Frozen section: An overview

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ABSTRACT

Frozen section is a specimen of tissue that has been quick-frozen, cut by microtome, and stained immediately for rapid diagnosis of possible malignant lesions. A specimen processed in this manner is not satisfactory for detailed study of the cells, but it is valuable because it is quick and gives the surgeon immediate information regarding the malignancy of a piece of tissue. Frozen: said of material kept at less than 0°C. Biological materials are frozen solid because of their high water content.

Key word: frozen section, tissue, quick frozen, Microtome, surgeon.

INTRODUCTION

Frozen section
A thin slice of tissue that is cut from a frozen specimen and is often used for rapid microscopic diagnosis section and a histologic section of tissue that has been frozen by exposure to dry ice.

The frozen section procedure is a pathological laboratory procedure to perform rapid microscopic analysis of a specimen. It is used most often in oncological surgery. The technical name for this procedure is cryosection (Gal AA, 2005).

The quality of the slides produced by frozen section is of lower quality than formalin fixed, wax embedded tissue processing. While diagnosis can be rendered in many cases, fixed tissue processing is preferred in many conditions for more accurate diagnosis. The intra operative consultation is the name given to the whole intervention by the pathologist, which includes not only frozen section but also gross evaluation of the specimen, examination of cytology preparations taken on the specimen and aliquoting of the specimen for special studies. The report given by the pathologist is usually limited to a "benign" or "malignant" diagnosis, and communicated to the surgeon operating via intercom.

Procedure:
The key instrument for Cryosection is the cryostat, which is essentially a microtome inside a freezer. The microtome can be compared to a very accurate "deli" slicer, capable of slicing sections as thin as 1 micro metre. The usual histology slice is cut at 5 to 10 micro metres. The surgical specimen is placed on a metal tissue disc which is then secured in a chuck and frozen rapidly to about −20 to −30 °C. The specimen is embedded in a gel like medium consisting of polyethylene glycol and polyvinyl alcohol; this compound is known by many names and when frozen has the same density as frozen tissue. At this temperature, most tissues become rock-hard. Usually a lower temperature is required for fat or lipid rich tissue. Each tissue has a preferred temperature for processing. Subsequently it is cut frozen with the microtome portion of the cryostat, the section is picked up on a glass slide and stained (usually with hematoxylin and eosin, the H&E stain). The preparation of the sample is much more rapid than with traditional histology technique (around 10 minutes vs 16 hours). However, the technical quality of the sections
is much lower. The entire laboratory can occupy a space less than 9-square-foot (0.84 m²), and minimal ventilation is required compared to a standard wax embedded specimen laboratory.

**Uses from frozen section**

The principal use of the frozen section procedure is the examination of tissue while surgery is taking place. This may be for various reasons:

1-In the performance of Mohs surgery - a simple method for 100% margin control of a surgical specimen.

2-If a tumor appears to have metastasized, a sample of the suspected metastasis is sent for cryo section to confirm its identity. If the tumor has metastasized, surgery is usually not curative, and the surgeon will choose a more conservative surgery, or no resection at all (Williams and Wilkins, 1999).

3-If a tumor has been resected but it is unclear whether the surgical margin is free of tumor, an intraoperative consultation is requested to assess the need to make a further resection for clear margins.

4-In a sentinel node procedure, a sentinel node containing tumor tissue prompts a further lymph node dissection, while a benign node will avoid such a procedure.

5-If surgery is explorative, rapid examination of a lesion might help identify the possible cause of a patient’s symptoms.

6-Rarely, cryo sections are used to detect the presence of substances lost in the traditional histology technique, for example lipids. They can also be used to detect some antigens masked by formalin (Zdamar et al., 2006).

7-The cryostat is available in a small portable device weighing less than 80 lb (36 kg), to a large stationary device 500 lb (230 kg) or more. The entire histology laboratory can be carried in one portable box, making frozen section histology a possible tool in primitive medicine (Wilson, 1905).

**Techniques of the frozen**

Good frozen section technique is learned gradually and only through experience. The resident should question the staff freely and attempt to observe all steps closely at first. He/she then should do as many things:

1. **Gross Tissue Examination:**
   This step is probably the most important step and unfortunately is one that many pathologists have not yet learned. The pathologist obtains gross details not from just looking at the tissue, but also from feeling it and cutting it, i.e., soft or gritty. The pathologist records all gross expressions, i.e., size, adhesions, weight, similar to the recording of microscopic features.

2. **Proper Communication with the Surgeons:**
   The intercom is located in the frozen section suite; the room where the surgeon is operating will be obtained from the Pathology Department secretary transmitting the request for frozen section. A list of operating room procedures appear the day before the planned surgery, and it is the responsibility of the resident and staff to be familiar with each case in advance. This means knowing what tissues have been removed previously, reviewing any previous diagnoses that our department has on file, and reviewing all previous slides on the patient. This is important because the present procedure may be related to previous ones. This "research" is the pathologist's equivalent of the medical history (Weiss et al., 2010).

3. **Embedding the tissue:**
   The selected piece of tissue is then placed on a metallic holder and must be oriented a certain way so that the future section will reveal proper spatial relationships, this orientation depends on the question being asked. Sometimes orientation is not important; at other times it is of paramount importance. The tissue is embedded in OCT mounting medium and is then placed either in cooled 2-methyl butane or the cryostat machine where it is properly frozen.

4. **Cryostat:**
   The machine, which cuts the tissue, is the cryostat. Certain things should be routinely checked in the operation of this machine:
a) Temperature:
The temperature should be at -20°F for most tissues. For tissues with a large fat component, -40°F is optimal. This temperature is critical for optimal sectioning. Too high, i.e., -10°F and the tissue will not stay frozen and firm and will not cut crisp.

• Too cold, i.e., -50°F and the tissue will crumble and become powder.

The Ideal tissue should cut like butter, smooth and in one piece.

b) Blade sharpness and angle:
The blade should be sharp and should be changed approximately once every 2 weeks. A dull blade cuts dull. Equally important is the blade angle. There is an optimal angle between blade and tissue:

• Too steep an angle and the tissue will crumble like it was too cold.
• Too shallow, then two things will happen. The section will alternately skip and not cut and then it will cut, but too thick.

5. Staining:
Once the tissue is on the slide it can be either air-dried or fixed in methanol. This depends on which staining procedure will be used. There are several stains available in the frozen section room and each has certain advantages. The choice of stain depends on what the pathologist is trying to demonstrate. The resident should practice all the stains and gain experience with their use.

6- Interpreting The Frozen Section:
The results of one's labor now come to a climax when the resident and staff sit at the double-headed microscope and discuss the slide and render a diagnosis.

Since rapid diagnosis takes precedence over everything else in the operating room, often times additional discussion and questions occur after the diagnosis has been rendered.

7. Controls:
In all science, controls are necessary. Since pathology is not an exact science, controls cannot be exact, but an attempt is made to check our frozen section accuracy. The tissue, which is frozen, is submitted for permanents and labeled "frozen section control." This should be kept separate from the other additional tissue submitted for permanents. In this way, the pathologist has a limited check on his frozen. If anything shows up on the permanents that is substantially different than the frozen, the surgeon or doctor taking care of the patient should be notified immediately. (Abbasi et al., 2006).

Advantages of frozen section biopsy
1-If more tissue is needed to make an accurate diagnosis, the surgeon is able to obtain an additional sample, avoiding a second operation.
2-If the tissue is determined to be cancerous and is amenable to surgery, the mass can be removed at that time.
3-If the tissue is determined to be benign (not cancerous), then the mass may not always need to be removed and the surgery can end.
4-The frozen section biopsy can help ensure that the mass being removed is the intended tissue for removal.
5-It can help ensure that the entire mass and its surrounding borders are removed.
6-It allows for the collection of proper tissue samples for further scientific research.
7-The surgeon and pathologist are able to collaborate to care for the patient.

CONCLUSION

The pathologist should always be conservative with frozen section, but accurate. A diagnosis of invasive breast carcinoma should be, in fact, invasive breast carcinoma with no hesitation if that is what the slide shows. Too many surgeons and too many pathologists imply an uncertainty in all frozen section and take the philosophy of waiting for permanents. If the frozen section is definite, and many are, a definite diagnosis should be made.

As in so many things, experience is no substitute for knowledge, but knowledge alone is no substitute for experience. It takes years of frozen section experience to become a good diagnostic frozen section pathologist, and the resident should not be discouraged at first, but instead be encouraged to ask the advice of the staff for help and assistance in the frozen section room.
REFERENCES