

Induction Checklist: Safe Use of the Cryostat (Leica CM3050S)

Trainee Information

- Name:
- Date:
- Trainer Name:
- Position (e.g., researcher, student):

Safety Protocols

- Ensure proper Personal Protective Equipment (PPE) is worn: gloves, lab coat, safety glasses.
- Understand the cryostat locking and unlocking mechanism (holding the key symbol to unlock).
- Safe handling and disposal of cryostat blades into a biohazard sharp container.
- Awareness of cryogenic temperatures and frostbite prevention measures.
- Cleaning and decontaminating the cryostat after use (use of 70% ethanol).
- Take regular breaks during use to prevent fatigue and ensure safe operation.
- Stay hydrated and maintain proper ergonomic posture to reduce fatigue and improve focus
- Emergency procedures and first aid in case of blade or cold injuries.

Cryostat Setup

- Reserve the cryostat ahead of time in the histology record book.
- Ensure the cryostat is clean before use
- Set cryochamber and object temperatures (CT and OT) according to the type of tissue.
- Equilibrate cryostat chamber and materials for at least 30 minutes before sectioning.
- Verify that the section thickness is correctly set
- Lock the handwheel and check the blade clearance angle (5°) before operation.
- Use OCT compound for frozen tissue block preparation and ensure correct orientation.
- Insert the disposable blade and cover it with the anti-roll plate before starting.

Frozen Tissue Block Preparation

- Correctly orientate fresh frozen tissue in a cryomould with OCT freezing medium.
- Ensure the OCT turns white before attaching the tissue block to the chuck.
- Clamp the frozen tissue block and adjust the specimen holder position.

Trimming Procedure

- Set trimming thickness.
- Ensure the handwheel is unlocked and trim the tissue block using a continuous forward motion.
- Remove unwanted trimmed sections (debris) with a large brush.
- Lock the handwheel after trimming is complete.

Sectioning Procedure

- Align the anti-roll plate with the blade for sectioning.
- Slowly and uniformly rotate the handwheel forward to produce an optimal section.
- Mount each section immediately onto a charged microscope slide to prevent curling.
- Gently flatten the section with a stiff brush before mounting onto the slide.
- Ensure each slide is pre-labeled and sections are evenly spaced on the slide.

Post-Use Clean-Up

- Remove all frozen tissue blocks and clean the cryostat chamber with 70% ethanol.
- Discard used blades into a biohazard waste sharp container.
- Empty the waste tray and dispose of debris in a biohazard bag.
- Decontaminate forceps and brushes by soaking them in diluted Trigene for 30 minutes.
- Wipe clean the cryostat surfaces (use dry tissue for anti-roll plate glass).
- Ensure it is locked before leaving.

Trainee Competency Confirmation

- Safe operation of the cryostat demonstrated.
- Tissue sectioning quality verified.
- Proper clean-up procedure followed.

Sign-Off

Trainer Signature:

Trainee Signature: