Date Published: 7-1-2018

Safe Handling of Cryogenic Materials Guidelines

Applies to: **(examples; Faculty,Staff, Students, etc)**

Faculty , Staff , Students

Policy Overview:

Issued: 07-01-2018

Next Review Date: 07-19-2022

Frequency of Review: Annually

Applies to University of Health Sciences and Pharmacy in St. Louis students, faculty, and staff working with cryogenic materials.

Definitions:

Term	**Definition**
Cryogenic Liquids	Liquefied gases with a boiling point lower than -130°F (-90°C) at an absolute pressure of 14.7 psi (101.3 kPa).
Boiling Point	The temperature of a substance at which the vapor pressure of the liquid equals the pressure surrounding the liquid and the liquid changes into a vapor.
Expansion Ratio	The volume of a given amount of that substance in liquid form compared to the volume of the same amount of substance in gaseous form, at room temperature and normal atmospheric pressure
OSHA Permissible Oxygen Levels	Any level between 19.5 percent and 23.5 percent oxygen. Any level above or below this range will cause respiratory problems up to, and including, asphyxiation and death.
Asphyxiation	Loss of consciousness or death caused by impairing normal breathing, as by gas or other noxious agents; choke; suffocate; smother.
Dewars	Liquid Dewar flasks are non-pressurized, vacuum-jacketed vessels, similar to a Thermos bottle. Dewars are designed with either loose-fitting caps or pressure relief valves, that prevents air and moisture from entering, yet allows excess pressure to vent.
Freezer Farm	Refers to a collection of multiple freezers, housed in one location.

Details:

1. Introduction
    a. The primary hazards of cryogenic liquids include both physical hazards such as fire, explosion, and pressure, but also health hazards such as chemical toxicity or severe frostbite and asphyxiation. Not only are some liquid cryogens flammable in gaseous phase but some are very strong oxidizers and under the right conditions, inert cryogenic gasses may condense oxygen from the atmosphere. This oxygen-rich environment in combination with organic, flammable, or combustible materials may be particularly hazardous. Pressure is also a hazard because of the large volume expansion ratio from liquid to gas that a cryogen exhibits as it warms and the liquid evaporates. This expansion ratio also makes cryogenic liquids more prone to splash and spatter, therefore skin and eye contact may occur. Contact with living tissue can cause frostbite or thermal burns, and prolonged contact can cause blood clots that have very serious consequences. All laboratory personnel should follow prudent safety practices when handling and storing cryogenic liquids.

Procedures:

1. Properties
    a. Boiling point and expansion ratios for some common cryogenic liquids:

Gas	Boiling Point (ºC)	Volume Expansion Ratio
Argon	-185.7	847:1
Carbon Dioxide	-78.5	553:1
Fluorine	-187.0	888:1
Helium	-269.0	757:1
Hydrogen	-252.7	851:1
Methane	-161.4	578:1
Nitrogen	-195.8	696:1
Oxygen	-183.0	860:1

2. Hazards
    a. Thermal (low temperature) - Contact with cryogenic liquid, its boil-off gases, or components cooled to these low temperatures can readily cause frostbite or cryogenic burns. Release of these cryogens into the work area can damage equipment and property (e.g., frozen water pipes, damaged flooring, damaged electrical cables and their insulation).
    b. Pressurization - Cryogenic fluids confined and allowed to warm can generate very high pressures. LN2 confined and allowed to warm to room temperature will generate a nominal pressure of 10,200 psig. The pressure similarly generated by LHe is 11,000 psig. Other cryogens behave in similar fashion. Dry ice (solid CO_2) can also generate hundreds of psig pressure if confined. The function of vent lines can be defeated by the formation of ice (from condensed moisture) in the vent line. With LHe, air or other gases can solidify to form this blockage. If a cryogenic fluid is subjected to a large amount of heat input, a flash vaporization can occur. This will result in a rapid pressure rise that can be described as a BLEVE (boiling liquid expanding vapor explosion).
    c. Venting - Required vents and pressure-relief devices must be vented to a safe location, which is determined with the following criteria: 1) The specific cryogen in question; 2) The volume and flow rates of the potential releases; 3) The potential hazards presented by accumulation of the gases or liquids being vented.
    d. Oxygen deficiency/asphyxiation - Cryogenic fluids have large liquid-to-gas expansion ratios, which means that any accidental release or overflow of these cryogenic liquids will quickly boil into gas and may create an asphyxiation hazard by displacing the oxygen content of the surrounding area. In the case of LN2, the nitrogen gas generated from malfunctioning equipment or spills will be cold and denser than ambient air. Even well-ventilated lab spaces that have pits or other low-lying (or recessed) areas could have the oxygen displaced by this cold, dense N_2 gas. Argon or carbon dioxide also present these heavier than air hazards. Large-volume sources used in small laboratory spaces or in poorly ventilated areas increase the asphyxiation hazard. Oxygen monitors may be advisable in some applications.
    e. Ice buildup - The temperatures associated with cryogenic liquids can easily condense moisture from the air and cause the formation of ice. This ice can cause components or systems to malfunction (e.g., can plug vent lines and impede valve operation) or can damage piping systems. In the case of LHe, air itself can freeze solid and block vent lines. Building exhaust systems that are accidentally cooled to LN2 temperatures can also be damaged by ice formation or the weight of the accumulated ice and the weight of the LN2 itself. The resultant runoff water when the ice melts can also present a hazard.
    f. Materials concerns - The low temperature of cryogenic liquids will adversely affect the properties of some materials, resulting in system or vessel failure. The process for selecting materials to construct vessels and piping systems for cryogen handling must consider the behavior of the materials at cryogenic temperatures. Carbon steels and other metals can become brittle and fracture easily at cryogenic temperatures. Common acceptable materials for construction include metals such as the 300 series stainless steels, some aluminum alloys, and copper or brass. Plastics, such as Tygon® tubing, become brittle and can easily fail in cryogenic applications. Be sure to consult the appropriate references when selecting materials for cryogenic applications. Even when the appropriate materials are selected, thermal stresses can lead to failure in some applications. Thermal gradients across a material or piping system or the rapid cool-down of a vessel can generate thermal stresses. Joining materials with dissimilar coefficients of expansion can also generate thermal stresses.
    g. Oxygen enrichment - LN2 is cold enough to condense the surrounding air into a liquid form. The concentration of O_2 in this condensed air is enhanced. This condensed “liquid air” can be observed dripping from the outer surfaces of uninsulated/ nonvacuum-jacketed lines carrying LN2. This “liquid air” will be composed of approximately 50% O_2 and will amplify any combustion/flammable hazards in the surrounding areas. Open dewars of LN2 can condense O_2 from the air into the LN2 and cause an O_2 enrichment of the liquid that can reach levels as high as 80% O_2. Air should be prevented from condensing into LN2 with loose-fitting stoppers or covers that allow for the venting of LN2 boil-off gas. Large quantities of LN2 spilled onto oily surfaces (such as asphalt) could condense enough O_2 to present a combustion hazard. LHe can also condense air into a liquid or even solid with an enriched O_2 content.
    h. Lifting, physical - Studies of accident statistics involving cryogenics commonly include back strains or other lifting injuries associated with dewars. Although not specifically cryogenic in nature, this hazard is appropriate to note as a hazard associated with cryogenic applications. Care should be taken when lifting and moving cryogenic dewars. The proper use of carts or hand trucks can help prevent these injuries. Alternately, the use of low-pressure liquid-transfer equipment and procedures can replace lifting and pouring operations. Appropriate footwear, such as steel-toed shoes is recommended.
    i. LN2/ionizing radiation field - Using LN2 in high ionizing radiation fields that can generate ozone or nitrogen oxides may cause a potential explosion hazard when the LN2 condenses quantities of oxygen from the atmosphere. The applicable

control measure is to minimize the accumulation of oxygen into the LN2 and to keep containers free of hydrocarbon contamination.
   j. Noise - Transfer or venting of cryogens can generate, in some cases, noise levels that may require hearing protection. Sound levels in excess of 150 dBA have been recorded during routine tank filling. A redesign of the equipment or procedure could also be addressed in these cases.
   k. Others - Other cryogenic fluids present specific hazards in addition to the above concerns. Examples include: LOX, with the added concerns of materials compatibility and cleanliness (hydrocarbon contamination), presents additional enhanced combustion hazards. LH2, with the added concerns of low ignition energy, proper bonding and grounding of equipment, and venting of boil-off gases, presents additional hazards of flammability and materials embrittlement.
   l. *Note: This list is not to be considered exhaustive. Seek additional guidance from the manufacturer or EH&S for a thorough hazard analysis and safe operation of these systems.*
3. Requirements for Use of Cryogens
   a. All near misses and accidents (e.g. spills and burns) must be reported to supervisor/PI immediately.
   b. All injuries related to the handling of cryogenic materials should be treated immediately.
   c. Personal Protective Equipment
      i. Face shield required. Cryogen liquids can splatter, possibly onto face and eyes. Safety glasses (even with side shields) or goggles are not sufficient to protect the face.
      ii. Cryogloves are required (gloves should be loose fitting, so they can be quickly removed if liquid pours into them, or they should be elastic cuff insulated gloves).
      iii. A splash resistant lab coat is required to minimize skin contact. A full-length splash resistant apron may also be worn.
      iv. Closed toe shoes that cover the entire foot.
      v. Skirts and shorts are strongly discouraged due to the potential splash hazard.
4. Rules and Precautions for Handling, Transporting, and Storing
   a. Due to high expansion ratios and potential for asphyxiation cryogenic materials must only be used/stored only in well-ventilated areas. Cryogenic materials and dry ice must never be stored in cold rooms.
   b. Never dispose of LN2 by pouring it on the floor or into a sink. It could displace enough oxygen and cause suffocation and cause damage to materials it contacts. (Nitrogen is colorless and odorless – the cloud that forms when you pour LN2 is condensed water vapor from the air, not nitrogen gas.)
   c. Do not allow any LN2 to touch any part of your body or become trapped in clothing near the skin.
   d. Do not touch any item that has been immersed in LN2 until it has warmed to room temperature.
   e. Never dip a hollow tube into LN2; it may spurt liquid.
   f. Metals to be used for equipment in LN2 must possess satisfactory physical properties at the low operating temperatures.
   g. Introduction of a substance which is at normal room temperature into a cryogenic liquid or gas is always somewhat hazardous. There is a violent evolution of gas, and there is likely to be considerable splashing of liquid.
5. Equipment and Areas Utilizing Cryogens:
   a. The following are some of the most common locations where cryogenic liquids are used:
   b. Nuclear Magnetic Resonance (NMR) rooms (or magnet rooms)
   c. Freezer farms
   d. Cold traps
   e. Cryo tubes
   f. Labs performing x-ray crystallography
   g. Tissue banks
6. NMR:
   a. There are two sets of cryogens required to operate an NMR, liquid nitrogen (LN2) and liquid helium (LHe). These are both used to keep the superconducting coils that power the magnet cooled below its critical superconducting temperature (Tc). The outside air is typically at around 20-25 degrees Celsius (293-298 Kelvins). The liquid helium and liquid nitrogen used in the NMR magnets are extremely cold. Helium liquefies at 4 K (-269°C), and nitrogen liquefies at 77 K. Be careful and stay out of the NMR room when the magnet is being filled with cryogenic gases. Prolonged contact with liquid nitrogen or even brief contact with liquid helium will cause frostbite. If the magnet quenches, the liquid helium and nitrogen inside the magnet may quickly boil off. Due to their large expansion ratios (nitrogen 695:1, helium 760:1), these gases can quickly displace all the oxygen in the NMR room and cause asphyxiation. The temperature of the room will also drop increasing the risk of hypothermia.
7. Cold Traps:
   a. Cold traps are used in instrumentation and other systems to prevent the introduction of liquids, vapors, or contaminants into the system. Single finger traps, U-traps, or other shaped lines are submerged in cryogenic liquids or slushes to provide a very low temperature surface on which the molecules condense. Cold traps also improve the achievable vacuum in systems by one or two orders of magnitude. Improperly managed cold traps can impair accuracy, destroy instruments, and present a physical or health hazard. Many of the slush mixtures used in cold traps are toxic or explosive hazards and may not be indicated in the literature. Cold traps exposed to atmospheric air may liquefy air resulting in an accumulation of liquid oxygen and nitrogen in the line. In the presence of organic materials this can result in a dangerous oxidation reaction. Also, if liquefied air is exposed to atmospheric pressure by opening a valve, the liquefied air will be pushed through the line by atmospheric pressure and when it contacts warmer plumbing, the liquid will rapidly expand to gas causing the gas to shoot through the line at nearly explosive velocities.
8. Cryotubes:
   a. Cryotubes used to contain samples stored under liquid nitrogen may explode without warning. Tube explosions are thought to be caused by liquid nitrogen entering the tube through minute cracks and then expanding rapidly as the tube thaws.

When thawing cryotubes, all PPE requirements must be met and the tubes should be slowly thawed inside a biological safety cabinet, heavy-walled container (e.g. desiccator) or behind a safety shield while it is thawing.

9. Freezer Farms:
    a. A "freezer farm" refers a collection of freezers that are housed in one location. Some of the freezers may use liquid nitrogen, either as a primary or back-up coolant. Because of the large amount of nitrogen being used, it is important that there be adequate ventilation in the area. An oxygen level alarm may also be required. Contact EH&S for assistance in determining ventilation and safety requirements. Storage of 10 or more liquid nitrogen cooled freezers must be reviewed by EH&S.
10. Storage:
    a. Work and storage areas should be well ventilated. Evaporation of the liquid cryogens will displace oxygen in the room and may present an asphyxiation hazard. Air contains about 21% oxygen and breathing air with less than 18% oxygen can cause dizziness and result in unconsciousness, injury and death.
    b. *Note: The cloud that appears when liquid nitrogen is exposed to air is condensed moisture in the atmosphere. Gaseous nitrogen is invisible.*
    c. Cryogenic liquids should be handled and stored in containers that are designed for the pressure and temperature to which they may be subjected. The most common container for cryogenic liquids is a double-walled, evacuated container known as a Dewar flask.
    d. Containers and systems containing cryogenic liquids should have pressure relief mechanisms.
    e. Cylinders and other pressure vessels such as Dewar flasks used for the storage of cryogenic liquids should not be filled more than 80% of capacity, to protect against possible thermal expansion of the contents and bursting of the vessel by hydrostatic pressure. If the possibility exists that the temperature of the cylinder may increase to above 30ºC (86°F), a lower percentage (i.e. 60% capacity) should be the limit.
    f. Dewar flasks should be taped or shielded with mesh to minimize flying glass and fragments should an implosion occur.
    g. Dewar flasks should be labeled with the full cryogenic liquid name and hazard warning information.
    h. Never modify pressure relief valves on cryogenic cylinders. Pressure relief and occasional venting of gas is necessary to prevent over pressurization and explosion of cryogenic cylinders.
11. Handling
    a. Handle cryogenic liquids carefully. The extremely low temperatures can freeze human flesh rapidly. When spilled on a surface the liquid tends to cover the surface completely. The gas issuing for the liquid is also very cold. Delicate tissue such as the eyes can be damaged by the cold which may be too brief to damage the skin.
    b. Never allow unprotected parts of your body to touch any objects cooled by liquid cryogens. Unprotected body parts in contact with vessels or pipes that contain cryogenic liquids may bond firmly to the skin and tear flesh if separation is attempted.
    c. Use tongs or proper gloves to handle objects that are in contact with cryogenic liquids.
    d. Appropriate personal protective equipment should be worn when handling cryogenic liquids. This includes special cryogen gloves, safety goggles, full-face shield, impervious apron or coat, long pants, and high topped shoes. Gloves should be impervious and sufficiently large to be readily removed should a cryogen be spilled. Watches, rings, and other jewelry should NOT be worn.
    e. Precautions should be taken to keep liquid oxidizers (i.e. oxygen) from contacting organic materials; spills on oxidizable surfaces can be hazardous. Do not use grease on any oxygen equipment, fittings, or containers.
    f. Keep all equipment clean. Keep storage areas clean and free of moisture or conditions that could damage or corrode containers.
    g. Transfers or pouring of cryogenic liquid should be done very slowly to minimize boiling and splashing. Use a phase separator or special filling funnel to prevent splashing. The top of the funnel should be partially covered whenever possible to shield splashes.
    h. Use small Dewars that are easily handled to transfer liquids. Use a cryogenic liquid withdrawal device for larger quantities from heavy containers.
    i. Keep Dewars upright and avoid rough handing. Do not transport Dewars in closed vehicles.
    j. Do not overfill containers. Overfilling above the neck tube can cause overflow when the neck tube core or cover is placed in the opening.
    k. Never use hollow tubes as dipsticks. When a warm tube is inserted into the cold liquid the liquid will spout from the top of the tube due to the rapid expansion of liquid cryogen inside the tube. Wood rods or solid metal dipsticks are recommended. Plastic rods will become very brittle.
    l. Keep cryogenic liquids and dry ice used as refrigerant baths open to the atmosphere. They should never be used in a closed system where they may develop uncontrolled or dangerously high pressure.
    m. Do not transfer liquid hydrogen in an atmospheric air environment. Oxygen from the air can condense in the liquid hydrogen presenting a possible explosion hazard.
    n. Never dispose of liquid cryogens in confined spaces or drains. Liquid nitrogen can be slowly poured onto gravel outside where it will evaporate safely without causing damage.
12. First Aid
    a. Asphyxiation:
        i. If a person becomes dizzy move them to a well ventilated area immediately.
        ii. If breathing has stopped apply artificial respiration or cardio pulmonary resuscitation (CPR) immediately.
        iii. Call Public Safety at 314-446-SAFE (7233) and keep them warm and calm and get prompt medical attention.
    b. Frozen Tissue:
        i. Restore tissue exposed to liquid cryogens to normal body temperature (98.6°F) as rapidly as possible.

ii. Remove or loosen clothing that may restrict blood flow to the frozen tissue. Rapid warming can be achieved by using warm water. Under no circumstances should the temperature exceed 112°F.
iii. Protect tissue from further damage or infection. Do not rub warming tissue before or after warming.
iv. Call Public Safety at 314-446-SAFE (7233) and keep them warm and calm and get prompt medical attention.

Responsibilities:

Position/Office/Department	Responsibility
Researchers	Must complete the annual laboratory safety training and the lab specific training and document training compliance on Moodle. Must wear all required personal protective equipment
Principal Investigator/Supervisor	Ensure that staff handling liquefied gases are instructed and trained as to the nature and the safe handling of cryogenic materials.

Resources:

University of Health Sciences and Pharmacy in St. Louis Personal Protective Equipment Policy

Safe Use of Cryogens Training Module on Moodle

Policy Contacts:

Name	Contact Information
Carlin Harp	314-446-8133
Eric Knoll	314-446-8375