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Lightsheet Tissue Intake - Photodocumentation and Tracking

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Optical Clearing of Tissue Human BioMolecular Atl...

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Protocol status: Working We use this protocol and it's working

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Abstract

In order to track tissue position and orientation each sample is photographed when received for lightsheet microscopy (Figure 1, A). Large samples (>1 cm³), such as the spleen in Figure 1, are cut into 2 mm sections (Figure 1, B) to facilitate tissue clearing, staining, and three-dimensional imaging. These tissues are batch processed and withdrawn from the lipid clearing pipeline as needed (Figure 1, C); at this time a novel identifier is assigned to the sample.

Guidelines

Photodocumentation, in our pipeline, occurs following tissue-hydrogel polymerization and multiple washes in PBS. This serves to remove trace fixatives for safer handling.

Materials

MATERIALS

🔀 50 ml conical tubes

Sourceps (tweezers), 12.5cm, Blunt End Bio Basic Inc. Catalog #FC003.SIZE.1

🔀 Razor blades Fisher Scientific Catalog #12-640

X Hexagonal Polystyrene Weighing Dishes Thermo Fisher Catalog #02202103

Safety warnings

Wash tissues of any fixatives, or other dangerous chemicals, prior to manipulating for photodocumentation.

- 1 Photograph whole tissue; capture from various angles if possible. A dissection ruler or grid should accompany each photograph.
- 2 Using a razor blade and forceps slice the tissue into sections approximately 2 mm wide. Try to slice in a continuous motion, while applying a gentle downward force.
- 3 Set aside tissue section and repeat Step 2 as needed. Maintain or keep track of slice order and orientation.
- 4 Photograph the sequential 2 mm slices, again using a dissection ruler or grid.
- 5 Retain all slices within one container for subsequent processing (lipid removal).