DEPARTMENT OF MOLECULAR AND CELLULAR BIOLOGY

SAFE OPERATING PROCEDURE

LIQUID NITROGEN HANDLING AND DISPENSING

Purpose:
To provide safe operation and procedural guidance on the dispensing and handling of liquid nitrogen for research laboratory staff and students in the Department of Molecular and Cellular Biology.

Training:

All students and staff within MCB working with liquid nitrogen should be familiar with the following procedures and precautions. It is mandatory that all users have proper training provided by your supervisor or technical staff.

Safety Precautions:

- ⚠ Liquid nitrogen is a colourless, odourless liquid with a boiling point of -196^{o}C. Splashes and spatter can cause cold burns and frostbite. Protect your face and skin, and always use gloved hands or tongs to manipulate items that have been in contact with liquid nitrogen.
- ⚠ The vapors coming from these liquids are also extremely cold and can produce burns. Exposure to these cold gases, which is too brief to affect the skin of the face or hands, may affect delicate tissues, such as the eyes.
- ⚠ Boiling and splashing will occur when filling a warm dewar or when inserting objects into the liquid Nitrogen. Always perform these operations slowly to minimize these hazards.
- ⚠ Never allow any unprotected part of your body to touch uninsulated pipes or vessels containing cryogenic liquids. The extremely cold material may stick fast to skin. Always wear insulated gloves and a face shield. Gloves should be loose enough that they could be removed rapidly if they came in contact with liquid nitrogen.
- ⚠ One of the main dangers associated with liquid nitrogen is the risk of asphyxiation when used or stored in poorly ventilated areas. Liquid nitrogen evolves nitrogen gas which is inert and non-toxic but there is a risk of asphyxiation in situations where high concentrations may accumulate and subsequently displace air from the room. Rapid evaporation of liquid nitrogen can displace oxygen. Keep the door to the room open when dispensing liquid nitrogen from bulk storage dewars.
- ⚠ If you spill any quantity of liquid nitrogen, evacuate everyone from the area, and allow time for the ventilation system to return oxygen levels to normal.
- ⚠ Always keep liquid nitrogen in vacuum jacketed/insulated container designed for storage of cryogenic liquids.
- ⚠ To avoid explosion of pressurized containers, liquid nitrogen vessels must be equipped with a means to relieve pressure (i.e. relief valves or loose fitting lid).

Notes:

- Liquid nitrogen is a hazardous material under WHMIS – containers must have a supplier or workplace label, and an MSDS must be available.
- It is not permissible for anyone to work in atmospheres with oxygen concentrations below 19.5%. Do not handle liquid nitrogen in closed or confined spaces as asphyxiation can occur quickly in small, poorly ventilated areas.

Operating Procedure:

Dispensing:

- ✓ Wear a face shield and insulated gloves when dispensing liquid nitrogen.
- ✓ Using a wrench, affix the elbow and connecting hose to the valve labeled 'liquid' on the bulk storage tank. Do not over-tighten.
- ✓ Ensure the phase separator is affixed to the end of the hose, and place the hose into the receiving dewar.
- ✓ With a gloved hand, open the liquid valve slightly at first. Air pulses may occur as the hose cools – when they subside you may open the valve fully.
- ✓ Monitor the level of liquid nitrogen in the receiving dewar. Do not fill past the base of the neck, as this can lead to failure in the insulation.
- ✓ Fill to the desired level, close the valve fully.
- ✓ With a gloved hand, carefully remove the hose from the receiving dewar. The hose may be very cold, so avoid striking it against anything.
- ✓ Allow the hose to warm up, then disconnect from the bulk storage tank with the wrench.

Transport of dewars through building

- ✓ When transporting a closed dewar within the building, no personal protective equipment is necessary.
- ✓ Use a suitable, stable dolly or hand truck for transport. Do not attempt to carry dewars from one location to another.

Retrieving samples from liquid nitrogen storage

- ✓ Wear a face shield and loose fitting insulated gloves when retrieving samples.
- ✓ Rapid changes in temperature can cause a rupture in a sample container.
- ✓ Remove lid of dewar, grasp the wire hook and maneuver the sample container to the centre of the opening.
- ✓ Slowly lift the sample container straight up, then tilt slightly to allow excess liquid to run before lifting the item fully out of the dewar.
- ✓ Gently set the sample container in a suitable location and retrieve the desired samples.
- ✓ When returning the sample container to the dewar, ensure the wire hook is affixed securely, then lower slowly to into the dewar to minimize splashing.
- ✓ Replace the lid.

Contingency Plans

Spills

- In the event of a spill, evacuate everyone from the area of the spill and keep the area unoccupied for 30 minutes. As the spilled nitrogen evaporates, it will displace oxygen, and it will take time for the ventilation system to return oxygen concentration to normal.
- For spills larger than 4L, contact EHS. Clearance air testing may be required prior to re-entering the area.

Hazardous Atmospheres

- Immediately stop what you are doing and leave the room if you begin to experience any symptoms of oxygen deficiency, such as:
  - Dizziness
  - Seeing spots
  - Rapid breathing
  - Poor coordination, giddiness or altered judgment

Skin or Eye Contact

- Skin – immediately flush area with warm (not hot) water. Remove any saturated clothing and if injury has occurred, dial x2000.
- Eyes – immediately flush the eyes for 15 minutes, and dial x2000 to get help.

First Aid

- Where inhalation has occurred, the victim (who may be unconscious) should be removed to a well-ventilated area. Rescuers should not put themselves at risk - a contaminated area should not be entered unless considered safe. Breathing apparatus may be required but should only be used by trained personnel.
- The person should be kept warm and rested whilst medical attention is obtained.
- If breathing has stopped then resuscitation should be commenced by a trained first aider.
- Where contact has occurred, the aim should be to slowly raise the temperature of the affected area back to normal. For minor injuries, clothing should be loosened and the person made comfortable. Clothing should not be pulled away from burned or frozen skin. The affected area should be doused with copious quantities of tepid water (40oC) for at least 15 minutes and a sterile burn dressing applied to protect the injury until the person can be taken to receive hospital treatment.

- **Canada Regulations**: Nitrogen, Refrigerated Liquid (7727_37_9)
  Listed on the Canadian DSL (Domestic Substances List)
  WHMIS Classification: Class A – Compressed Gas