

**LOCAL JOINT HEALTH AND SAFETY COMMITTEE
DEPARTMENT OF BIOMEDICAL SCIENCES**

STANDARD OPERATING PROCEDURE

for Rm: 3658 Bld: Ext. 3

1. SAFE USE AND MAINTENANCE OF THE CRYOSTAT INSTRUMENT

Effective Date: June 30, 2002

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Purpose: To promote the safe and proper use of the cryostat during sectioning of frozen tissue

Approvals Required: Faculty Supervisor, Local JHSC, EHS

2. DEFINITIONS:

The **CRYOSTAT**, also called a freezing microtome consists mainly of a microtome contained within a refrigerated cabinet designed to operate between -5°C to -40°C. Recent models have external controls so one can operate them without having hands inside the cabinet. They also have an antiroll device which enables flat sections to be cut easily in the cold environment. Most components of the cryostat are made of stainless steel. During cutting, the stage holder assembly arm that holds the specimen stage goes up and down as you rotate the handwheel. At the end of the up stroke it moves forward an increment of what it was adjusted to (e.g. 8 microns). In this example, each time the cutting arm goes down, 8 microns of the tissue will hit the knife edge cutting it. In frozen microtomy, most tissue that is handled and manipulated is unfixed tissue.

It is imperative that one maintains a high degree of bacteriological discipline and be fully aware of the hazards of infectious materials. This microtome is **NOT** designed to be used for biohazards and other infectious materials.

3. REQUIREMENTS:

- a) OSHA Sections 25(1), 25(2), 27(2), 42.
- b) All users of the cryostat are required to undergo proper training in frozen sectioning with the technician in charge of the equipment or other personnel who have previous experience in frozen sectioning.
- c) All users must read the operators manual located in the room and must familiarize themselves with the parts of the cryostat and their function.
- d) All users are required to log in usage time and indicate the source of funding towards the applicable users fee.
- e) Equipment failure and other maintenance or problems with the equipment are to be reported immediately to the technician in

charge. Equipment and work area is to be left clean and in good order at all times.

- f) Users are required to bring their own tools and supplies (e.g. forceps, embedding compound, microscope slides, brush, kimwipes etc.)
- g) **Before using the Cryostat, familiarize yourself with the location of the FIRST AID KIT, the nearest PHONE and all EMERGENCY EXITS from this room and building.**

ADDITIONAL SAFETY PRECAUTIONS:

- a) **Wear a labcoat and non-latex gloves as basic protection.**
- b) Use a **surgical mask** to avoid exposure to lyophilized tissue dusts.
- c) Make sure cryostat is in the **LOCKED** position anytime you are not actually cutting (rest position is on locked)
- d) Avoid distraction while cutting.
- e) Avoid handling very cold metal surface (e.g. specimen stage) with bare hands. Use gloves or forceps.
- f) **Avoid using fingers to remove tissue from the knife.** Use a brush, forceps or microscope slides.
- g) If using liquid nitrogen or dry ice, read the SOP on Use of Cryogenics, the University of Guelph Safety Policy Manual, and the CSLT Laboratory Safety Guideline.

4. DESCRIPTION OF THE TASK:

- a) Adjust chamber temperature to -20°C , the usual cutting temperature for most tissue. It may take up to 30 minutes for the chamber temperature to reach -20°C .
- b) Ascertain that the handwheel is in the **locked** position before working inside the cryostat chamber.
- c) With the handwheel in the locked position, install specimen stage (complete with previously mounted tissue) into the specimen stage holder. Tighten specimen stage using clamp screw.
- d) Adjust angle of specimen stage holder by loosening specimen stage holder clamp and using specimen stage alignment grip, so that when cutting the block face of your tissue is not tilted but would drop as parallel as possible in relation to the knife edge. Tighten specimen stage holder clamp.
- e) With handwheel in the **locked** position secure knife to knife holder.
CAUTION: KNIVES ARE VERY SHARP AND MAY CAUSE DEEP FINGER CUTS.
- f) Now you can start cutting your tissue.
- g) When finished cutting, leave handwheel in **LOCKED** position. Remove all leftover samples for proper disposal in your laboratory or take to the incinerator in the Pathobiology Department. Clean up cryostat. Put disposable knives and other sharp objects in proper disposal containers designated for sharps. If you have to leave a

knife for future use inside the chamber please leave a **CAUTION NOTE** for others on the outside of the cryostat.

- h) After proper cleanup, adjust chamber temperature back to -10°C, turn knife temperature off and specimen holder temperature off.
- i) Leave all specimen stages inside the chamber at all times. Do not remove specimen stages.

5. CONTINGENCY PLAN AND REPORTING:

In case of emergency, call ext. 52000 for assistance 24 hours a day. In case of injury, seek first aid, inform your supervisor and complete an "Injury/Incident Report Form" available from the Administrative Assistant, Department of Biomedical Sciences **NO MATTER** how minor the injury may appear to be. This report is to be faxed to Environmental Health and Safety 824-0364 or you can phone ext. 53132 to make a report during working hours.

6. WASTE MANAGEMENT AND ENVIRONMENTAL RESPONSIBILITY:

All leftover tissue is to be disposed of properly through your own laboratory or brought to the incinerator, Department of Pathobiology. No biological specimen should be left in the Cryostat. All sharp objects e.g. hypodermic needles, broken glass, pipettes, microscope slides etc. are to be cleared away and disposed of properly in designated Sharps or By-Pass containers in your own laboratory. Refer to University Safety Policy 851.08.15 regarding disposal of sharp objects.

7. REFERENCES:

Instruction Manual for Leitz 1720 Cryostat (located in Rm. 3656)
Ontario Health and Safety Act and the CSLT Laboratory Safety Manual (both located at the Safety Desk, Rm. 2634, Biomedical Sciences.
University of Guelph Safety Policy Manual, - on line at www.uoguelph.ca/HR/ehs. Refer to Policies 851.08.05 (Cryogenics) and 851.08.15 (Sharps)

8. DISTRIBUTION OF COPIES:

Technicians, Graduate Students, Project Students, other University of Guelph employees using the equipment.
Dr. _____ Faculty Supervisor
Environmental Health and Safety
Local JHSC, Department of Biomedical Sciences

Written By: Luisito David Staff Technician

Approved By: _____ Faculty Supervisor (signature)

Date: June 24, 2002