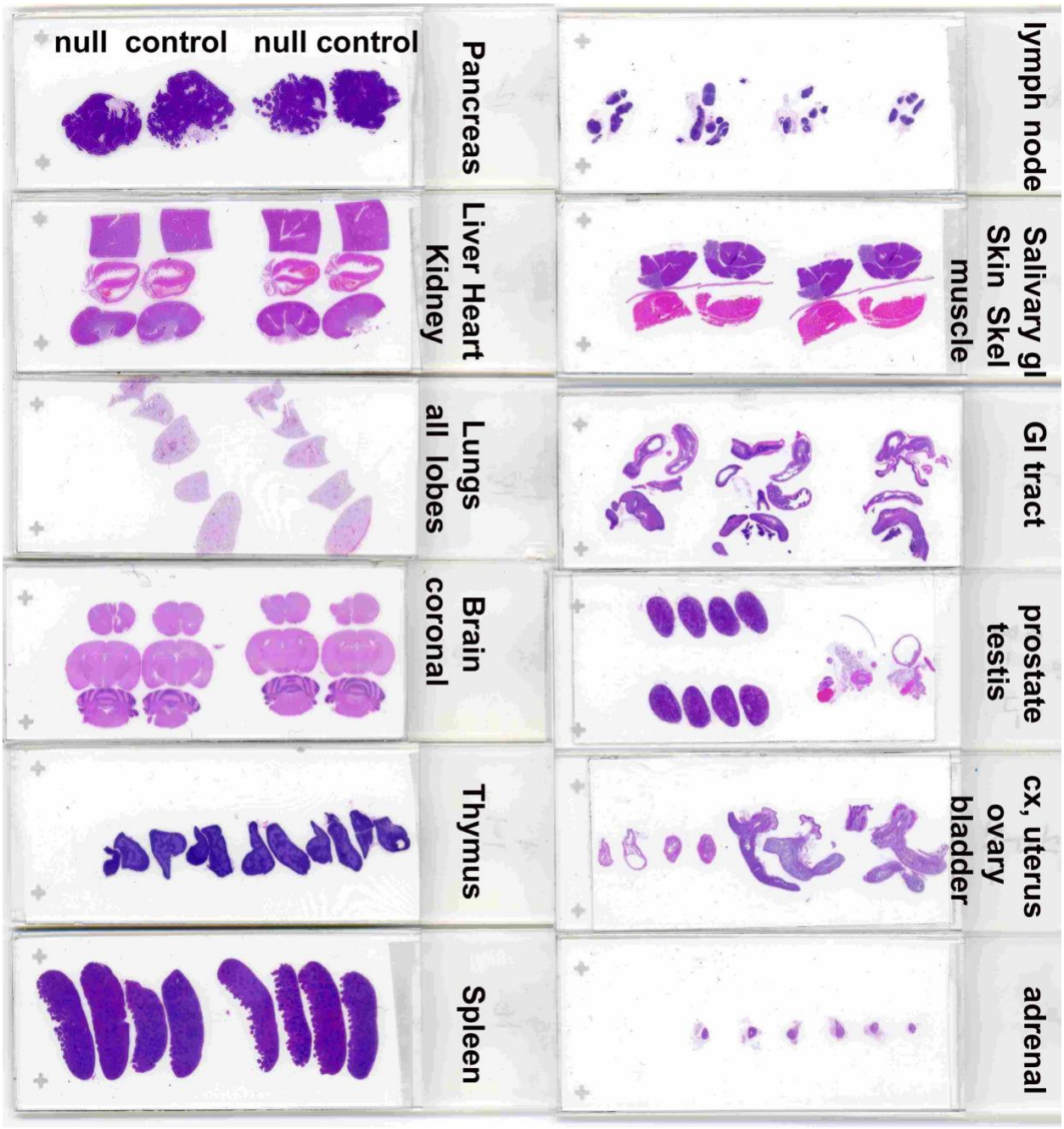
Summary of Optimum Handling of Mouse Tissues

When tissues are removed from the body, the process of autolysis occurs rapidly. In order to preserve tissue morphology for histopathologic examination, it is important to immediately FIX (to process and embed in wax) OR FREEZE tissues promptly after removal from the body.

Mouse Organs on Slides



Optimum Handling of Mouse Tissues

- Frozen sections are best used for immunohistochemistry assays because morphologic detail is usually not well preserved. See "**Freezing and Fixing Tissue**" in the Protocols section of the website.
- **Brain**: Prior to opening the skull to remove the brain, first perfuse the animal with PBS and then with 10% buffered formalin. Perfusion is best for examination of the brain, as it helps to avoid introducing artifacts during removal from the skull.
- **Skin, Pancreas, Spleen, Thymus**: Flatten between sponges in labelled cassettes and fix for processing, embedding, and sectioning. After processing, the skin is carefully embedded on edge.
- **Kidneys, Liver, Heart**: Halve the kidneys and place flat surface down, with a square segment of liver, and the two halves of the heart in labelled cassettes and fix for processing, embedding, and sectioning.
- **Ovaries/Testes, Adrenals, Lymph Nodes**: Place in labelled cassettes and fix for processing, embedding, and sectioning. The testes need to be fixed in Bouin's solution for at least 6 hours before transferring to 70% alcohol in labelled cassettes. To generate good sections, these small organs are processed using a short protocol.
- **Gut**: The stomach and esophagus should be opened to allow fixative to penetrate and each half laid flat in a labelled cassette. The small intestine should be sampled with Peyer's patches included from different places. The openings at each end of the tube selected will allow fixative to penetrate. The colon must be opened and rolled before fixing, processing, embedding, and sectioning (see "**Preserving Organs for Histopathologic Examination**" in the Protocols section of the website).
- **Bones**: The bones need to be decalcified. If immunostains are needed on bone marrow, the bones should be decalcified in EDTA.
- **Tumor Samples**: In order for the fixative to work well, tumor samples should be sliced thinly and placed in labelled cassettes for fixing, processing, embedding, and sectioning.
- If **immunostains** are planned:
    a. **Do not fix for more than 24 hours** in 10% buffered formalin before proceeding to immerse in 70% alcohol in labelled cassettes for fixing, processing, embedding, and sectioning.
    b. Make sure the **positive and negative controls** are processed similarly. If tissue culture cells are the positive and negative controls, plan to grow up 100 million (10_8) cells, wash away media, and resuspend each pellet of cells in 10% buffered formalin for 24 hours. Then, centrifuge each set of cells into a pellet and embed in 1% agarose in water. Place the solidified agarose with the pellet of cells between sponges in labelled cassettes for processing, embedding, and sectioning. Two or more sets of cells may be sections onto slides for use as negative and positive controls in immunohistochemistry.
- **Adipose Tissue**: Frozen sections are difficult. The best morphology is obtained if the adipose tissue is enclosed between tissue cassette sponges and placed in properly labelled cassettes and fixed in 10% buffered formalin for 24 hours before being transferred to 70% alcohol for processing, embedding, and paraffin sections.