

NCSU-CVM Histology Lab

Research and Teaching Sample Submission Guidelines

Following these guidelines ensures your submission will be completed efficiently and with utmost regard to quality and reproducibility. You are welcome and encouraged (!) to make an appointment with laboratory staff to review protocols and discuss your project before it begins. For assistance, contact the Histology Lab Manager or laboratory staff 919-513-6390.

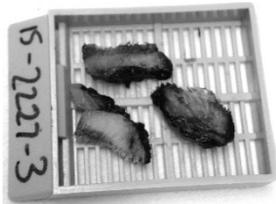
GROSS TRIMMING AND FIXATION

Untrimmed tissues are accepted when in an adequate volume of fixative. Please record the approximate date collected so we can ensure your sample is appropriately fixed. Tissues trimmed into cassettes should be submitted in the correct fixative or 70% ethanol in a sealed, spill proof container and accompanied by a submission form. Specimens not submitted into cassettes or incorrectly trimmed samples will incur an additional trimming fee. **Do not use blue, green or pink cassettes.** These colors are reserved for Diagnostic Specimens.

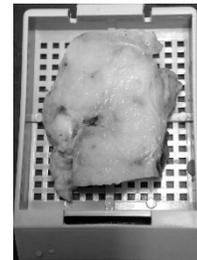
Tissues will be embedded in the orientation they are placed in the cassette. The surface which faces down (bottom of the cassette) will be microtomed at full-face unless otherwise indicated. Do **NOT** cram tissues into the cassette. The specimen requires room within the cassette on all sides to allow for fluid transfer during processing. Use a cassette with the appropriate-sized holes to promote fluid transfer. Your tissue should not be at risk of floating out of the cassette, however use the largest sized hole which still safely contains your sample. Cassettes **MUST** be labelled with either pencil or histology marking pens. Sharpies or Sarstedt lab markers are NOT acceptable, ethanol steps during paraffin processing remove all traces of their ink. Your label must be **legible and concise** for transcription on the slide. We recommend you include a printed spreadsheet list of each cassette label with your submission form for quality control.

For Example:

YES



NO



Tissues containing bone or calcified areas must be separated from soft tissue samples, submitted in a separate container of fixative and labelled as such. The lab will decalcify them in the appropriate decalcification solution before they can be processed.

If you are requesting IHC or expect to perform IHC in the future, it is best to standardize your protocol for trimming (uniform tissue size) and interval of fixation (10% NBF is preferred). Variation in sampling and fixation protocols among samples results in variable IHC staining, tissue adhesion and makes comparative studies difficult to interpret. The penetration rate of 10% NBF is dependent on tissue thickness, temperature, and fixative volume. For a typical sample 5-10mm, prolonged exposure (greater than 72hrs) to formaldehyde is not recommended, however, inadequate fixation (less than 24hrs) will not preserve the tissue well enough to withstand the rigors of IHC staining. Our IHC protocols have been validated to control tissues processed here under these conditions. If your samples have been fixed under different conditions, we cannot guarantee desired results. Tissues which are trimmed into cassettes AND adequately fixed can be transferred to 70% ethanol for storage until processing can be completed. Simply rinse cassettes with water before transfer to ethanol.

SECTIONING AND STAINING

Step sections = two or more sections, taken at a known interval apart

Serial sections = two or more sections, taken consecutively or sequentially

A routine H&E = a single, 5um section on a regular glass slide, stained with Hematoxylin and Eosin