United Lincolnshire Hospitals NHS
NHS Trust

Safe Handling and Utilisation of Liquid Nitrogen (LN_2) SOP 19

SOP Title	Safe Handling and Utilisation of Liquid Nitrogen (LN_2)
SOP No.	SOP 19
Author	Andrew Sloan
Consulted Departments	Lincolnshire Clinical Research Facility, Research and Development, Trust consultants and research staff.
Lead Manager Sign and Print Name	Dr. Tanweer Ahmed Director of LCRF and Head of Research and Development (Signature on file)
Date published	11/02/2015
Review date of SOP	04/02/2017
Version 1.1 (11/02/2015)	

Version History Log

This table should detail the version history for this document. It should detail the key changes when a version is amended.

Version	Date Implemented	Details of key changes
1	05/02/2015	-
1.1	11/02/2015	• LCTU updated to LCRF in consulted departments section • Dr Tanweer Ahmed 'R&D Manager' corrected to 'Head of R&D' • 'Applies to' section amended to include 'researchers'

Purpose:

The purpose of this SOP is to describe the safe handling of liquid nitrogen (LN_2) in the cryogenic preparation of donated human tissue samples in certain clinical research trials.

It relates to the handling and use of 500 mL of liquid nitrogen or less.

Background:

Liquid nitrogen is nitrogen that is cold enough to exist in liquid form. It is used for many cooling and cryogenic applications.

- Liquid nitrogen is the liquefied form of the element nitrogen (N) that is commercially produced by fractional distillation of liquid air.
- Liquid nitrogen is usually denoted as LN_2, LN, or LIN.
- Liquid nitrogen has the UN number 1977.
- At normal pressure, liquid nitrogen boils at 77 K (−195.8°C/−320.4°F).
- The liquid to gas expansion ratio of nitrogen is 1:694, therefore liquid nitrogen boils to fill a volume with nitrogen gas very quickly.

Liquid Nitrogen: General Uses

- Freezing and transport of food products.
- Cryopreservation of biological samples.
- Coolant for superconductors, vacuum pumps, and other materials and equipment.
- Cryotherapy to remove skin abnormalities.
- Shielding materials from oxygen exposure.
- Cooling materials for easier machining or fracturing.

Applies to:
Researchers, research nurses/radiographers and research managers. Supervised students.

Relevant SOP documentation:
Control of Substances Hazardous to Health Regulations 2002
Human Tissue Act 2004 (see SOP 17 for further details)
Local COSHH Assessment Form (PHB) (attached)
Local Risk Assessment (PHB) (attached)
Local Guidelines for Decanting Liquid Nitrogen, Dermatology Unit (PHB) (attached)

Definitions:
SOP – Standard Operating Procedure
PHB – Pilgrim Hospital Boston
CRF – Clinical Research Facility

Policy:

Liquid Nitrogen Safety

- Liquid nitrogen is cold enough to cause severe frostbite upon contact with living tissue, especially skin surfaces. Wear proper personal protective equipment (PPE) when handling liquid nitrogen to prevent contact or inhalation of the extremely cold vapour. Make sure exposed skin is covered i.e. long-sleeved lab coat (preferably cuffed at the sleeve ends), gloves, long trousers, no open-topped footwear.
- Because it boils rapidly, the phase transition from liquid to gas can generate a lot of pressure very quickly. Do not enclose liquid nitrogen in a sealed or screw-top container, as this may result in bursting or an explosion. Receptacles must be fit for purpose (i.e. dewar flask) and should only be transported in the minimum volume required for the task.
- Adding a lot of nitrogen to the air reduces the relative amount of oxygen. This can result in an asphyxiation risk. Cold nitrogen gas is heavier than air, so the risk is greatest near the ground. Use liquid nitrogen in a well-ventilated area.
- Liquid nitrogen containers may accumulate oxygen which is condensed from the air. As the nitrogen evaporates, there is a risk of violent oxidation of organic matter.

Responsibilities:

Cold Burns

Extremely low temperatures can freeze flesh rapidly. When spilled on a surface, liquid nitrogen tends to cover it completely, cooling a large area. The gas forming from the liquid is also extremely cold. Delicate tissue, such as eyes, can be damaged by an exposure to cold gas alone which would be too brief to affect skin.

Unprotected body parts contacting objects cooled by liquid nitrogen may stick fast. This may result in injuries by flesh being torn whilst attempt to withdraw from the object. Small splashes of liquid nitrogen will run off bare skin due to a vapour layer forming between the skin and the liquid. This must never be relied upon.

Asphyxiation

Liquid nitrogen rapidly vaporises to gas that is approximately 700 times the liquid volume. By displacing air, the gas has the potential to kill by asphyxiation. When the oxygen concentration in air is sufficiently low, a person can become unconscious without any warning symptoms.

Over Pressure

Since liquid nitrogen boils rapidly, users must ensure that it is never used in a closed system. Therefore do not use thermos flasks or screw-top bottle/containers to store liquid nitrogen, as this presents an explosion risk.

Embrittlement

Many ordinary materials cannot withstand cryogenic temperatures. Laboratory plumbing is one example, therefore never dispose of cryogenic liquids down the drain. Materials exposed to cryogenic temperatures for long periods or which have undergone periodic warming and freezing should be examined for cracks and crazing.

Precautions

Local rules for liquid nitrogen safety (see Policy number: ULH-H&S- COSHH 1 – version 1.0. (See attached)

Local rules exist for handling Liquid Nitrogen in association with specialist equipment. All such local rules must be adhered to. Additional guidelines can be found in the attached BOC information sheet.

Containers

Use only containers designed for low-temperature liquids. Cryogenic containers (e.g. dewar flasks) are specifically designed and made of materials that can withstand the rapid changes and extreme temperature differences encountered in working with liquid nitrogen. Even so, these special containers should be filled SLOWLY to minimise the internal stresses that occur when any material is cooled. Excessive internal stresses can damage the container.

• Do not cover or plug the entrance opening of any liquid nitrogen dewar.

• Do not use any stopper or other device that would interfere with venting of gas.

Cryogenic liquid containers are generally designed to operate with little or no internal pressure. Inadequate venting can result in excessive gas pressure which could damage or burst the container. Check the unit periodically to be sure that venting is not restricted by accumulated ice or frost.

Protective Clothing

When using or decanting liquid nitrogen, a face shield or safety goggles must be used. Always wear appropriate gloves when handling anything that is, or may have been, in immediate contact with liquid nitrogen. Use tongs to withdraw objects immersed in the liquid, and handle the object carefully. Do not put hands (even in the best gloves) into liquid nitrogen. Appropriate gloves are thermal protective gloves which are specifically designed for cryogenic use preferably with close fitting ribbed cuffs to prevent liquid nitrogen from spilling inside the glove. Inadequate protective clothing can absorb the liquid nitrogen and result in even more severe burns than would otherwise have resulted. Additionally, when handling liquid in open containers, it is advisable to wear closed-top shoes. Trousers should be worn outside the shoes.

Training

Safety precautions must be followed to avoid potential injury or damage. Do not attempt to handle liquid nitrogen until you fully understand the potential hazards, their consequences, and the related safety precautions.

Handling of Liquid Nitrogen

Decanting of Liquid Nitrogen

Never overfill dewars. Spillage damages flooring and may cause injury. Insert objects (e.g. forceps) into liquid nitrogen slowly to avoid splashing. Great care should be exercised to ensure that space is left to replace lids/tops on dewars especially those that close a considerable distance into the vessel.

Maintenance of Dewars.

Small dewars should be visually inspected each time they are refilled and any defects must be reported to the line manager. LCRF Researchers may do this as only small containers will be used (containing <500 mL of liquid nitrogen)

Condensed moisture or frost on the outer shell of a refrigerator or dewar and abnormally rapid evaporation of the liquid nitrogen are indications of vacuum loss. If vacuum loss is evident or suspected, transfer the materials stored in the unit to another refrigerator as soon as possible and remove the unit from service.

Transport

Never carry more than one container, which should not contain more than 500 mL of liquid nitrogen. Keep unit upright at all times and hold with both hands, do not run or walk hastily. Wear insulated gloves. Goggles not required if container is closed as this can distort vision whilst walking. Avoid tipping the container or laying it on its side, which can cause spillage of liquid nitrogen. It may also damage the container and any materials stored in it. Rough handling can cause serious damage to dewars. Gently place container onto the laboratory worktop on arrival at the place of use, ensuring it is situated to the rear of the bench. Do not 'visit' other working areas on the way to the lab and avoid crowded areas of the hospital if possible.

Disposal

Never dispose of cryogenic liquids down the drain. Ordinary materials may not be able to withstand cryogenic temperatures without failure. Laboratory plumbing is a common example (Newcastle University, UK). Allow waste liquid nitrogen to evaporate naturally in a well-ventilated area using fan extraction. This is available in the research lab at PHB. The BOC guidelines can be consulted for further advice and information for disposal of liquid nitrogen.

First Aid

Skin/Eye Contact.

Immediately flush thoroughly with copious quantities of tepid water (the water must not be hotter than 44° C). In case of frostbite spray with water.

DO NOT apply any form of direct heat.

DO NOT rub affected parts either before or after warming.

Move the casualty to a warm place (22° C).

Arrange for the casualty to be transported to A&E without delay. While waiting for transport:

- Remove or loosen restrictive clothing.
- Continue to flush the affected area with copious quantities of tepid water.
- Protect any frozen parts with bulky, dry, sterile dressings. Do not apply to tightly.
- Keep patient warm and at rest.
- Ensure ambulance crew/hospital is advised of details of accident and first aid treatment already administered.
- The casualty should not smoke, nor drink alcohol

Anoxia (should not apply when using <500 mL)

Attempts to rescue affected persons from confined spaces or where oxygen deficient atmospheres may be present should only be made by those trained in the use of breathing apparatus and confined space entry procedures. The Fire Brigade should be called in all instances where a trapped person requires rescue. If a person seems to become dizzy or loses consciousness while working with liquid nitrogen, move to a well-ventilated area immediately. If breathing has stopped, apply artificial respiration. Keep warm and at rest. The patient should neither smoke, nor drink alcohol

References:

The Health and Safety Executive (HSE): *COSHH Basics*. (2014) Available from: http://www.hse.gov.uk/coshh/basics.htm

Newcastle University (United Kingdon): Handling of Liquid Nitrogen: Guidance and Information Sheet. Available from: https://www.safety.ncl.ac.uk/uploads/Handling%20of%20Liquid%20Nitrogen.pdf

This SOP will be reviewed every 2 years unless changes to legislation require otherwise
Current versions of all SOPs are located on the LCRF website – users are responsible for ensuring that they are using the most up-to-date version.