

SOP Title: Use and Maintenance of the Liquid Nitrogen Freezer

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1. PURPOSE

- 1.1. The purpose of this procedure is to describe the proper use and maintenance of the Liquid Nitrogen (LN₂) Freezer.

2. SCOPE

- 2.1. This procedure applies to the Human Papillomavirus (HPV) Serology Laboratory located at the Advanced Technology Research Facility, room C2007.

3. REFERENCES

- 3.1. CryoPlus 1 Storage System 7400 user manual
3.2. ISM-176: Safe Handling of Liquid Nitrogen

4. RESPONSIBILITIES

- 4.1. The Research Associate, hereafter referred to as analyst, is responsible for reviewing and following this procedure.
4.2. The Scientific Manager or designee is responsible for training personnel in this procedure and reviewing associated documentation.
4.3. The Quality Assurance Specialist is responsible for quality oversight and approval of this procedure.

5. REAGENTS, MATERIALS AND EQUIPMENT

- 5.1. Liquid Nitrogen Freezer
5.2. LN₂ Measuring Stick (VWR, Cat# 97049-658 or equivalent)
5.3. Liquid Nitrogen
5.4. Cold Resistant Gloves

6. HEALTH AND SAFETY CONSIDERAIONS

- 6.1. Proper safety precautions should be taken while working in a laboratory setting. This includes, but is not limited to, proper protective equipment such as lab coats, safety glasses, closed-toe shoes, and non-latex gloves.
6.2. Extreme temperature hazards. Use appropriate cold resistant gloves when handling samples or performing maintenance in the liquid nitrogen freezer.

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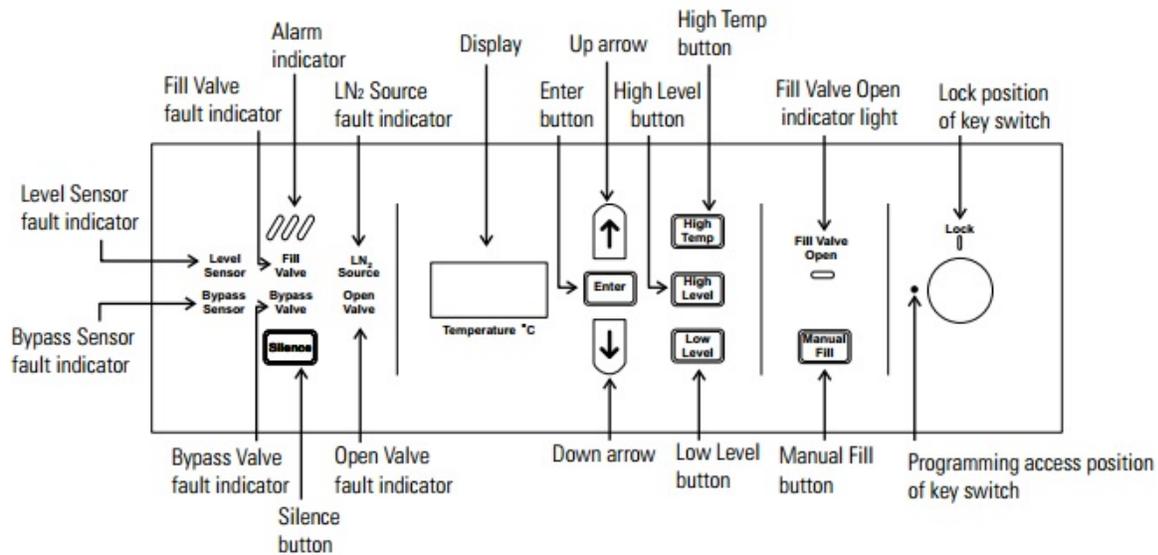
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- 6.3. Refer to “ISM-176: Safe Handling of Liquid Nitrogen” for safety guidelines regarding handling liquid nitrogen tank.

7. OPERATION

7.1. Control Panel

Figure 1: Liquid Nitrogen Freezer Control Panel



- 7.1.1. **Enter** button, used to send programming changes to the microprocessor.
- 7.1.2. **Up and down** arrows that change the high and low level settings when making programming changes, are also used to change the high temperature alarm set-points.
- 7.1.3. **High Level** button, changing the level at which the system stops filling.
- 7.1.4. **Low Level** button, pressed to change the level at which the system starts filling.
- 7.1.5. **High Temp** button, changing the temperature at which the high temperature alarm activates.
- 7.1.6. **Manual Fill** button, pressed to manually fill the tank. The level must be at least 1-1/4 inches below the high level set-point to start a manual fill.
- 7.1.7. **Fill Valve Open** light, indicating the fill valve is open.

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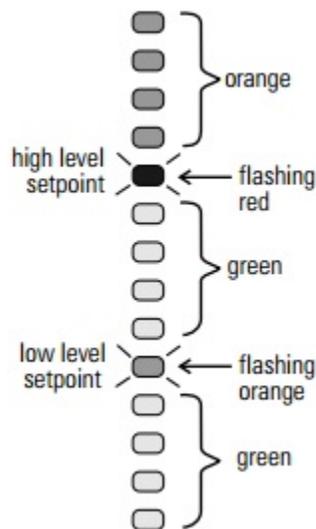
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7.1.8. **Key** switch, used to allow program changes when the key is in the (.) Programming Access position and to protect the system from tampering or accidental button presses when the key is in the Lock position.

7.2. Fill Level Bar Graph

Figure 2: Liquid Nitrogen Freezer Fill Level Bar Graph



7.2.1. **Orange (steady):** indicates the remaining space above the high level set-point.

7.2.2. **Orange (flashing):** Under normal conditions, the flashing upper LED is the high level (stop fill) set-point, and the flashing lower LED is the low level (start fill) set-point.

7.2.3. **Green:** indicates the actual liquid Nitrogen level.

7.2.4. **Red (steady):** indicates the amount of space below or above set-point from the actual liquid level.

7.2.5. **Red (flashing):** indicates that the liquid level is above or below the level set points. A possible alarm condition is pending.

7.2.6. **Single Red (flashing):** after a fill operation indicates that the liquid level is at the high level set-point. This is not an alarm condition.

8. USE AND MAINTENANCE

8.1. Do not leave the lid of the unit open for extended periods of time.

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- 8.2. The liquid nitrogen freezer is monitored by the Rees alarm system. If there is an alarm notification and the unit is not operational, then an entry must be recorded.
- 8.3. The target level of LN₂ in the liquid nitrogen freezer is between the low level setpoint and high level setpoint, as illustrated in Figure 2. Visually check the LN₂ level daily.
- 8.4. If the LN₂ level is low (more than half past the defined limits on the digital gauge), turn the Key to the Programming Access position and press the Manual Fill button. The unit should activate a filling process within 5 seconds; if not, verify the LN₂ tank valve is completely open.
- 8.5. The liquid nitrogen freezer can house up to four racks (Labeled 1, 2, 3, and 4). Do not load material into the bottom-most three slots of each rack.
- 8.6. Changing the Liquid Nitrogen Tank
 - 8.6.1. Obtain a full tank from the Logistics Support group.
 - 8.6.2. Close the "Liquid" valve on the LN₂ tank by turning completely in a clock-wise direction.
 - 8.6.3. Disconnect the steel braided line from the valve with an adjustable wrench.
 - 8.6.4. On the empty tank, initial and date the "Empty" tank tag section and tear off the "In Use" and "Full" section of the tag. The "In Use" and "Full" section tag may be thrown away if the Tank was in use for more than 7 days. If fewer than 7 days, alert the manager to trend Tank issues.
 - 8.6.5. Replace the empty tank with the full tank.
 - 8.6.6. Connect the braided steel line to the full tank at the "Liquid" valve.
 - 8.6.7. Hand tighten the connector; finish tightening using an adjustable wrench.
Note: Do not overtighten or use Teflon tape.
 - 8.6.8. Open the "Liquid" valve on the tank by turning completely in a counter-clockwise direction.
 - 8.6.9. Initial and date the "In Use" tank tag section.
 - 8.6.10. Return empty tank to the Logistics Support group.
- 8.7. Quarterly Manual Measurement of Liquid Nitrogen level
 - 8.7.1. Record the level of LN₂ using the unit bar graph on "HSL_EQ_009.01: Liquid Nitrogen Freezer Maintenance Form."

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8.7.2. Open unit and carefully lower the LN₂ measuring stick to the bottom for a few seconds.

8.7.3. Remove the measuring stick; a white frost forms to indicate the highest point of LN₂. Record measurement on HSL_EQ_009.01.

8.7.4. The level must be within two inches of the set level indicated on the front of the instrument. If outside the two inch tolerance, contact Facilities, Maintenance and Engineering (FME) or a contracted vendor to fix the problem. If the samples need to be removed, the manager will coordinate a suitable temporary storage freezer.

8.8. Annual Calibration

8.8.1. FME or a contracted vendor certify and perform calibration of the liquid nitrogen freezer.

9. ATTACHMENTS

9.1. Attachment 1: HSL_EQ_009.01: Liquid Nitrogen Freezer Maintenance Form

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Attachment 1: HSL_EQ_009.01: Liquid Nitrogen Freezer Maintenance Form

Frederick National Laboratory for Cancer Research <i>sponsored by the National Cancer Institute</i>		HPV Serology Laboratory Standard Operating Procedure Form	
Form Title: Liquid Nitrogen Freezer Maintenance Form			
Document ID: HSL_EQ_009.01		Version: 2.0	
Associated SOP: HSL_EQ_009		Effective Date:	
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Quarterly Maintenance

Quarter	Display Level (in.)	Measured Level (in.)	Performed by/date	Reviewed by/date
1				
2				
3				
4				

Unscheduled Maintenance

Date	QE Number	Activity Performed	Recorded by/date	Reviewed by/date

QA Reviewed by/date: _____

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