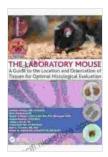
Guide to the Location and Orientation of Tissues for Optimal Histological Analysis

Histological analysis is a fundamental technique in biomedical research and clinical diagnostics, providing microscopic insights into tissue structure and pathology. Proper localization and orientation of tissues are crucial for accurate and meaningful histopathological interpretation. This guide provides comprehensive instructions on the best practices for tissue handling, embedding, and sectioning to ensure optimal histological analysis.

Tissue Collection and Handling

1. **Immediate Preservation:** Tissues should be immediately preserved in a suitable fixative upon surgical removal or biopsy. Formalin (10% neutral buffered formalin, NB) is the most commonly used fixative and provides excellent preservation of tissue morphology. 2. **Proper Fixation:** Tissues should be immersed in fixative in a volume ratio of at least 10:1 (fixative:tissue) and processed within 24 hours to avoid over-fixation or autolysis. 3. **Tissue Orientation:** If possible, note the tissue's orientation at the time of collection. This information will guide subsequent embedding and sectioning. 4. **Tissue Trimming:** Remove excess fat and connective tissue around the tissue to facilitate processing.



The Laboratory Mouse: A Guide to the Location and Orientation of Tissues for Optimal Histological

Evaluation by Lee McIntyre

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Tissue Embedding

1. **Tissue Dehydration:** Tissues are dehydrated through a series of graded ethanol solutions (e.g., 50%, 70%, 95%, and 100%). Ethanol removes water and facilitates wax infiltration during embedding. 2. **Wax Infiltration:** Dehydrated tissues are infiltrated with molten paraffin wax or other embedding media in a tissue processor. The wax fills the spaces left by ethanol and provides rigidity for sectioning. 3. **Embedding:** Infiltrated tissues are embedded in wax blocks using embedding molds. The orientation of the tissue should be maintained during this step if possible. 4. **Trimming and Sectioning:** Embedded wax blocks are trimmed to expose the tissue of interest. Thin sections (typically 5-10 micrometers) are cut using a microtome and placed on glass slides for staining and analysis.

Tissue Sectioning

1. Longitudinal and Cross-sectional Planes: Tissues can be sectioned in two main planes:

a. **Longitudinal plane:** Sections are cut parallel to the tissue's long axis, providing an elongated view.

b. **Cross-sectional plane:** Sections are cut perpendicular to the tissue's long axis, providing a cross-sectional view. 2. **Orientation Markers:** If the tissue's orientation is not known, orientation markers (such as suture threads or India ink) can be placed on the tissue before embedding. 3.

Serial Sectioning: Serial sections are a series of successive sections taken from the same block, allowing for three-dimensional visualization of the tissue.

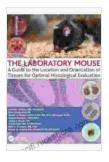
Special Considerations for Specific Tissues

1. Eye: Embed in paraffin, section at 4-6 micrometers, and stain with hematoxylin-eosin (H&E) or Masson's trichrome. 2. Brain: Embed in paraffin or frozen sections, section at 5-10 micrometers, and stain with H&E or Nissl stains. 3. Lung: Inflate with formalin or inflate and embed in low-melting-point agarose, section at 5-10 micrometers, and stain with H&E or elastic van Gieson stain. 4. Liver: Embed in paraffin or frozen sections, section at 5-10 micrometers, and stain with H&E or elastic van Gieson stain. 4. Liver: Embed in paraffin or frozen sections, section at 5-10 micrometers, and stain with H&E or Masson's trichrome. 5. Kidney: Embed in paraffin, section at 3-5 micrometers, and stain with H&E or periodic acid-Schiff (PAS) stain.

Troubleshooting

1. **Tissue Crumbling:** Over-fixation, incomplete dehydration, or inadequate wax infiltration can cause tissues to crumble during sectioning. 2. **Tissue Folding:** Improper tissue handling or orientation during embedding can lead to tissue folding, obscuring the true tissue structure. 3. **Tissue Loss:** Improper sectioning techniques or a dull microtome blade can result in tissue loss and poor-quality sections. 4. **Orientation Issues:** If the tissue's orientation is not known or maintained during embedding, it can be difficult to interpret the histological sections accurately.

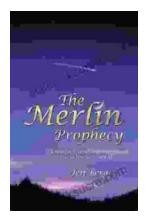
Proper tissue location and orientation are essential for optimal histological analysis. By following the guidelines outlined in this guide, researchers and clinicians can ensure accurate interpretation of tissue structure and pathology, leading to better diagnoses and treatment outcomes. It is important to note that special considerations may be necessary for specific tissues, and consultation with an experienced histologist is recommended in such cases.



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