ROLE OF FROZEN SECTION IN SURGICAL PATHOLOGY

COMMON PROBLEMS

1. Specimen identification
2. Specimen orientation
3. Benign v/s malign
4. Artifacts
5. Cutting of the fat
6. Temperature regulation
7. Infectious specimen

During frozen section

1. Tissue selection and representative area from organ/tissue sent
2. Type of the tissue should cut at proper temperature
3. Time consideration
4. Proper communication with the operating surgeon

TECHNIQUES

- FROZEN SECTION:
  1. CRYOSTAT
     - Microtome in side the chamber
     - Microtome is in constant temperature control

  1. FREEZING MICROTOME
     - Tissue fixed separately
     - Microtome is not under temperature control

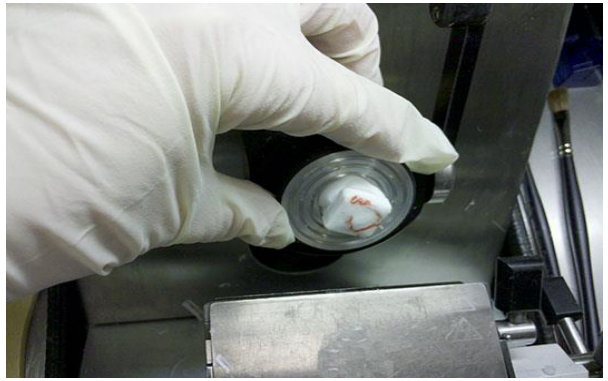
Cryostat

Sectioning (done in cryostat)

Once it froze,the tissue part is place onto a ‘chuck’ and fix it at chuck holder, to do 4 sections at 4 micron thickness.

1)

2)



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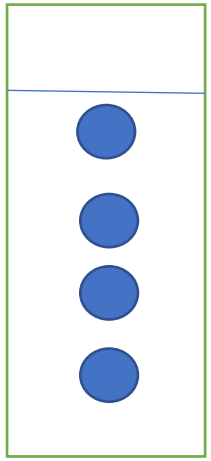


FROZEN SECTION EMBEDDING MEDIUM

FSC 22 Frozen Section Media

FSC 22® is a **water soluble** embedding compound used in **frozen sectioning**.

The compound bonds and encapsulates tissue specimens to the object holder for cryosectioning.

It provides excellent **sectioning consistency** with minimal curling of sections at a working temperature of -20 °C.

FSC 22 is available in clear or in light blue for better visualization of small specimens.

It is highly recommended for surgical pathology laboratories.

FSC 22 Mounting Media

It consist of poly ethylene glycol and poly vinyl alcohol.

What pathologist should know before frozen section

1. Availability of cryostat machine
2. Should received request form well in advance with,
    - Brief history of the patient
    - Clinical diagnosis
    - Radiological findings

1. What surgeon wants to know.?
2. What surgeons have sent?
3. How to contact them?

INDICATIONS

- Plan the work up of the specimen. Certain studies should be evaluated prior to fixation.
  - Cytogenetics
  - Flow cytometry
  - Special stains
- When tissue for banking may need to be sampled.

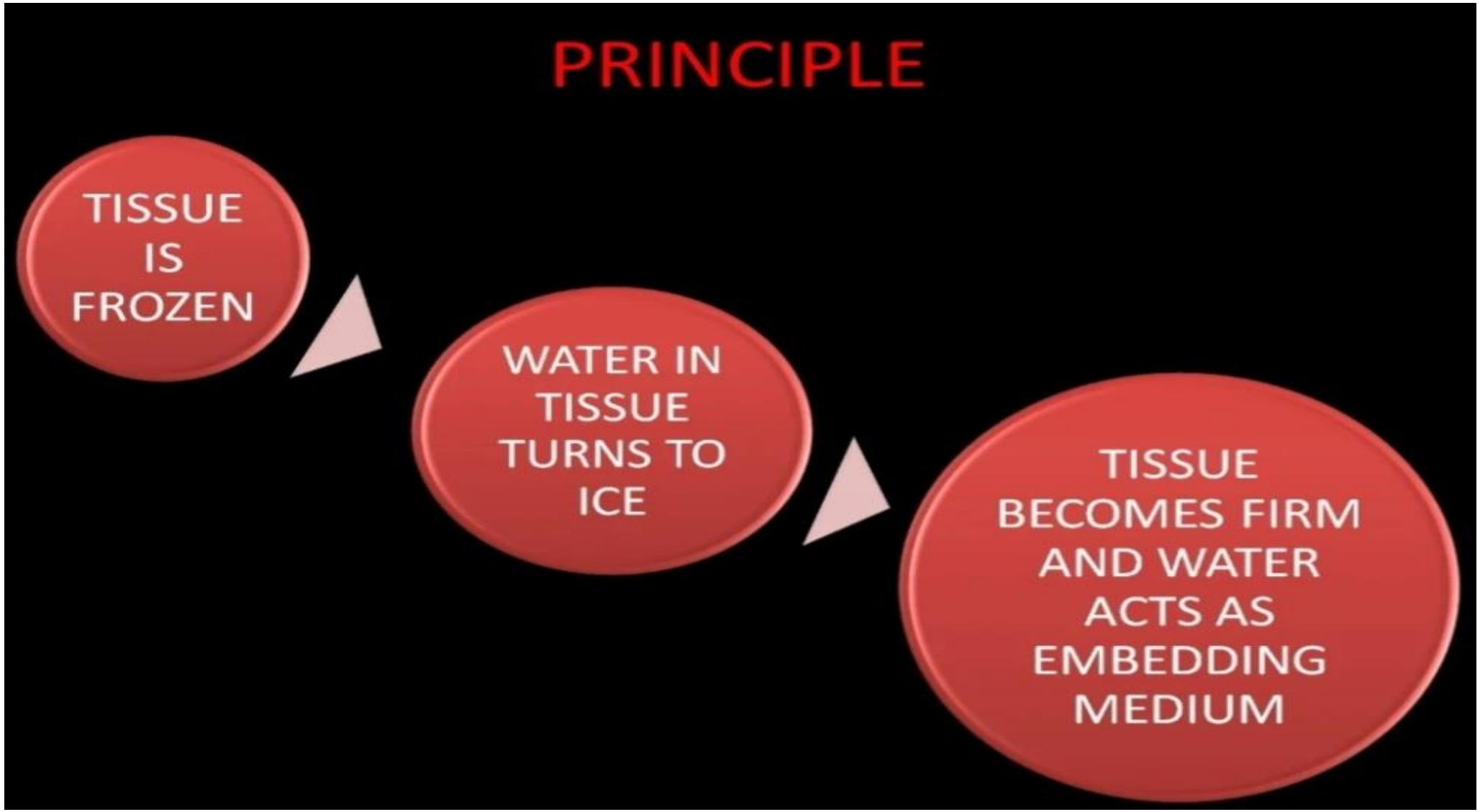
MARGINS

- Technically difficult
- Multiple section from the edge of the surgical section is in common practice
- Margins of Head and Neck tumors may be particularly very difficult

IDEAL TEMPRATURE CHART

TABLE 1.1 The Ideal Temperature for Cutting Cryostat Sections Varies with Amount of Matrix and Lipid Content of the Tissue

	Suggested Temperature for Sectioning				
Organ	−30	−25	−20	−15	−10
Brain				+	+
Fat	+	+			
Cartilage				+	+
Liver				+	+
Skin		+	+		


PRINCIPLE
TISSUE IS FROZEN
WATER IN TISSUE TURNS TO ICE
TISSUE BECOMES FIRM AND WATER ACTS AS EMBEDDING MEDIUM

FIXATION

- For fast result, it requires to cut sections of frozen but unfixed tissue.
- 'Short period of fixations before freezing makes section cutting easier and yields better sections' (especially for fat/mucin containing tissue)
- Ideal fixative is 10% formal-saline for 10 mins at 60° C
- Important freezing agents,
  - Co2 gas (-60)
  - Solid Co2 (-70)
  - Aerosol spray (-50)
  - Thermo-electric cooling(-40)
  - Refrigeration

SELCTION OF TISSUE

1. Tissue sample should represent the specimen
2. Should not contain any necrotic area
3. 3-4 mm is ideal section
4. Sample once collected should be frozen immediately

CUTTING

- Any microtome (In Our Lab?)
- Tissue should be in frozen state while sectioning.
- 'COLD KNIFE'- is a device where specimen and microtome knife cooled but sectioning done at room temp.

POINTS TO REMEMBER WHILE SECTION CUTTING

- Frozen section requires well oriented specimen with flat cutting surface with a sufficient rim of embedding medium.
- Fat should be the last thing to hit the blade or should hit the blade by itself.
- The most critical aspect of the tissue should be perpendicular or diagonal to the blade.

POINTS TO REMEMBER WHILE SECTION CUTTING

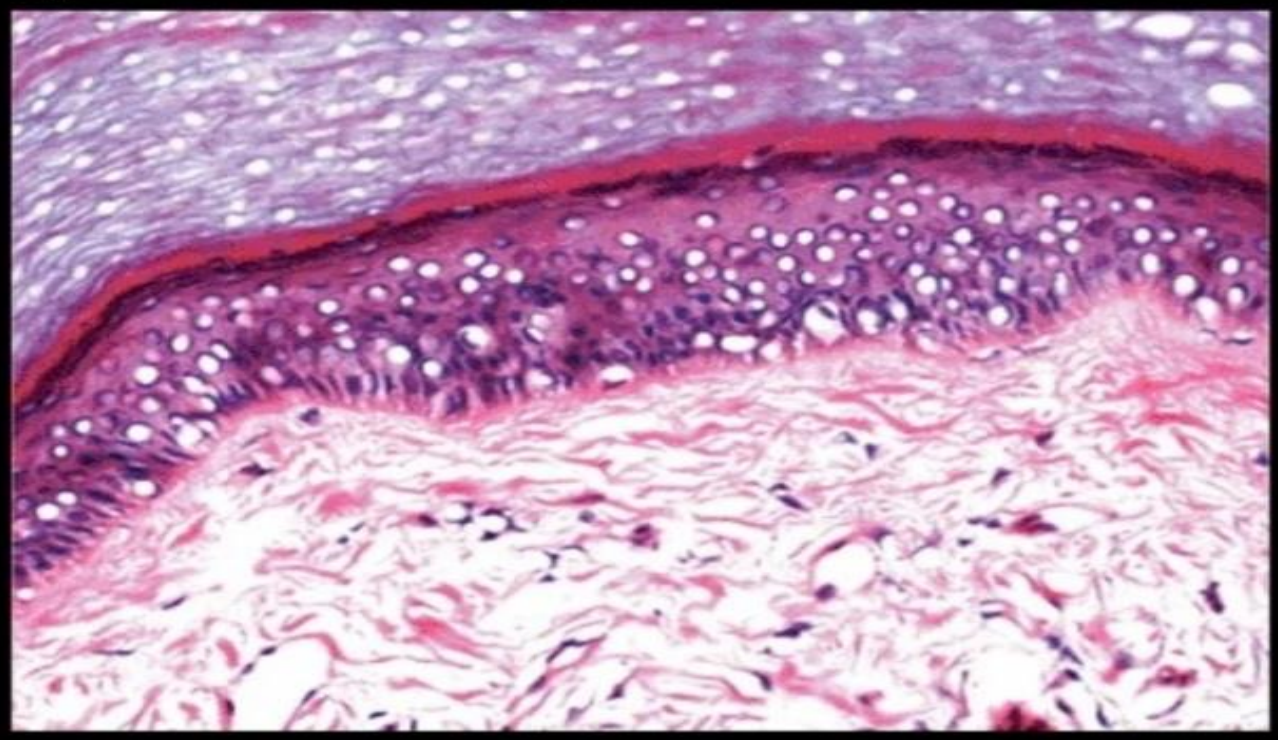
- Epithelial and mucosal lined tissues such as skin, GI, bladder, uterus and cervix should be oriented with the plane of the epithelium perpendicular to the blade.
- Trimming the block should be done to a depth at which the complete desired tissue face is available for the frozen section.
- The optimum cutting temperature of most of the tissue is -15 to -20° C
- Anti role plate placed 0.5 mm above the knife and it prevents sections from curling

STAINING

1. Rapid H&E Method
2. Toludine blue
3. Methylene blue
4. Methyl violet for amyloid
5. Oil red O for fat
6. PAS

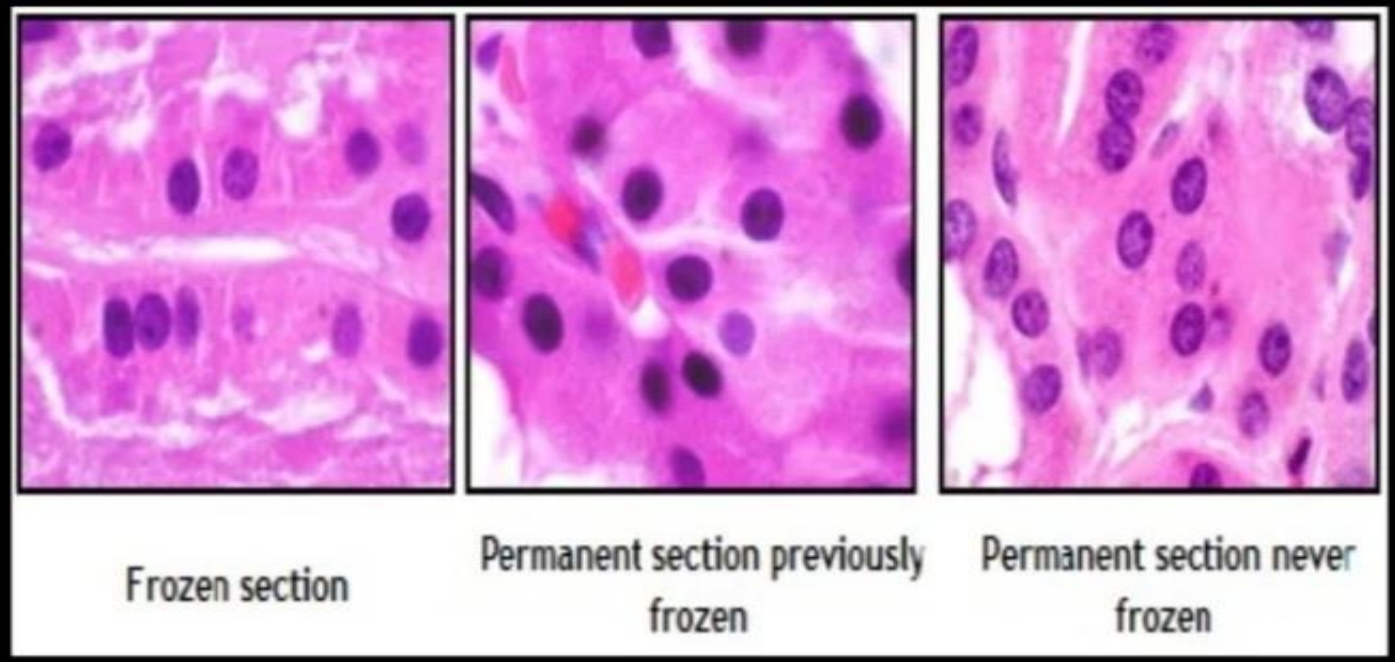
COMMON PROBLEMS - Artifacts

- Freezing artifact
  - It is a chemical property of water, that water will expand on freezing.



COMMON PROBLEMS - Artifacts

- Nuclear chromatin changes
  - The chromatin is more condensed and hyper chromatic.



IMPORTANT

- Reporting time: 20 minutes (In 90% of cases)
- Specimen should be sent fresh; without formalin.
- In some case the pathologist may make touch preparation of the received specimen.

INDICATIONS

- To provide a diagnosis that will allow the surgeon to make an intra-operative decision regarding further surgeries.
  - To avoid subsequent surgical procedure
  - To make primary diagnosis when pre-op diagnosis is not available
- Assess margins when additional excision to attain negative margins is an option.
- Assess adequacy of diagnostic tissue in a biopsy specimen from an open or a complicated procedure.

Disinfection of Cryostat

Cleaning done after each session

↓

Wear safety gloves

↓

Remove the blade from its holder

↓

Remove all the remnants of trimmed and cut tissues of previous frozen section cases

↓

Wipe with 1% Hypochlorite for 10 – 20 min

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Wipe Dry

↓

Spray/Wipe with 70% alcohol

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Within 10 min wipe with 100% alcohol

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Dry and close the lid

SUMMARY

- Frozen section are for intra-operative management decision making.
- Give the details as much required
- Do not give unnecessary detail or explaination. Report should be relevant to the surgeon's query.
- Ask for the more information if required.
- TAT should be maintained.


THANK
YOU