

# Chapter 2

## Technical Considerations of EUS and EUS-FNA

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### Echoendoscopes

Echoendoscopes have a transducer mounted in front of the optic lens and are available in two different designs, radial and curvilinear, depending on the orientation of the transducer (Figs. 2.1, 2.2).

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FIG. 2.1. Echoendoscope with radial transducer. (Olympus GF-UE 160, Olympus America, Center Valley, PA)



FIG. 2.2. Echoendoscope with linear transducer. (Olympus GF-UC140P, Olympus America, Center Valley, PA)

A radial scanning echoendoscope produces a 360° real-time view perpendicular to the shaft of the echoendoscope (Fig. 2.3). A linear-array instrument produces a real-time, sector-shaped image parallel to the shaft of the echoendoscope (Figs. 2.4, 2.5). This imaging plane with the linear instrument allows for visualization of the entire length of a needle, which is advanced through the working channel of the echoendoscope whereas a radial device only displays a cross-section of the needle (a small spot) where the needle crosses the imaging plane (Fig. 2.3b). For this reason, virtually all endoscopic ultrasound fine-needle aspiration (EUS-FNA) is performed using linear-array instruments. For colorectal EUS, rigid probes for the rectum and flexible forward-viewing echocolonoscopes are available. Doppler (color) ultrasound allows for visualization of blood flow in vascular structures. Acoustic contact of the ultrasound (US) probe and the wall of the gastrointestinal (GI) tract are typically obtained through a water-filled balloon that surrounds

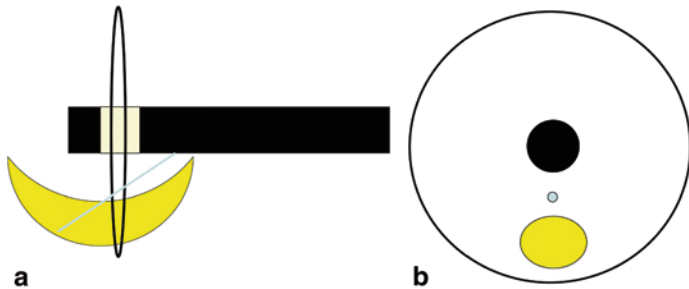


FIG. 2.3. A radial scanning echoendoscope produces a 360° real-time view perpendicular to the shaft of the echoendoscope (*B* shows expected ultrasound image with scope positioned as in *A*)



FIG. 2.4. A linear-array instrument, the preferred choice for fine-needle aspiration (*FNA*), produces a sector-shaped image in a plane parallel to the endoscope. This allows visualization of the entire length of a needle, which has been advanced into the banana

the probe or less frequently by filling the GI lumen with water. Samples can be obtained effectively from small lesions irrespective of the organ affected. Tumors <5 mm in diameter can be detected and sampled by the use of high-resolution echoendoscopes.

## Needle Type and Caliber

Several manufacturers, including Wilson-Cook, Boston Scientific, Olympus, and Medi-Globe, produce commercially available specialized needle assemblies for use with EUS. Our gastroenterology

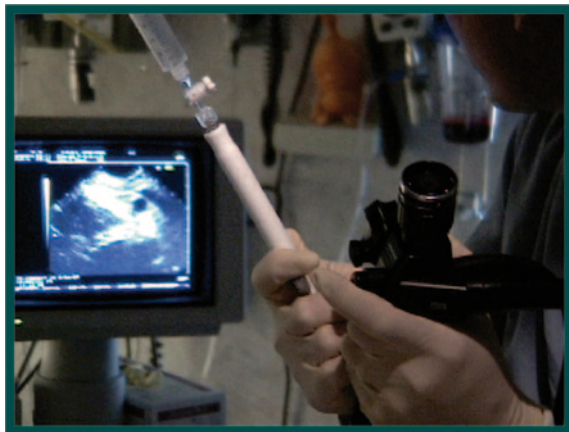


FIG. 2.5. The real-time image is visualized on a sector-shaped sound field on a computer monitor

team typically uses 19-, 22-, and 25-gauge Wilson-Cook needles with an excursion of up to 8 cm beyond the distal end of the 125-cm-long echoendoscope. A 25-gauge needle is routinely used for the sampling of most lesions and produces sufficient quantities of minimally bloody specimens. When needed, a 19-gauge needle is used for obtaining tissue fragments from suspected stromal tumors and for performing immunohistochemical (IHC) stains in cell-block slides. About 22- and 19-gauge needles may be used for draining of fluid collections and sampling of mediastinal lymph nodes with sclerosis. Ancillary studies for lymphoma work-up are performed successfully in aspirates, with use of a 25-gauge needle. The use of a 19-gauge Trucut needle may result in more frequent complications (i.e., infection and bleeding) and may not increase the accuracy, particularly when mediastinal lesions are evaluated; however, the approach does appear safe and accurate for the sampling of intestinal wall and abdominal masses.

## Role of the Endosonographer

EUS identification, staging, and FNA, particularly of pancreatic masses, are technically challenging and require special training. Accuracy is operator dependent and correlates with experience. EUS competence is difficult to achieve even for advanced endoscopy trainees. The current recommendation of the American Society for Gastrointestinal Endoscopy (ASGE) for EUS competence is 150 EUS procedures; however, this number may not be sufficient and most trainees may require 250 or more. Factors that influence this variable rate include cognitive and technical skills of the operator and station/organ examined, i.e., esophagogastric versus pancreatobiliary imaging; other factors are availability of cases in the trainer center and continuous access to perform EUS exams. Ideally, EUS-FNA competence starts once EUS competence is reached. EUS-FNA sampling accuracy has been shown to increase from 33 to 91% following a 2-month period of formal mentored training. The ASGE recommends a minimum of supervised 50 EUS-FNA procedures, 25–30 of which should be aimed to pancreas masses to reach the level of competence. The ASGE recommendation of 25–30 supervised EUS-FNA procedures for the diagnosis of pancreatic adenocarcinoma is supported by a study showing that there was a significant increase in the sensitivity of EUS-FNA diagnosis of pancreatic cancer, which plateaued at 80–90% after 30 procedures. EUS-FNA cytology interpretation errors during the initial learning phase are primarily due to inadequate specimens.

## The Aspiration Technique

The needle selection should be based on the physical characteristics and anatomic location of the target lesion.

### a) Solid lesions

One of the basic principles of aspiration of solid lesions is that the thicker the needle is and the more negative pressure applied, the more bloody and diluted the sample can be. Therefore for most

cases, the tendency is to use one of the smallest caliber needles available: a 25-gauge needle.

A 25-gauge needle is usually initially utilized in most solid lesions. It is better to have the stylet in place completely occluding the needle lumen until the target lesion is reached. Having the stylet occluding the needle lumen before reaching the target tumor avoids carryover of nonlesion material/contaminants. These contaminants include gastrointestinal mucosal epithelial fragments, fluids from the lumen, or any tissue fragments from organs trespassed with the needle before reaching the tumor. These nondiagnostic elements cause marked dispersion of the diagnostic material, which gets diluted, swollen, and blurred by fluid or widely scattered between numerous non-lesional tissue fragments making it more difficult to interpret.

Once the stylet is completely removed, it is more useful to do the aspiration without suction (the cellular material gets in the needle lumen by the action of capillary forces due to the fast movement of the needle within the lesion). The use of a 25-gauge needle allows the operator to have a “feel” of the consistency of the lesion. On occasions when there is no too much physical distance to move the needle within the lesion or when after the first pass a very hard lesion is felt, application of negative pressure with a syringe is recommended.

When the needle is in the lesion, it is recommended to move it back and forth as fast as possible within the target lesion. Keeping the needle still or longer than 5–10 seconds (depending on Doppler examination) within the lesion often causes the specimen to clot or be too bloody resulting in “sausages” of blood and fibrin obscuring the diagnostic elements, which are difficult to smear onto a glass slide and interpret on cytological rapid on-site interpretation. When clotting occurs, attempts to make smears should be avoided and the material should be placed in fixative and submitted to the pathology laboratory to be processed as a cell block.

The consistency of the lesion felt by the operator who is moving the needle is also important in deciding the next step. For example, in gastric wall lesions, a soft consistency generally narrows the differential diagnosis to lipomas, neuroendocrine tumors, or heterotopic pancreatic tissue. Gastrointestinal stromal tumors (GISTs), Schwannomas, and leiomyomas are generally firm rather than soft.

Solid masses for practical purposes could be approached in three ways based on their consistency: hard, gritty or rubbery, and soft.

**Hard masses:** Most of these lesions are associated with a very fibrotic or desmoplastic stroma. In these cases, the lesion “grabs” the needle and it is difficult to move the needle within the lesion; not uncommonly the needle gets bent because of the attempts to move the needle fast. The best approach is to use a 22- or a 19-gauge needle and perform the FNA applying suction. The smears are generally not very cellular but are often diagnostic. Several passes might be required. It is advised to try as much as possible to sample different areas of the mass when diagnostic material is not obtained with the first attempt. The use of a 19-gauge needle is recommended when GIST is suspected to obtain material for a cell-block preparation and perform immunostains and molecular tests in the paraffin-embedded material.

**Gritty or rubbery masses:** These lesions could be sampled using a 25-gauge needle, with or without suction. The movement of the needle within the lesion has to be as fast as the size and location of the tumor allow it. Generally, 1–3 passes provide enough diagnostic material. It is important to keep in mind that when additional studies might be required (e.g., metastatic carcinoma of unknown origin, metastatic lung adenocarcinomas where epidermal growth factor receptor (EGFR) testing might be requested, metastatic colorectal cancer where *KRAS* gene mutations are often needed, etc.), additional passes up to the discretion of the cytopathologist and interventional gastroenterologist are often required.

**Soft masses:** These lesions are usually sampled with a 25-gauge needle without suction. After the first pass when the tumor type is identified, subsequent passes are triaged according to the ancillary test required (e.g., flow cytometry for lymphomas or cell-block material for immunostains).

#### b) Cystic lesions

The larger, stiffer, and more awkward 19-gauge needle is reserved for aspirating/draining larger cysts as it allows for a faster procedure and the aspiration of viscous contents. It may also be used for obtaining tissue fragments from the cyst wall.

Some needles come with a stylet whose tip is beveled to the needle tip whereas others come with a protruding ball-tip stylet. The ball-tip version might protect the endoscope channel should the needle be deployed accidentally. In general, the ball-tip stylet must be withdrawn by about 1 cm before puncture in order to “sharpen” the needle. Following puncture, the stylet is pushed in to extrude any plugs of extraneous tissue. A prospective trial comparing FNA with and without stylet tried to answer the question of whether the use of the stylet is needed or not. The general conclusion was to use the stylet during the first pass and do the others without stylet. In our practice, we still use the stylet for all passes.

It is paramount to remember that the use of thinner needle, less dwelling time in vascular lesions, and no or minimal suction yields less bloody but diagnostic samples.

## Number of Passes

In general, the number of passes is inversely proportional to the experience of the endosonographer and the presence of the cytopathologist during the procedure. Without a cytopathologist in attendance, 5–7 passes should be made for pancreatic masses and miscellaneous lesions, 3–5 for lymph nodes, and 2–3 for liver metastases to ensure a high degree of certainty for making a correct diagnosis (80% sensitivity and 100% specificity). However, these guidelines in preference to rapid on-site smear evaluation are associated with a 10–15% reduction in diagnostic accuracy, extra procedure time, increased risk, and need for an increased number of needles, at considerable expense. We must emphasize that rapid on-site evaluation of adequacy is a better means of assessing the number of passes needed to obtain diagnostic material than is adherence to a set number of needle passes.

## Role of the Cytopathologist

EUS-FNA cytology interpretation requires a high level of communication and trust between the endosonographer and the cytopathologist. The clinical and imaging data provided by the endosonographer are paramount for the cytopathologist final diagnosis. The cytopathologist should know the route that the needle travels to be aware of possible carryover of contaminants at the time of microscopic evaluation of the sample. Ultimately, it is the cytopathologist's role to tell the endosonographer when to end lesion sampling. This interaction is highly beneficial for patient management.

## Specimen Handling

Briefly, the step-by-step procedure is as follows:

1. The sample is deposited on a glass slide by the endosonographer. The use of the stylet or air flushing is recommended if the sample is from a solid lesion. If the sample is from a cystic lesion, use the stylet to deposit the first few drops and make smears (one DiffQuik® for rapid interpretation and one ethanol-fixed for later stain) or use liquid-based cytology solution. If the needle gets clogged with blood clot, use the stylet and place the material in a glass slide and then transfer it to a container with formalin for cell-block processing; do not attempt to make smears.
2. Visible tissue fragments are separated from the blood, "picked up" with the edge of another glass slide, and smeared onto one or more slides. This results in a concentrated specimen that can be examined immediately under the microscope for on-site evaluation.
3. Air-dried smears fixed with methanol are stained with the DiffQuik® stain for immediate reading; 95 % ethanol-fixed smears are stained with the Papanicolaou or H&E stain for later reading. One smear for each staining per pass is usually prepared.
4. Rapid on-site evaluation of the DiffQuik®-stained smear for specimen adequacy and preliminary diagnosis is encouraged.

5. Let the remainder of the bloody sample clot on the glass slide. Submit the blood clot in formalin for cell-block processing. The methodology is the same as for FNA of masses in superficial organs.
6. Additional passes are requested if indicated for additional purposes, i.e., cell block, cell marker analysis, ultrastructural studies, cytogenetics, cultures for infectious agents, molecular techniques, and research protocols.

## Rapid On-Site Evaluation

Rapid on-site smear evaluation (ROSE) by the cytopathologist optimizes the EUS-FNA diagnostic accuracy and minimizes the technique's insufficiency rate (i.e., sampling failure and poor handling/preparation of aspirated material), resulting in a reduced likelihood of complications for the patient. Thus, it is recommended that a cytotechnologist or cytopathologist be in attendance for all aspiration procedures to assess specimen adequacy and give a preliminary diagnosis which, in many instances, is the definitive diagnosis (Fig. 2.6).

ROSE increases the diagnostic yield and sensitivity to near or higher than 90%, and the specificity and positive predictive values to near 100%. However, it requires special training and can be time-consuming. A preliminary diagnosis may guide further clinical investigation or treatment, and it may determine whether ancillary studies are needed for a more specific diagnosis. It appears that lesion characteristics, aspiration site, or tumor type do not significantly influence the diagnostic results. Negative or nondiagnostic EUS-FNA interpretation requires correlation with clinical and/or imaging findings for a decision on further appropriate management. In general, it has been shown that a reduction in rate of nondiagnostic specimens is observed when ROSE is performed; however, the range of this rate is broad (as low as 2–36% and higher).

Other factors that influence diagnostic yield include experience of the gastroenterologist, number of passes, and sample processing. It has been suggested that when EUS-FNAs of pancreas masses are performed by experienced endosonographers, ROSE may not

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