NEW YORK STRUCTURAL BIOLOGY CENTER

Standard Operating Procedure **For Work with Liquified Ethane**	Created: 02/09/2019 Reviewed: 02/09/2019

Chemical name/class: **Cryogens (Liquid ethane)** **CAS #:**

1. **Circumstances of Use:**

Ethane is a gas at room temperature. It is stored in a pressurized tank and used to freeze samples for Cryo-EM. To prepare for use, ethane gas is slowly released into a container cooled with liquid nitrogen and allowed to liquify. Once liquid, it is cooled to near-freezing point by liquid nitrogen. Samples to be frozen are rapidly plunged into liquid ethane

2. **Potential Hazards:**

General:

- Since liquid nitrogen us used to liquify the ethane, all hazards and circumstances for cryogens also apply to liquid ethane. In addition, the following hazards exist.
- During cooling, it is possible for ethane to solidify at the end of the tip where gas is released. This will stop gas flow and cause pressure buildup in the tubing.
- Frostbite/cryoburns; Because of the extreme low temperatures even very short exposures of unprotected skin or eyes can cause severe frostbite or loss of vision. This effect is stronger for liquid ethane than liquid nitrogen since the ethane will stick to tissue and freeze immediately, unlike nitrogen which will vaporize and form a gas barrier.
- Spattering: Spillage of liquid nitrogen onto liquid ethane will cause the ethane to immediately freeze. This can cause it to spatter and spray frozen and liquid ethane out of the cup.
- Flammability: Ethane gas is flammable. While nside the nitrogen cup, the local atmosphere is depleted of oxygen so fire is not a hazard. However when warming up the ethane gas will mix with room air and create a potential fire hazard.
- Consult the Safety Data Sheet (SDS) and

Toxicity:

- N/A

Reactivity:

- N/A

3. **Engineering Controls:**

- Use cryogens in well ventilated areas.
- When appropriate install or use oxygen monitors.
- For small rooms; oxygen monitors will be integrated with HVAC fans so than upon a low oxygen condition fans will automatically switch to “exhaust mode”.
- Safety pressure relieve valves will be installed on piping or containers between two valves or any other location where liquid cryogen may become trapped.
- A fume hood should be available for release of the ethane gas after the experiment.

4. **Work Practice Controls:**

- Do not touch Cryogenic materials, or tools in contact with Cryogens, with bare skin or disposable gloves (see PPE requirements below)
- Use tongs or similar tools to immerse and remove objects from liquid Cryogens.
- Do not alter or disable the pressure-relief mechanisms/valves as installed by the manufacturer.
  How not to do it: Texas Dewar Accident Summary

- Do not alter/modify the Cryogen containers as received from the vendor.
- Do not ride in elevators with Cryogen containers.
- Warm the ethane in a fume hood after the experiment. Do not allow to warm up in a closed room.

5. **Personal protective equipment (PPE):**

- Loose-fitting, thermal-insulated gloves (not intended for full immersion purposes) that are meant for incidental contact **must** be available to all personnel using Cryogens. No metal jewelry, watches, or rings should be worn while handing Cryogens.
- Laboratory safety glasses must be worn at all times when handling or preparing liquid ethane.
- In cases where the arms or torso may be exposed to liquid suspensions or dry particles, wear a chemically-compatible laboratory coat that fully extends to the wrist.

6. **Transportation and Storage:**

- Cryogen dewars should be moved at a walking speed with container under control at all times.
- If a Cryogen container does not have wheels, use appropriately designed dollies or carts.
- For Cryogen containers with wheels, rotate the container as it goes over thresholds or transitions. Rotation helps to prevent tipping.
- Cryogen containers should be stored in well ventilated areas.

7. **Waste Disposal:**

- Liquid ethane should be allowed to evaporate in a fume hood. Do not dump or pour on the floor, this will cause rapid evaporation and create a fire hazard due to flammability.
- Excess liquid nitrogen can be allowed to evaporate in well ventilated areas.
- Do not dump or pour excess nitrogen on the floor, this will cause rapid evaporation potentially displacing oxygen and/or damage flooring.
- Do not dispose of Cryogens into sinks or plumbing as this may cause damage.
- Do not put excess Cryogens into a sealed container, this may result in an explosion hazard.
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8. **Exposures/Unintended contact:**

- For symptoms of asphyxia such as headache, drowsiness, dizziness, excitation, excess salivation, vomiting or unconsciousness remove personnel to fresh air. Seek medical attention.
- If breathing is difficult, call 911 from any phone to request assistance. In the event breathing has stopped administer CPR. Seek medical attention.
- In the event of exposure to cryogen liquids or gases warm body part as quickly as possible with tepid water. Do not use hot water or dry heat. Under no circumstances should the frozen body part be rubbed, even after rewarming. Flush eyes with tepid water for at least 15 minutes. Seek medical attention.
- When safe to do so, inform LSO of exposure.
- An incident report found at: http://www.nysbc.net/twiki/bin/view/Staff/IncidentReports should be completed within 24 hours. Follow-up medical attention should be sought.

9. **Spill Procedure:**

- For small spills of cryogens; evacuate the immediate area and allow cryogen to evaporate and dissipate.
- For large spills, vacuum jacketed pipe failures, tank/dewar failures or any uncontrolled release of Cryogens; evacuate the building and call FDNY.
- Consult SOP for Liquid Nitrogen Bulk Tank Operation.
- Consult SOP for Activation of Oxygen Monitor.
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10. **SEMC Specific Instructions**

   a. Before starting, wear appropriate personal protection: goggles and long sleeves. Wear appropriate lab footwear: no open-toed shoes. Goggles or protective glasses must be worn throughout the experiment.

   b. Pre-cool the tip for ethane dispensing in liquid nitrogen. This will help it liquify faster. Pre-cool the ethane cup with liquid nitrogen.

   c. Place the tip in a pre-cooled cup. Open gas valve to start dispensing ethane. The flow should be low, otherwise the gas will come out too fast to liquify.

   d. When the ethane is liquifying, a low whistling sound can be heard. If gas flow is too fast, warm gas will not liquify in the cup. Adjust flow accordingly.

   e. Fill until cup is full. Close gas valve on ethane tank when done.

   f. If the ethane freezes, a metal rod can be used to melt it.

   g. When finished with the experiment, bring the ethane cup to the fume hood. Let it evaporate there.

11. **Training of personnel:**

All personnel are required to complete the online General Lab Safety session thru the OESO website. This session includes an introduction to general chemical safety. All personnel shall read and fully adhere to this SOP for safe handling of Cryogens, the SDS, and shall document that they have read these documents by signing and dating the SOP.

“I have read and understand this SOP. I agree to fully adhere to its requirements.”

Last	First	Signature	Date
