Once your samples are collected, they need to sectioned and mounted onto an MMI MembraneSlide prior to laser microdissection. The procedure for cryosectioning is relatively simple and can be performed rather quickly. Observe the cryostat manufacturer’s and/or institutional safety guidelines.

Materials:

- Frozen embedding media
- Empty slide box
- Dry ice
- Cryostat and disposable blades
- Specimen mount
- MMI MembraneSlides (PN: 50102, 50103)
- Poly-L-Lysine
- Gelatine
- Agarose

Note: Frozen sections must be either fixed immediately, kept in the cryostat until all sectioning is completed, or stored at -80°C (or dry ice) immediately to prevent thawing. The frozen section slide must be on dry ice at all times to avoid degradation of molecules of interest in downstream analysis (RNA, DNA, Protein)

Method:

Cryostat Preparation:

1. Clean the cryostat with proper cleaning agents
2. Insert new sterile blade
3. For fresh frozen and frozen embedded block specimens, use a drop of frozen embedding media to adhere the sample to the specimen mount - make sure you are adhering the specimen in the correct cutting position (cutting surface should be parallel to the blade)
4. Allow frozen block with specimen to equilibrate to the cryostat temperature for about 20 minutes
5. Place empty slide box in cryostat to equilibrate to cryostat temperature (-20°C).

Use of Membrane Support Plates:

To prepare the slide thaw the section by placing the membrane support plate at room temperature in the indentation of the membrane slide. Fix the section on the flat side of the slide

Coating of Slide to increase adherence (optional):

Additional coatings (poly-L-Lysine, Agarose, or Gelatine) are recommended for tissues that are fatty, hard, fibrous, or contain cartilage/bone. The most common method is coating the mmi Membrane Slide with 0.1% poly-L-Lysine solution. Incubate the slides for 1 hour at room temperature or 30 minutes at 37°C. Gelatine or agarose can be used as well by preparing a 0.01% solution and incubating the slides using the above guidelines

Leading the way in Micromanipulation

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