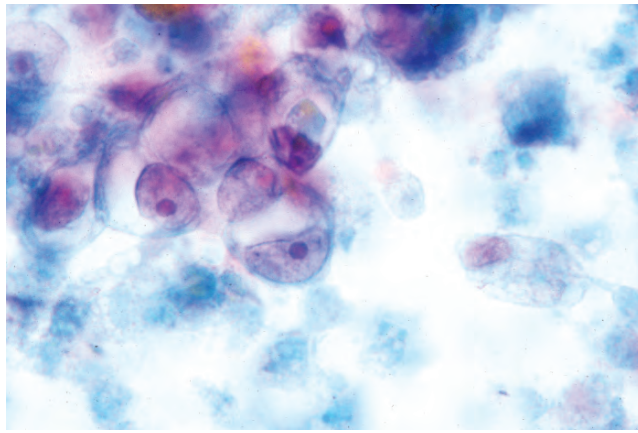


FIG. 12.15. Sebaceous carcinoma. Markedly atypical cells with enlarged oval nuclei, prominent nucleoli, and abundant clear cytoplasm. (Smear, Papanicolaou.)

- Clear cell carcinoma
- Sebaceous lymphadenoma
- Sebaceous carcinoma
- Lipoma
- Metastatic renal cell carcinoma

Ancillary Techniques

If material is available for making a cell block or alternatively using cytologic preparations (e.g., cytospins, thin-layer preparation), immunohistochemical stains can demonstrate the biphasic pattern of epithelial-myoepithelial carcinoma. The inner (luminal) ductal cells are immunohistochemically positive for wide-spectrum keratin, low molecular weight keratin, and EMA, while the outer (abluminal) myoepithelial cells are immunoreactive for smooth muscle actin, calponin, p63, and S-100. A PAS stain can also be used to demonstrate the abundant glycogenated cytoplasm which is sensitive to diastase treatment. As mentioned previously, when assessing an aspirate of a clear cell tumor, it is often helpful to apply histochemical stains to determine the nature of the clear

TABLE 12.1. Useful histochemical stains for evaluating clear cell tumors.

Histochemical Stain	Target	Tumor
Oil red-o	Lipid	Sebaceous adenoma, Sebaceous carcinoma, Lipoma
PAS	Glycogen	Myoepithelioma, Epithelial-myoepithelial carcinoma
PAS + Diastase	Zymogen granules	Acinic cell carcinoma
PTAH	Mitochondria	Oncocytoma Warthin tumor
Mucicarmine	Mucin	Mucoepidermoid carcinoma
Alcian Blue	Mucin	Mucoepidermoid carcinoma

cytoplasm (Table 12.1). Unfortunately, there are no specific markers for salivary gland primary tumors; therefore, immunohistochemical panels and histochemical stains need to be individually geared towards specific questions of differential diagnosis. This is especially true when the differential diagnosis includes the possibility of metastatic disease. Since a majority of patients with a secondary tumor of the salivary gland will have a history of malignancy, a focused immunocytochemical panel can be performed. Represenative immunostains would include TTF-1 for lung carcinoma, thyroglobulin (and TTF-1) for thyroid carcinoma, S-100, HMB-45, and MART-1 for aspirates suspicious for malignant melanoma, CK20 for colorectal carcinoma, RCC and CD10 for renal cell carcinoma, and CD45, CD20, and light chain markers for B-cell lymphoma (Table 12.2).

Clinical Management and Prognosis

The prognosis for epithelial-myoepithelial carcinoma is excellent. Clinical management usually involves surgical resection with negative margins. Local recurrences are seen in approximately 30%–40% of cases (range: 23%–80%), and can be seen up to 3 decades after initial treatment. The rate of metastatic disease is low, ranging from 5%–25%. Deaths from epithelial-myoepithelial carcinoma are infrequent, with a reported range from 0–40%; the 10-year survival rate is over 80%. The most important factors predictive of aggressive behavior include tumor size, margin status,

TABLE 12.2. Selected immunohistochemical stains for evaluating secondary tumors.

Tumor	Immunohistochemical Stain
Malignant melanoma	S-100, HMB-45, MART-1 (A103)
Renal cell carcinoma	CD10, renal cell carcinoma marker, vimentin
Lung	TTF-1; Synaptophysin+ and keratin 20 – for small cell carcinoma
Breast	GCDFP-15, ER, PR
Thyroid	Thyroglobulin, TTF-1
Colorectal	CK20, CDX-2
B-cell lymphoma	CD45, CD20, kappa and lambda light chains

angiolymphatic invasion, tumor necrosis, and myoepithelial anaplasia. Most secondary tumors involving the salivary glands are treated by combined modalities, including surgical excision to control the tumor locally, consideration of concurrent neck dissection, and postsurgical radiation therapy. Loco-regional rates of recurrence, however, are high. The overall average 5-year survival rate for all secondary tumors is 60%–79%. The 5-year survival rate for metastatic head and neck squamous cell carcinoma is 48%, and for malignant melanoma it is 57%. For secondary tumors from infraclavicular sites, the prognosis is usually poor, especially when the salivary gland represents only one of multiple sites. Overall, clinical management and prognosis vary depending upon the specific type of tumor and extent of disease.

Suggested Reading

Batsakis JG, Bautina E. Metastases to major salivary glands. Ann Otol Rhinol Laryngol 1990;99:501.

Layfield LJ, Glasgow BJ, Goldstein N, Lufkin R. Lipomatous lesions of the parotid gland: potential pitfalls in fine needle aspiration biopsy diagnosis. Acta Cytol 1991;35:553–556.

Milchgrub S, Gnepp DR, Vuitch F, Delgado R, Albores-Saavedra J. Hyalinizing clear cell carcinoma of salivary gland. Am J Surg Pathol 1994;18:74–82.

Nuyens M, Schupbach J, Stauffer E, Zbaren P. Metastatic disease to the parotid gland. Otolaryngol Head Neck Surg 2006;135:844–848.

Saqi A, Giorgadze T, Eleazar J, Remotti F, Vazquez M. Clear cell and eosinophilic oncocytomas of salivary gland: cytological variants or parallels? Diagn Cytopathol 2007;35:158–163.

Schroeder WA, Stahr WD. Malignant neoplastic disease of the parotid lymph nodes. Laryngoscope 1998;108:1514–1519.

Seethala R, Barnes L, Hunt JL. Epithelial-myoepithelial carcinoma: a review of the clinicopathologic spectrum and immunophenotypic characteristics in 61 tumors of the salivary glands and upper aerodigestive tract. Am J Surg Pathol 2007;31:44–57.

Wang B, Brandwein M, Gordon R, Robinson R, Urken M, Zarbo RJ. Primary salivary clear cell tumors – A diagnostic approach. Arch Pathol Lab Med 2002;126:676–685.

Index

Page numbers followed by f indicate figures; t, tables.

N