Frozen Section Pathology

Diagnostic Challenges

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Preface

Multidisciplinary care teams and continuing medical education programs are largely subspecialty focused in the modern era. Although a subspecialty structure dominates the practice of surgical pathology at many institutions, intraoperative consultation often remains a general rotation with a somewhat unpredictable nature. However, the nature of the procedure, goal of the frozen section, and impact of tumor staging on management are all important and subspecialty dependent. Naturally, surgeons develop relationships with pathologists acquainted with their subspecialties, but may find themselves interacting with pathologists outside their areas of expertise in the frozen section laboratory. In this situation, a lack of rapport combined with an incomplete understanding of the situational needs on the part of the pathologist can lead to lapses in communication that influence immediate patient care. The editors of this book have first-hand understanding of the challenges faced by subspecialists who find themselves faced with difficult frozen sections outside their areas of expertise. The goal of this book is to provide guidance regarding the approach to common scenarios encountered in the frozen section laboratory while underscoring diagnostic pitfalls and providing the proper level of diagnostic information to ensure clear communication. The book is organized according to organ system with additional chapters discussing the roles of digital pathology and molecular assays. Each chapter is extensively illustrated to highlight key points that facilitate interpretation and highlight areas for potential error. We hope that trainees understand the need for mastery of this unique diagnostic tool, and that pathologists who cover frozen section can convert practical information provided into diagnostic improvements in this essential activity.

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Technique of Intraoperative Consultation

1

Alain C. Borczuk

Introduction

Intraoperative consultation represents a workflow that allows for real-time macroscopic and, more often, microscopic evaluation of tissue specimens during surgical procedures. Its role is to provide guidance that might influence the surgical procedure or, in some instances, a rapid diagnosis to expedite the administration of a therapy [1]. The reasons for a frozen section can vary and at times dynamically so; an unexpected finding can shift the focus from a staging or margin assessment to a diagnostic question. As a result, an understanding of the procedure and its purpose, coupled with the specifics of the particular sample, is critical. Surgical pathology has become increasingly subspecialized, but the realities of practice in many pathology departments require a team effort for frozen section coverage that includes maintaining competency in areas of general pathology outside of the individual subspecialty of the pathologist. This is made more complicated by the fact that surgery is subspecialized, and the pathologist covering the procedure may be known to a different subspecialty team. As a result, the surgeon operating may not be familiar with the covering pathologist, and this can at times impact communication. Understanding the clinical scenarios and knowing the pitfalls help to bolster confidence and improve communication. The goal of this book is to present the uses of intraoperative consultation across different specialty areas, to help the diagnostic pathologist negotiate what has largely remained an exercise of general pathology in an increasing subspecialized medical world.

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Practical Approach to Frozen Section

While the indications for a frozen section are quite varied, it is useful to divide them into main categories. Among the most frequent ones performed is for staging of cancer. This can include parameters that affect TNM staging, such as nodal station sampling, and R-factor parameters related to margin assessment. Margin assessment also encompasses macroscopic examination. Frozen section impacts the primary diagnosis, and, thus, recognition of neoplastic and non-neoplastic conditions requires maintenance of proficiency in many areas of a variety of organ systems. This includes the "surprise" frozen section, such as a nodule or abnormality discovered during a procedure in which little preoperative evaluation has been performed. Frozen sections may be requested to identify specific tissue types, such as parathyroid tissue in a patient with hypercalcemia or undergoing thyroid surgery.

The history of frozen section evaluation demonstrates the origin of its importance in surgical procedures [2]. As surgery became more sophisticated, the range of diseases treated became more complex. This was especially true in the treatment of cancer, and the recognition of cancer in real-time during surgery gained importance. Unfortunately, techniques that allowed tissue sectioning and staining were either too slow or insufficiently detailed to facilitate real-time intraoperative consultations. However, emergence of tissue freezing and sectioning techniques in combination with the application of optimal vital dyes led to discovery of effective intraoperative methods for tissue evaluation [3]. These methods gained wide acceptance and are now essential to many operations.

The steps of intraoperative consultation for the pathology laboratory begin with preparation [4–6]. Since the majority of surgical procedures are scheduled, it is possible to generate a list of cases with potential frozen sections, based upon procedure type. At a minimum, the electronic medical record can be leveraged to determine prior imaging and in-house biopsy pathology or alternatively a clinical history of prior disease, including malignancy. In a setting where whole digital slide imaging is available, the prior pathology may be accessed electronically; however, the file archive may also be interrogated with relevant glass slides retrieved. Another preparatory step involves the confirmation that real-time systems of slide review, such as robotic microscopes, are operational in laboratories that have remote coverage.

Preparation also focuses on laboratory readiness. The microtome-cryostats should be examined for temperature and its blades replaced. Materials used for frozen sections should be restocked. Fixatives, staining reagents, mounting medium, slides, and coverslips need to be replaced or restocked as appropriate. These efforts assure that when the tissue examination request comes through, the turn-around time is minimized.

Communicating the need for frozen section and specimen transport to pathology are also important considerations. Some facilities have the frozen section room close to the operating room, but not necessarily near the pathology department. The logistics of assembling the team of pathologist and/or technician, pathologist

assistant, and resident can be a challenge and efforts should be made such that personnel issues do not impact turn-around time. Here again, leveraging remote viewing technology can simplify some steps, but these considerations may be site-specific. The planning of frozen section capability during hospital expansions of surgical suites is essential.

The pathology laboratory must have a method for accessioning and labeling frozen section specimens upon their receipt, as these are often the first materials received for the particular patient on a given day. The specimen part needs a unique identifier, and sequential labeling of additional specimens has to be maintained, usually as assigned by the laboratory information system. This information, along with the requisition, must be then shared with the pathology team.

The pathologist is then presented with a macroscopic examination and a diagnostic question. In some instances, the question is either explicitly stated on the requisition or implied by nature of the procedure. In other circumstances, interrogation of the electronic medical record makes it clear what is required. Macroscopic examination is critical, especially when clarification from the surgeon, either by phone or in-person evaluation, is required. Some surgeons regularly visit the frozen room to facilitate this communication. Ambiguous scenarios include nodal sampling, unoriented specimens with multiple potential margins, tumor identification versus margin assessment, or specimens that contain orienting sutures without explanation. While these questions can take some time to answer, addressing them prior to specimen handling can avoid later delays and performance of unnecessary additional frozen sections.

Once the tissue is weighed, measured, and evaluated macroscopically, an area is selected for cryostat freezing. Selection is important in order to demonstrate the desired pathology. Areas to be selected may include a mass lesion, its relationship to a resection margin, and an interface with normal tissue. For many samples, it is important to remove adipose tissue because it cannot be effectively cryosectioned; abundant adipose tissue can impact the quality of the frozen section. This is especially important when lymph nodes are grossly embedded in adipose tissue or are especially fatty.

Tissues may be frozen upright or inverted. The goal of both methods is to orient the tissue such that the cut surface reveals the pathology. While this sounds straightforward, the geometry of the tissue can make this difficult. For example, a long flat structure may need to be oriented on edge in order to see the margin. Tissues are embedded in optimal cutting temperature (OCT) mounting medium, which is a clear viscous liquid at room temperature that becomes solid, white, and opaque when frozen at −20 °C. A tissue can be manipulated and reoriented in OCT prior to freezing, but its position can be difficult to maintain while in the liquid viscous material. Once placed in the cryostat, the OCT hardens, and tissue cannot be repositioned. Inverted freezing involves use of a cryomold with application of OCT that allows for some degree of tissue repositioning along the cutting surface. The OCT is then placed to cover the tissue; a metal platform is placed over it and left to freeze, creating an OCT-embedded frozen tissue block on a metal platform. Upright freezing places the

tissue on an OCT covered chuck, repositioning tissue placement in OCT and covering with a flat metal dehydrator to freeze, dehydrate, and create a flat cutting surface. In general, frozen tissue without OCT will adhere to the cold metal and cannot be repositioned. It is important to wait until the tissue and OCT are completely frozen when using either method. Incompletely frozen tissue can be difficult to cut, and it may separate from the metal base, resulting in significant delays.

Once the block is completely frozen, the metal base of the chuck is mounted on the cryostat, and the tissue is oriented such that it can clear the cutting blade of the microtome. It is best to place long strips of tissue perpendicular to the blade, as folds occur in the plane of the knife, and this minimizes folds in the plane of the tissue being cut. Most sections are cut between 4 and 6 microns. Cut sections are mounted onto a warm slide and immediately fixed. While staining protocols vary, a hematoxylin and eosin stain is generally performed. Slides are subsequently dehydrated with either xylene or xylene substitutes, covered with adhesive, and protected with a coverslip.

During this process it is important to maintain specimen identification as well as part and block identification. This can be done by inserting a paper label during the freezing process or labeling the block directly within a marker. Slides are labeled with two patient identifiers as well as the part, block, and slide number. The slide number indicates which slice was taken first, which may not represent a full cut surface of the frozen tissue.

The slide is then evaluated by the pathologist. Considerations include correlating the gross appearance of the slide findings, determining whether the tissue section is full face and representative, and assessing the technical quality of the slide. The first section is often adequate, and a diagnosis can be rendered. Deeper sections can be performed to improve staining quality, eliminate tissue folds, obtain a full tissue surface, or better assess atypical foci.

Reporting frozen sections in real time can be performed with a verbal or written report delivered to the operating room. Some facilities may have a fax or electronic system, but most facilities use phone calls as a verbal reporting system. Phone discussions should include verification of the diagnosis with verbal read-back and documentation of the names of persons delivering and receiving the report. This avoids the misunderstanding of a verbal report, especially when phrases include both positive and negative meaning (e.g., no carcinoma seen versus carcinoma seen). Some pathologists prefer terminology that is unambiguous such as negative or positive, and this may be a better practice to avoid confusion [7]. The entire intraoperative consultation can be done quite rapidly with a turnaround time of approximately 20 minutes or less for an uncomplicated case.

There are several well-known pitfalls of frozen section evaluation. Small tissues can be difficult to handle due to apprehension that the tissue will be lost during sectioning. Taking additional sections while generating a full face can mitigate against this concern. Melanocytes of melanoma in situ can be difficult to recognize in frozen sections; epidermal margins of melanocytic lesions should not be performed. Cortical bone often poses difficulty when sectioned with a

microtome-cryostat, and even tissues that contain numerous bony fragments can produce sufficient technical difficulty as to render a frozen section impossible to interpret. Adipose tissue can be especially problematic in that it will not produce a section unless it is significantly abnormal, such as fat infiltrated with tumor. If the margin of a specimen is predominantly composed of adipose tissue, it can be difficult to assess. Changing the cutting temperature or attempting a thicker section can improve the section quality in some cases.

It may be better to avoid frozen sections when evaluating adipose or bony tissues; touch preparations can be attempted in these cases [8]. It is important to think about this before freezing, as such cytologic preparations cannot be performed after the tissue is frozen. Cytologic preparations can be performed quickly and can be a very useful tool [9]. These can be done as touch preps, scrapes, and smears. Rapidly fixed or air-dried slides can be stained with a Diff-Quik stain, whereas rapidly fixed slides can also be stained in the same H&E process as the frozen section. Certain features, such as mucin or the matrix of mixed tumors, can be readily identified using cytologic preparations. Nuclear features, such as the inclusions and chromatin patterns of papillary thyroid carcinoma, can be better preserved with such methods. Perinuclear clearing of plasma cells and other cytoplasmic features are more easily identified on touch preparations. Cytologic preparations can also be used to assess cellularity and decide how to partition a specimen.

Squash preparations avoid some of the artifacts of frozen sections and accentuate cellular architecture by creating a more three-dimensional view [10]. These are most often used in neuropathology and will be covered in Chap. 10.

The record of a frozen section is kept as the glass slide, but the frozen tissue itself can be fixed, thereby serving as a permanent section for comparison. This is an important quality assurance tool, as the morphologic features present in the permanent section are better demonstrated than those of the frozen section slide. Retention of the paraffin block enables the performance of immunohistochemistry or even molecular testing as needed. However, tissue morphology of frozen and subsequently thawed tissue is inferior to that of nonfrozen tissue that is immediately fixed. Therefore, tissue selection requires careful judgment to include as much of the lesion as is needed for diagnosis, but not to freeze so much lesional tissue that a final diagnosis is impeded. This is particularly important when dealing with small tumors for which distinctions between benign and malignant entities requires excellent morphology. This is also an excellent reason to discourage unnecessary frozen sections that are requested to appease curiosity but do not change the surgical procedure.

Conclusions

Frozen section is a rapid and diagnostically accurate technique that can be used to immediately affect surgical management of the patient. The use of intraoperative consultation has evolved over time, but it remains a mainstay of rapid diagnosis in numerous subspecialty areas.

References

1. Brender E, Burke A, Glass RM. JAMA patient page. Frozen section biopsy. JAMA. 2005;294(24):3200.
2. Gal AA. The centennial anniversary of the frozen section technique at the Mayo Clinic. Arch Pathol Lab Med. 2005;129(12):1532–5.
3. Wilson LB. A method for the rapid preparation of fresh tissues for the microscope. J Am Med Assoc. 1905;45(23):1737.
4. Arcega RS, Woo JS, Xu H. Performing and cutting frozen sections. Methods Mol Biol. 2019;1897:279–88.
5. Peters SR. A practical guide to frozen section technique. New York: Springer; 2010. xi, 193 pp.
6. Davis DA, Pellowski DM, William Hanke C. Preparation of frozen sections. Dermatol Surg. 2004;30(12 Pt 1):1479–85.
7. Nakhleh RE. Quality in surgical pathology communication and reporting. Arch Pathol Lab Med. 2011;135(11):1394–7.
8. Rahman K, Asif Siddiqui F, Zaheer S, Sherwani MK, Shahid M, Sherwani RK. Intraoperative cytology--role in bone lesions. Diagn Cytopathol. 2010;38(9):639–44.
9. Salem AA, Douglas-Jones AG, Sweetland HM, Newcombe RG, Mansel RE. Evaluation of axillary lymph nodes using touch imprint cytology and immunohistochemistry. Br J Surg. 2002;89(11):1386–9.
10. Hamasaki M, Chang KHF, Nabeshima K, Tauchi-Nishi PS. Intraoperative squash and touch preparation cytology of brain lesions stained with H+E and diff-quik: a 20-year retrospective analysis and comparative literature review. Acta Cytol. 2018;62(1):44–53.

Quality Assurance in Frozen Section

2

Alain C. Borczuk

Background

Intraoperative consultation is a real-time preliminary macroscopic or histologic diagnosis that enables active decision-making during medical procedures, most often surgical procedures. Such intraoperative diagnoses can be performed as macroscopic examinations only but more often combine a macroscopic examination with a histologic one. Overall, this is an accurate diagnostic tool, although knowledge of the pitfalls by the pathologist and refinement of the appropriate clinical scenarios for its use assures diagnostic value and minimizes errors that can impact type and extent of surgery. In order to gain this experience, individual pathologists as well as pathology laboratories need to incorporate the assessment of intraoperative consultation into both individual performance parameters as well as institutional quality programs.

Existing Guidelines

There are several organizations that survey laboratories and provide specific guidance for the evaluation of frozen sections and intraoperative consultation. The College of American Pathologists quality assurance checklist has areas of focus for laboratories that perform these procedures. These items include staining quality and reagent changes to maintain uniform technical quality, as well as issues that pertain to maintaining patient identification in the process of rapid assessment, especially when there are multiple slides and parts per case. Since the frozen section block often needs to be removed from the cutting area in the course of the frozen section,

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a method of temporary block labeling is needed to avoid mix-ups. In laboratories with barcoding through the laboratory, the frozen section may require temporary labels during and after the procedure which need to be made permanent for processing.

The other aspects of frozen section involve real-time reporting which is often verbal, given the location of the operating room and pathology department. With verbal reporting, issues of patient identification and accuracy of the frozen report need to be addressed, often with methodology regarding verbal readback. One checklist item also requires that the pathologist have the opportunity to speak with the procedure operator. In the case of written reports, a methodology for receipt of the report in real-time is needed, along with acknowledgment of receipt.

It is a provision of a quality assurance program that frozen section tissue be evaluated in comparison with the permanent sections prepared from the same block. This requires that the frozen tissue be thawed and processed into a formalin-fixed paraffin embedded block and sectioned in conjunction with the overall case evaluation. While not essential, the ideal quality program has a different pathologist reviewing the frozen-permanent correlation than the pathologist who performed the intraoperative consultation. The frozen section diagnosis should be documented in the final report, and in the case of a discrepancy, the nature of the discrepancy noted. This can include the reason for the false-positive, false-negative, or incompatible diagnosis. Overall the accuracy of frozen section can be evaluated by the individual pathologist for peer performance evaluation but also be assessed in the aggregate as part of a quality improvement program. Such activities can lead to institution-specific rates of false positive, false negative, and deferral which would correspond to the specific case mix. There are no specific benchmarks for these rates.

While quality probes indicate a turnaround time for uncomplicated cases at 20 minutes as a reasonable benchmark, specific checklist items monitoring turnaround time have been eliminated. It is up to the individual laboratory to decide the utility of monitoring this parameter and to develop institutionally acceptable levels for turnaround time.

Accuracy: False Positives and Incompatible Diagnoses, False Negatives, and Deferrals

Studies analyzing the accuracy of frozen section have included general surgical pathology as well as studies of organ-based subspecialty pathology. While accuracy has been reported in various ways, diagnostic errors are categorized as false positives and false negatives. False-positive diagnoses are largely interpretative errors resulting in an incompatible diagnosis (e.g., degree of malignancy or a malignant diagnosis but with different treatment implication) or an overcall of malignancy for a benign diagnosis. False-negative diagnoses include interpretative and sampling errors. Deferrals are neither false-positive nor false-negative but instead indicate a limitation in obtaining a definitive diagnosis.

Frozen section is a highly accurate technique, but its performance is influenced by a variety of factors. Judicious use of intraoperative consultations, deferral to permanent, careful sampling of gross lesions, and experience of the pathologist can all influence the value of frozen sections. In addition, certain subspecialty areas have particular pitfalls which need to be recognized by the frozen section pathologist [1].

While published definitions of accuracy vary, 96–97% accuracy overall is reported when limited to cases in which a diagnosis is rendered [2, 3]. For example, in a large general frozen section series, an overall concordance rate of 95% was reported, but after a deferral rate of 1%, the accuracy was 96.1% comprised of a false-negative rate of 3.7% and a false-positive rate of 0.2%. Deferrals were common when frozen sections involved soft tissue tumors; lymph nodes accompanying breast cancers were a source of false-negative interpretations resulting from sampling. In a series of over 24,000 cases [4], the accuracy of frozen section was reported at 97.8% with 1.6% false negatives, largely due to sampling, and an additional 0.5% that were incompatible, with 0.1% with a significant incompatible result. In most instances, false-negative rates exceeded false-positive rates substantially, as false-negative rates are impacted by the limitations of sampling inherent in the technique [5]. These series are in line my institutional experiences in that false-negative rates substantially exceed false-positive rates, and that sampling is the most common root cause of discrepancy. It is of note that while not specifically mentioned in these studies, sampling can be impacted by technical challenges such as lymph nodes rich in adipose tissue. Careful macroscopic examination can to a degree partially mitigate against this pitfall.

While false-positive and false-negative rates are generally stable within a department and consistent between departments, deferral rates can vary substantially. This experience warrants a careful quality improvement program that divides deferrals by either organ system or scenario. For example, it is recognized that malignant or benign characterization of soft tissue lesions can lead to error without the ancillary studies used on permanent sections material; such lesions may be routinely deferred. This statement may also apply to lymphoid lesions in which ancillary studies are routinely performed or thyroid follicular lesions that may require complete capsule sampling. Such a strategy of providing adequacy or margin information without characterizing the actual lesion can lead to a deferral that is warranted and appropriate. As a result surgical pathology experience in frozen section shows that deferral rates can vary considerably based on specimen mix and that deferral rates of over 5% do not necessarily indicate poor diagnostic ability [5]. In such instances, active QA programs to monitor trends in deferral rates as well as the type of cases deferred will assure that deferral is used appropriately.

Certain areas in which primary diagnosis without preoperative biopsy remains the standard of care, for example, in lung lesions, discrepancies that are incompatible with permanent section will occur at higher rate and have the potential to be clinically relevant errors. As biopsy techniques become more prevalent and safer, the role of frozen section will continue to change. Preoperative biopsies will reduce

the number of intraoperative false positives and incompatibles but may also shift the role of frozen section from primary diagnosis to assessment of stage through lymph node sampling or margins. In that scenario, given the higher false-negative rate than false-positive rate inherent to the technique, quality improvement programs need to re-focus their attention on the root cause of false negatives and attempt to improve or direct sampling. Examples of such innovations have led to sentinel node sampling and protocols around lymph node sectioning and leveling. One series that examined additional levels in margin assessment, while finding little overall impact, discovered that positive detection was improved [6].

Special Considerations for Subspecialty Areas

The use of frozen section in non-melanoma skin margins is commonplace and has specific guidelines, including the College of American Pathologists checklist. In a non-melanoma skin pathology study of 300 cases, 83% of diagnoses were concordant between frozen and permanent sections, with higher concordance rates for basal cell carcinoma (95%). The importance of the technique is turnaround time and rapidity, although the false-negative rate was reported as an area for improvement [7].

Accuracy of 97.1% [8] was reported for pathologists in gynecologic pathology frozen section. False positive and false negatives were compared based on subspecialization, with relatively good sensitivity and specificity for benign tumors and specificity for malignant tumors, but with differences in interpretation for borderline tumors. Interestingly, in a separate study, borderline ovarian tumors [9] were noted as having a diagnostic frozen to permanent correlation of 90.6%, indicating higher complexity in this area. Large tumors with borderline areas were also a source of error in another series [10]. The accuracy of frozen section was high with 97.5% concordance, but there was a small potential to underestimate malignancy, including in mucinous tumors, leading to a recommendation to sample multiple blocks in mucinous tumors [11]. It is clear that frozen section evaluation of ovarian tumors can distinguish benign from borderline or malignant tumors but with errors in borderline versus malignant categories [12]. This type of observation can provide guidance for the general pathologist performing frozen sections to help decide when to submit more tissue and importantly, when to consult a subspecialty pathologist.

In a series of 1796 head and neck frozen sections, 3.6% were discordant, with 1.9% false negatives, largely due to sampling and 1.1% false positive, which were largely interpretative [13]. In some arenas such as parathyroid identification, the accuracy level is very high with over 99% concordance [14]. Knowledge of pitfalls, such as morphologic overlap between thyroid and parathyroid in rare instances, can help avoid errors by introduction of consultation, additional frozen sections, or use of smears.

In a study of sentinel node pathology in breast cancer, a false-negative rate of 4.9% was reported, with three quarters of cases due to sampling, including errors of individual tumor cell and micrometastasis. Only 0.3% represented false positives [15]. In breast pathology, while frozen section was more commonplace prior to the year 2000, the use of screening techniques, reduction in the size of index lesions, and the use of preoperative biopsy have reduced utilization of frozen section independent of accuracy and turnaround time [16, 17].

A high level of sensitivity (98%) and specificity (94%) is reported for nervous system frozen sections, and they are especially good at detecting pathology with a positive predictive value of 99%. However, diagnostic difficulties do remain in this area with a proportion of incompatible pathology [18].

The use of frozen section in the evaluation of margins in genitourinary and hepatobiliary pathology is commonplace and yields highly accurate results [19, 20] when compared to permanent sections. In a study of pancreatic margin pathology, while accuracy was good, the study questioned the role of frozen section as residual disease was not associated with overall survival [21].

Impact of QA Monitoring in Frozen Section

An examination of a large inter-institutional Q-probes program in pathology [22] supported the view that monitoring could lead to reductions in discordant rates over time. Turnaround time could be improved to meet a 20-minute benchmark by a focus on multipart specimens, obtaining prior material for surgical patients, and consultations [23].

However, some subspecialty cases of frozen section lack diagnostic accuracy [24], and their use is therefore subject to specific situations, such as frozen section in follicular lesions of the thyroid. Such data can be used by the frozen section service to determine best use of these techniques. For example, preoperative fine needle aspiration can reduce the need for frozen section considerably in thyroid follicular lesions [25], an area for which frozen section is known to have low sensitivity.

Touch Preparations

The use of touch preps and smears can be valuable in frozen section and intraoperative assessment, reducing deferral and discrepancy rates [26]. In some instances, this approach can replace frozen section [27] although this may be dependent on the experience of the pathologist with tissue examination versus cytology. However, in lesions of the central nervous system as well as in the evaluation of lymph nodes and thyroid nodules, cytologic preparations are extremely valuable.

Telepathology in Frozen Section

The use of telepathology systems includes virtual slide systems (review of a whole slide image) and robotic microscopes – live view systems with remote pathologist control. The discordance rates between glass slide and robotic technologies are low, reportedly only 0.35% in one study [28]. In one series deferral rates were different between the two approaches. In a series of 5233 cases of whole slide image review versus local review, a turnaround time of under 30 minutes, a false-positive rate of 0.04%, and false-negative rate of 0.19% were achieved with whole slide imaging, which was comparable to results of local review. Deferral rates were also comparable. However, case mix did vary, with more frozen sections of the central nervous system, lung, and pancreas from the local rather than regional hospitals [29].

A comparison of virtual slide to robotic microscopy showed [30] a similar performance in accuracy and deferral rates for the two modalities, with a markedly reduced turnaround time for a virtual slide. In addition, virtual slide review allowed for faster consultation by image sharing. One pitfall was the occurrence of mid-case technical problems in three cases. Although these events were infrequent, this resulted in prolonged turnaround time to reporting and required emergency slide transport to the pathologist.

In another series, comparing pre- and post-digital pathology implementation, turnaround time was unchanged, although one failure was reported [31]. The accuracy for whole slide imaging was evaluated in neoplastic and non-neoplastic disease with equally good results in accuracy and turnaround time [32].

Conclusion

While frozen section remains a highly accurate procedure, experienced pathologists can learn to identify situations prone to error and adapt by submitting more sections, cutting more levels, obtaining real-time second opinions, or defer to permanent sections, as needed. An understanding of the clinical scenario helps in communicating the necessary level of lesion classification without overcall. Maintaining an active quality assurance program is a best practice for establishing institution-specific baselines. Improvements in accuracy, documentation, and turnaround time are all elements of a successful program. The incorporation of new technology, such as robotic microscopy and whole slide images, is promising and appears to be as accurate as primary glass slide interpretation for frozen section diagnosis.

References

1. Wen MC, Chen JT, Ho WL. Frozen-section diagnosis in surgical pathology: a quality assurance study. Kaohsiung J Med Sci. 1997;13:534–9.
2. Adhikari P, Upadhyaya P, Karki S, Agrawal CS, Chettri ST, Agrawal A. Accuracy of frozen section with histopathological report in an institute. JNMA J Nepal Med Assoc. 2018;56:572–7.

3. Winther C, Graem N. Accuracy of frozen section diagnosis: a retrospective analysis of 4785 cases. APMIS. 2011;119:259–62.
4. Ferreiro JA, Myers JL, Bostwick DG. Accuracy of frozen section diagnosis in surgical pathology: review of a 1-year experience with 24,880 cases at Mayo Clinic Rochester. Mayo Clin Proc. 1995;70:1137–41.
5. Oneson RH, Minke JA, Silverberg SG. Intraoperative pathologic consultation. An audit of 1,000 recent consecutive cases. Am J Surg Pathol. 1989;13:237–43.
6. Olson SM, Hussaini M, Lewis JS Jr. Frozen section analysis of margins for head and neck tumor resections: reduction of sampling errors with a third histologic level. Mod Pathol. 2011;24:665–70.
7. Onajin O, Wetter DA, Roenigk RK, Gibson LE, Weaver AL, Comfere NI. Frozen section diagnosis for non-melanoma skin cancers: correlation with permanent section diagnosis. J Cutan Pathol. 2015;42:459–64.
8. Bige O, Demir A, Saygili U, Gode F, Uslu T, Koyuncuoglu M. Frozen section diagnoses of 578 ovarian tumors made by pathologists with and without expertise on gynecologic pathology. Gynecol Oncol. 2011;123:43–6.
9. Shah JS, Mackelvie M, Gershenson DM, Ramalingam P, Kott MM, Brown J, Gauthier P, Nugent E, Ramondetta LM, Frumovitz M. Accuracy of intraoperative frozen section diagnosis of borderline ovarian tumors by hospital type. J Minim Invasive Gynecol. 2019;26:87–93.
10. Basaran D, Salman MC, Boyraz G, Selcuk I, Usubutun A, Ozgul N, Yuce K. Accuracy of intraoperative frozen section in the evaluation of patients with adnexal mass: retrospective analysis of 748 cases with multivariate regression analysis. Pathol Oncol Res. 2015;21:113–8.
11. Wang KG, Chen TC, Wang TY, Yang YC, Su TH. Accuracy of frozen section diagnosis in gynecology. Gynecol Oncol. 1998;70:105–10.
12. Kung FY, Tsang AK, Yu EL. Intraoperative frozen section analysis of ovarian tumors: a 11-year review of accuracy with clinicopathological correlation in a Hong Kong Regional hospital. Int J Gynecol Cancer. 2019;29:772–8.
13. Layfield EM, Schmidt RL, Esebua M, Layfield LJ. Frozen section evaluation of margin status in primary squamous cell carcinomas of the head and neck: a correlation study of frozen section and final diagnoses. Head Neck Pathol. 2018;12:175–80.
14. Westra WH, Pritchett DD, Udelsman R. Intraoperative confirmation of parathyroid tissue during parathyroid exploration: a retrospective evaluation of the frozen section. Am J Surg Pathol. 1998;22:538–44.
15. Poling JS, Tsangaris TN, Argani P, Cimino-Mathews A. Frozen section evaluation of breast carcinoma sentinel lymph nodes: a retrospective review of 1,940 cases. Breast Cancer Res Treat. 2014;148:355–61.
16. Bianchi S, Palli D, Ciatto S, Galli M, Giorgi D, Vezzosi V, Del Turco MR, Cataliotti L, Cardona G, Zampi G. Accuracy and reliability of frozen section diagnosis in a series of 672 nonpalpable breast lesions. Am J Clin Pathol. 1995;103:199–205.
17. Scheiden R, Sand J, Tanous AM, Knolle U, Capesius C, Wagnon MC, Faverly D. Accuracy of frozen section diagnoses of breast lesions after introduction of a national programme in mammographic screening. Histopathology. 2001;39:74–84.
18. Balsimelli LBS, Oliveira JC, Adorno FA, Brites CA, Bublitz GS, Tavares LCC, Coelho K, Stall J, Franca PHC. Accuracy of intraoperative examination in central nervous system lesions: a study of 133 cases. Acta Cytol. 2019;63:224–32.
19. Cioc AM, Ellison EC, Proca DM, Lucas JG, Frankel WL. Frozen section diagnosis of pancreatic lesions. Arch Pathol Lab Med. 2002;126:1169–73.
20. Kates M, Ball MW, Chappidi MR, Baras AS, Gordetsky J, Sopko NA, Brant A, Pierorazio PM, Epstein JI, Schoenberg MP, Bivalacqua TJ. Accuracy of urethral frozen section during radical cystectomy for bladder cancer. Urol Oncol. 2016;34:532 e531–6.
21. Dikmen K, Kerem M, Bostanci H, Sare M, Ekinci O. Intra-operative frozen section histology of the pancreatic resection margins and clinical outcome of patients with adenocarcinoma of the head of the pancreas undergoing pancreaticoduodenectomy. Med Sci Monit. 2018;24:4905–13.

22. Raab SS, Tworek JA, Souers R, Zarbo RJ. The value of monitoring frozen section-permanent section correlation data over time. Arch Pathol Lab Med. 2006;130:337–42.
23. Novis DA, Zarbo RJ. Interinstitutional comparison of frozen section turnaround time. A College of American Pathologists Q-Probes study of 32868 frozen sections in 700 hospitals. Arch Pathol Lab Med. 1997;121:559–67.
24. Grisales J, Sanabria A. Utility of routine frozen section of thyroid nodules classified as follicular neoplasm. Am J Clin Pathol. 2020;153:210–20.
25. Bronner MP, Hamilton R, LiVolsi VA. Utility of frozen section analysis on follicular lesions of the thyroid. Endocr Pathol. 1994;5:154–61.
26. Cimic A, Mironova M, Khoury-Collado F, Salih Z. Cytologic smears improve accuracy of frozen sections of ovarian tumors in the community practice settings. Cytojournal. 2019;16:10.
27. Liu Y, Silverman JF, Sturgis CD, Brown HG, Dabbs DJ, Raab SS. Utility of intraoperative consultation touch preparations. Diagn Cytopathol. 2002;26:329–33.
28. Dunn BE, Choi H, Recla DL, Kerr SE, Wagenman BL. Robotic surgical telepathology between the Iron Mountain and Milwaukee Department of Veterans Affairs Medical Centers: a 12-year experience. Hum Pathol. 2009;40:1092–9.
29. Huang Y, Lei Y, Wang Q, Li D, Ma L, Guo L, Tang M, Liu G, Yan Q, Shen L, Tong G, Jing Z, Zhang Y, Deng Y. Telepathology consultation for frozen section diagnosis in China. Diagn Pathol. 2018;13:29.
30. Evans AJ, Chetty R, Clarke BA, Croul S, Ghazarian DM, Kiehl TR, Perez Ordonez B, Ilaalagan S, Asa SL. Primary frozen section diagnosis by robotic microscopy and virtual slide telepathology: the University Health Network experience. Hum Pathol. 2009;40:1070–81.
31. French JMR, Betney DT, Abah U, Bhatt N, Internullo E, Casali G, Batchelor TJP, West DG. Digital pathology is a practical alternative to on-site intraoperative frozen section diagnosis in thoracic surgery. Histopathology. 2019;74:902–7.
32. Cima L, Brunelli M, Parwani A, Girolami I, Ciangherotti A, Riva G, Novelli L, Vanzo F, Sorio A, Cirielli V, Barbareschi M, D'Errico A, Scarpa A, Bovo C, Fraggetta F, Pantanowitz L, Eccher A. Validation of remote digital frozen sections for cancer and transplant intraoperative services. J Pathol Inform. 2018;9:34.

Intraoperative Evaluation of the Gastrointestinal Tract

3

Erika Hissong and Rhonda K. Yantiss

Introduction

Intraoperative consultation for diseases of the luminal gut can facilitate the immediate management of surgical patients in several situations. Most of the tubular gut is easily accessible to endoscopic evaluation, and, thus, many patients have an established diagnosis prior to surgery. The most common indication for intraoperative consultation of the tubular gut is evaluation of resection margins in the setting of neoplasia. Frozen sections may also be used to establish a primary diagnosis when lesions are not amenable to endoscopic evaluation, intraoperative findings raise the possibility of malignancy, or incidental peritoneal nodules are detected during cancer operations. Frozen sections are performed in the latter situation to exclude the possibility of metastatic disease; many patients with stage IV disease do not benefit from resection of the primary tumor. Lastly, the intraoperative evaluation of patients with possible Hirschsprung disease relies heavily on pathologists to identify the extent of aganglionosis in order to optimize extent of resection. It is important that pathologists offer timely, accurate diagnoses with full understanding of the specific concerns to be addressed in all of these scenarios. The purpose of this chapter is to discuss the situations in which pathologists may be asked to perform intraoperative consultations for surgeons treating patients with diseases of the tubular gut.

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Evaluation of the Esophagus

Surgical resection of the esophagus and/or gastroesophageal junction usually occurs in the setting of malignancy. While squamous cell carcinoma occurs anywhere in the esophagus and remains the most common esophageal malignancy worldwide, its incidence in Western countries is lower than that of adenocarcinoma. Esophageal and gastroesophageal adenocarcinomas now comprise >50% of all malignant esophageal neoplasms in the United States and occur almost exclusively in association with Barrett esophagus [1]. Both esophageal squamous cell carcinoma and adenocarcinoma show a male predominance. Virtually all tumors are preoperatively diagnosed with endoscopic examination, fine needle aspiration biopsy, and/or mucosal biopsy, often in combination with endoscopic ultrasound and other radiologic studies to determine clinical stage prior to operative management. Most patients with stage III or IV disease are offered chemotherapy and/or radiation, and some of those with stage II or III tumors are treated with neoadjuvant therapy prior to definitive resection. Superficially invasive tumors confined to the mucosa or submucosa may be amendable to wide local excision via endoscopic submucosal dissection, and some T2N0 tumors may even be managed with local excision and systemic therapy [2–4]. These local excisional specimens are rarely sent for frozen section analysis because most patients are followed closely with endoscopic surveillance regardless of margin status. On the other hand, frozen section plays a much larger role in the evaluation of margins of esophagectomy resection specimens. The presence of a positive resection margin is an independent predictor of disease recurrence and is associated with decreased overall survival [5–9]. Both proximal and distal margins may be assessed depending on the tumor location. While negative distal margins are often achievable, the nature of the surgical procedure (e.g., low intrathoracic resection versus more extensive resection with cervical anastomosis) may limit the amount of proximal esophagus that can be removed. For this reason, the proximal esophageal margin is more likely to be close to the tumor than the distal margin. Radial margins are not assessed by frozen section analysis in most cases because preoperative staging generally determines tumor resectability.

Squamous cell carcinomas often arise in association with high-grade dysplasia, which imparts a corrugated appearance to the esophageal mucosa (Fig. 3.1). Squamous dysplasia features a lack of surface maturation and an atypical population of cells with enlarged, hyperchromatic nuclei, single-cell necrosis, and increased mitotic activity (Fig. 3.2a, b). There is often an abrupt transition between the atypical population and the adjacent non-lesional epithelium. Invasive carcinomas appear as raised mucosa with nodules or ulcers (Fig. 3.3). Infiltrating cells are epithelioid or spindled with abundant eosinophilic cytoplasm and overtly malignant cytoplasmic features with or without keratinization. High-grade glandular dysplasia is rarely treated by esophagectomy in the modern era but can be present in the background mucosa of patients with Barrett-associated adenocarcinomas (Fig. 3.4).

Intraoperative macroscopic examination can be used to estimate the tumor clearance and may even be used in lieu of microscopic examination when bulky tumors are present distant from the resection margins. Most surgical guidelines recommend

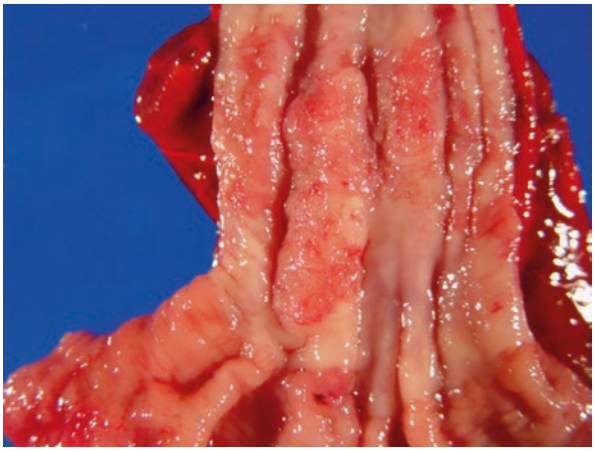

Fig. 3.1 A large plaque of high-grade squamous dysplasia produces thickened, slightly nodular esophageal mucosa with a granular appearance. The gastroesophageal junction is essentially normal

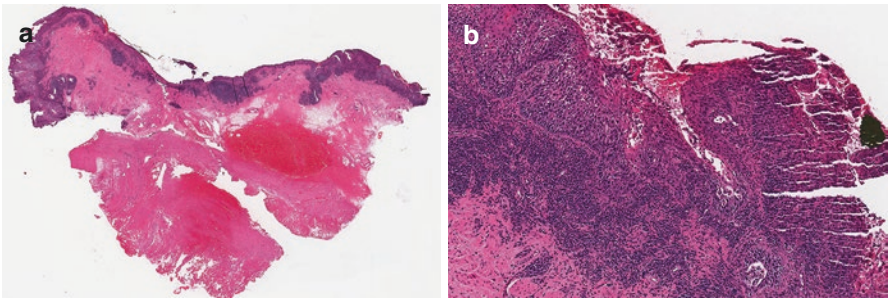

Fig. 3.2 Squamous cell dysplasia extends to the proximal margin of an esophagectomy specimen. An atypical population of squamous cells expands the mucosa (**a**). Lesional cells contain enlarged, hyperchromatic nuclei with irregular membranes (**b**)

a tumor clearance of at least 3 cm, although larger tumors and those with poor differentiation require greater macroscopic distance because microscopic extension can exceed the macroscopic tumor size [9–12]. This is especially true among neoadjuvantly treated patients with diffusely infiltrating tumors that show a partial response to therapy. In fact, as many as 50% of patients who receive neoadjuvant therapy have no or only minimal residual macroscopic tumor at the time of resection but still have residual microscopic disease [13, 14]. While the clinical benefit of intraoperative microscopic margin assessment in every case has yet to be fully elucidated, this is currently the practice at many institutions. Treated tumors show extensive regression with variable amounts of residual tumor deep within the wall or in the submucosa (Fig. 3.5a, b). Neoadjuvant therapy can also cause significant reactive atypia in non-neoplastic cells that simulates dysplasia or carcinoma. However, dysplasia generally shows an abrupt transition to more clearly benign epithelium, and carcinomas are poorly circumscribed with infiltrative edges. On the other hand, benign glands show a lobular arrangement in the mucosa and submucosa.

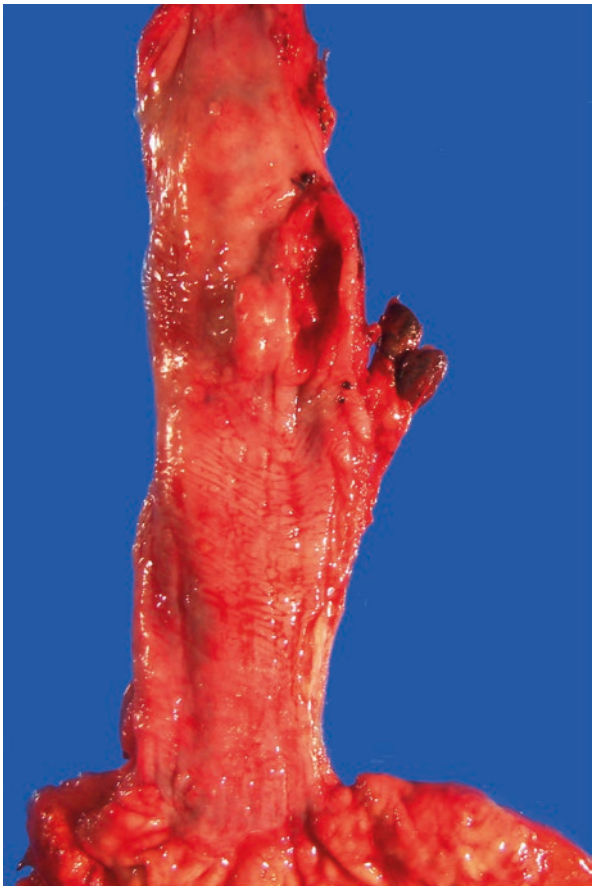

Fig. 3.3 Invasive squamous cell carcinoma appears as an ulcerated mass with adjacent nodule in the mid esophagus

Although cautery and frozen section artifacts can preclude a definitive diagnosis of invasive carcinoma, Barrett esophagus-related dysplasia, and squamous dysplasia, false-negative interpretations usually result from sampling errors. There are no recommendations regarding whether margins should be entirely frozen or if representative sections closest to the tumor are sufficient for intraoperative diagnosis [15]. The proximal esophageal resection margin is generally frozen entirely en face, whereas most pathologists selectively freeze only the region of the distal gastric margin closest to the tumor. Older data suggested that false-negative rates for gastric margins assessed in this fashion ranged from 9% to 21% [16]. However, a recent study re-examining gastric margin sampling during intraoperative consultation reported similar positive margin rates when comparing representative sampling to submission of the entire resection margin [17].

Intraoperative frozen section analysis may also be requested to rule out metastatic disease when enlarged lymph nodes are found outside the regional nodal basin or nodules are identified on the peritonealized surfaces, diaphragm, peripheral lung, or liver capsule during surgical exploration at the time of esophagectomy.

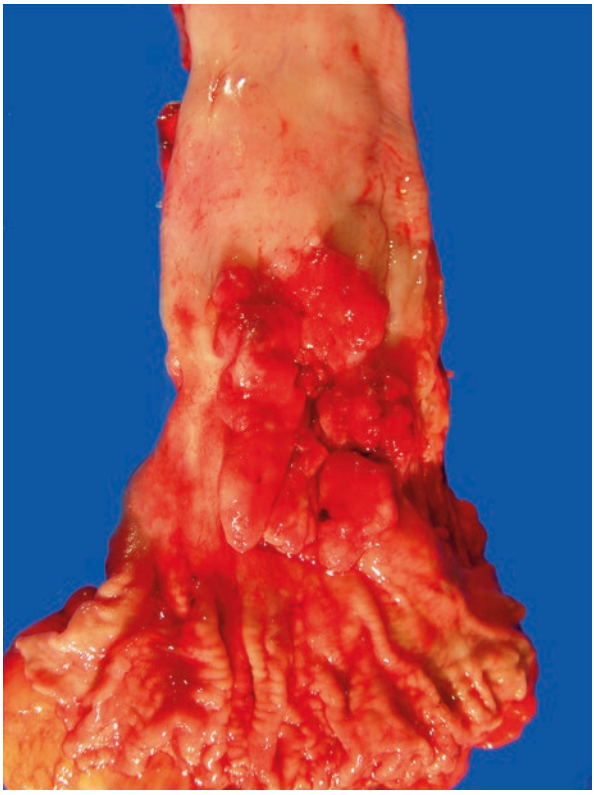

Fig. 3.4 Solid nodules of invasive adenocarcinoma are present at the gastroesophageal junction. The background mucosa is erythematous and thickened due to the presence of high-grade glandular dysplasia

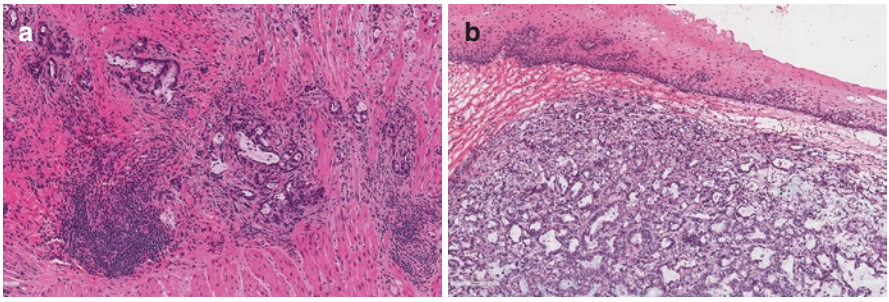

Fig. 3.5 Ill-defined aggregates of malignant glands are present in the wall at the resection margin of a neoadjuvantly treated patient (**a**). A residual nodule of malignant glands is present in the submucosa (**b**)

Non-neoplastic lesions including prominent pneumocytes, mesothelial cells, fat necrosis, organizing hematomas, fibrosis, and benign hepatic lesions, such as bile duct hamartoma or bile duct adenoma, can simulate the appearance of metastatic carcinoma as discussed in Chaps. 4 and 9.

Non-epithelial or mural-based esophageal tumors may be submitted for intraoperative consultation to establish a primary diagnosis in some cases. Leiomyoma is the most common mesenchymal tumor of the esophagus, often occurring in the mid- to lower esophagus and arising from the muscularis propria (Fig. 3.6). These tumors are generally well-circumscribed and composed of bland, pink, spindled cells with cigar-shaped nuclei lacking cytologic atypia, necrosis, and mitotic figures (Fig. 3.7). Peripheral nerve sheath tumors and granular cell tumors are rarely submitted for intraoperative consultation, in which case the frozen section diagnosis may simply be "benign spindle cell neoplasm" without further classification. Tumor cellularity, nuclear pleomorphism, mitotic activity, and tumor necrosis are features of malignancy [18, 19]. Gastrointestinal stromal tumors rarely occur in the esophagus. They may be morphologically similar to their gastric counterparts but are often more cellular with increased mitotic activity and necrosis (Fig. 3.8a, b). Metastases to the esophagus should be kept in mind with any mural-based lesion, particularly in the setting of a previously diagnosed malignancy.

Fig. 3.6 Esophageal leiomyomas are often multiple, mural masses with overlying normal mucosa

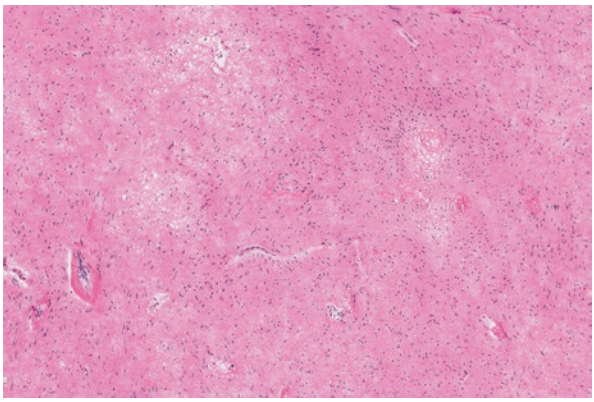

Fig. 3.7 Esophageal leiomyomas are paucicellular and contain bland spindle cells with abundant eosinophilic cytoplasm. Cytologic atypia, mitotic activity, and necrosis are lacking

Stomach

Intraoperative consultations are often obtained to evaluate surgical resection margins of gastric carcinomas, most of which have been previously diagnosed and staged. Although there are no universally agreed upon rules regarding assessment of gastric margins, the Japanese Gastric Cancer Association defines adequate tumor clearance as 2 cm for carcinomas invasive of the submucosa, 3 cm for tumors that extend into or beyond the muscularis propria, and at least 5 cm for tumors with an infiltrating growth pattern [20]. While macroscopic evaluation of tumor distance can be used in lieu of frozen section analysis, this practice may miss cases with subepithelial extension of diffuse-type carcinomas [12]. Perpendicular sections including tumor and margin are recommended for tumors within 2 cm of the margin; en face margins are likely sufficient for cases with more than 2 cm of clearance upon gross examination (Fig. 3.9a, b). Diffuse carcinomas are more likely to be present in the gastric wall than the mucosa; clear visualization of the wall is essential for accurate frozen section interpretation. Foci of increased cellularity or single infiltrating cells with hyperchromatic irregular nuclei and cytoplasmic vacuoles are typical of poorly differentiated carcinoma (Fig. 3.10a, b). Other features, such as desmoplasia, nuclear hyperchromasia, or perineural/lymphovascular invasion are helpful diagnostic features when present. Neoadjuvantly treated tumors may feature acellular mucin pools and fibrosis, although the presence of acellular mucin alone does not indicate a positive margin. Intraoperative consultation may also be obtained when peritoneal nodules or liver lesions are detected during gastric resection. Poorly differentiated carcinoma cells can be inconspicuous and simulate inflammation, especially when present in peritoneal soft tissue (Fig. 3.11). The presence of a desmoplastic response around clustered cells with more cytoplasm than would be expected of inflammatory cells is a helpful clue, especially if cytoplasmic mucin is present.

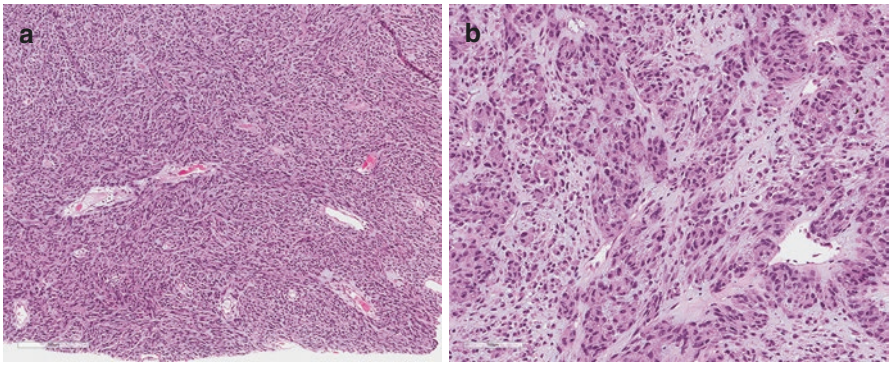

Fig. 3.8 Gastrointestinal stromal tumors of the esophagus tend to be highly cellular neoplasms (**a**). They may contain spindled or epithelioid cells with variably myxoid stroma (**b**)