

STANDARD OPERATING PROCEDURE **Indiana CTSI Specimen Storage Facility**

TITLE: STANDARD OPERATING PROCEDURE FOR THE LIQUID NITROGEN SYSTEM AND LIQUID NITROGEN FREEZER ROOM OPERATIONS

CHAPTER:

2-Facility

Issue Date: 12.28.2022

SOP #:

SF-2-2.13

SUPERSEDES SOP #: N/A

Effective Date: 02.01.2023

AUTHORED BY: Indiana CTS/ SSF Staff

DATE: 12-27-22

APPROVAL: Robert W. Orr Jr Digitally signed by Robert W. Orr Jr Date: 2022.12.28 11:27:57 -05'00'

_DATE: 12/28/2022

Indiana CTSI SSF Director

QA APPROVAL: _ Anne Kaiser

DATE: _12/28/2022

Ouality Compliance Specialist

1. REVISION

- 1.1. Significant changes incorporated in this version include:
 - 1.1.1. All references to Praxair were changed to "Praxair/Linde", since Linde purchased Praxair.
 - 1.1.2. Revised Section 4 to define alarm system management terminology and that alarming & response is managed by SF-2-4 and/or other validated alarm system & applicable SOP(s).
 - 1.1.3. All references to SF-2-4 and Siemens revised to align with Scope.
 - 1.1.4. Add Dräger PointGard 2100 monitor, related equipment manual, and Dräger PointGard Sensor (as applicable) in Step 5.3, Section 6.1.1.3.3, and Appendix A Step 4.4.
 - 1.1.5. Step 5.13 adds clarification of compressed air tank "for E-Stop", to reduce confusion.
 - 1.1.6. Replaced redundant Section 6.1.1.3.3 O₂ Monitor settings with a reference to Appendix A, where settings are also defined.
 - 1.1.7. Added Section 6.1.3.2 directives for Wall-Mounted O2 Monitor troubleshooting.
 - 1.1.8. Step 6.2.2.1.5 directive to record all values in whole numbers was moved to Appendix C.
 - 1.1.9. Section 6.2.2.2 & Step 6.3.2.6.1 negative pressure directives revised to reflect new procedure.
 - 1.1.10. Step 6.2.2.3.1 streamlined by directing response to OOS pressure to Section 6.2.4.2.
 - 1.1.11. Corrected E-Stop air tank supply pressure upper limit in Sections 6.1.2.6 & 6.2.3.2.6.
 - 1.1.12. Add Section 6.2.3.2.10 response to high pressure on the E-Stop supply pressure gauge.
 - 1.1.13. Section 6.2.4.2 response to OOS pressure strengthened to improve safety.
 - 1.1.14. Sections 6.3.2 revised and 6.3.3 created to define entry directives for when Wall-Mounted O₂ Monitors are functioning or malfunctioning.
 - 1.1.15. Clarify in Section 6.3.2 that observer must be stationed directly outside the LN₂ room.
 - 1.1.16. Add Step 6.3.2.1.2 directive to record name of CFS after-hours watcher on Visitor Log.
 - 1.1.17. Added Step 6.3.4.3.1 directing that due to splashing risk while filling dewars with liquid nitrogen, donning a full face shield while filling dewars is preferred.
 - 1.1.18. Section 6.4 completely rewritten, reconfiguring alarm response directives to improve understanding, compliance, safety, and eliminate redundancy with Appendix F.

- 1.1.19. Step 6.4.6.7 revised to include reference to the Sensit personal O₂ monitors & SOP SF-3-3.
- 1.1.20. Section 7: Added Dräger technical support phone number & PointGard 2000 Manual.
- 1.1.21. Section 9: Added Appendix I, updated page totals, and updated Appendix titles
- 1.1.22. Appendix A revised to clarify that E-Stop activation occurs per pressure *in the LN₂ piping* and to add PointGard II and PointGard 2100 alarm parameters.
- 1.1.23. Appendix C revised to include directive to document values in whole numbers, remove erroneous LN₂ plumbing inspection remnant directive, and revise negative pressure verification procedure.
- 1.1.24. Appendix F completely rewritten to facilitate comprehension, compliance, and as follows:
 - 1.1.24.1. To reflect immediate response to alarm condition. Appendix I created to define SSF alarm response once personnel have evacuated low O₂ area.
 - 1.1.24.2. Reinstated the option to call 911 OR IUPUI Police
 - 1.1.24.3. Added response directives for C158A wall-mounted O₂ monitor alarm.
 - 1.1.24.4. CBP alarm response defined on page 2 of Appendix F as follows:
 - 1.1.24.4.1. Alarm response consolidated removing redundancy with Appendix H
 - 1.1.24.4.2. Add response to discovery of malfunctioning wall-mounted O₂ monitor.
 - 1.1.24.4.3. Add reminder not to enter LN₂ Room unless trained observer is stationed at LN₂ freezer room for safety purposes
 - 1.1.24.4.4. Further define SSF contact information

1.1.25. Appendix H revised as follows:

- 1.1.25.1. Top section revised to align with CBP training appendices in other SOPs and define criticality of LN₂ freezer room safety.
- 1.1.25.2. Added response to inadequate negative pressure at LN₂ freezer room entry.
- 1.1.25.3. Added that LN_2 freezer room entry is prohibited unless a trained watcher is stationed at the LN_2 freezer room entrance
- 1.1.25.4. Corrected Don a Personal O₂ Monitor directive to indicate that the trained watcher ensures **all personnel** working in the LN₂ room wear an O₂ monitor.
- 1.1.25.5. PPE directive corrected indicating that personnel wear approved PPE per universal precautions, removing limiting directive "when accessing samples".
- 1.1.25.6. Added Step 1.1.17 recommendation to don full face shield when filling dewars.
- 1.1.25.7. Clarified dewar fill reporting directives for billing purposes.
- 1.1.25.8. Move all alarm response directives to Appendix F to eliminate redundancy.
- 1.1.25.9. To improve understanding of safety procedures, clarify that Wall-Mounted O₂ monitors measure and report O₂ levels in the vicinity of the unit and personal O₂ monitors measure and report O₂ levels around the user.
- 1.1.25.10. Added wall-mounted O₂ monitor malfunction directive
- 1.1.26. Appendix I created to define alarm response when one or both wall-mounted O₂ Monitors are malfunctioning, and alarm response directives once personnel have evacuated low O₂ area were streamlined and relocated from Appendix F to Appendix I.

2. PURPOSE

2.1. This Standard Operating Procedure (SOP) defines the liquid nitrogen system utilized in the Indiana CTSI Specimen Storage Facility (SSF). This procedure satisfies guidance set forth in ISBER.

3. PRINCIPLE

3.1. The SSF has a hard-plumbed, vacuum-insulated, liquid nitrogen (LN₂) system that connects a 6000-gallon bulk tank to freezers to provide liquid and vapor phase nitrogen storage as a service to approved biorepositories and to provide bulk LN₂ to other IUPUI entities. This SOP defines the process for monitoring and maintaining the LN₂ system to include routine maintenance of the piping

system and safety components including emergency shut-off (E-Stop) and low oxygen alarm. Communications with the supplier, Praxair/Linde, are critical to the maintenance of the LN₂ system and are addressed in this SOP.

4. SCOPE

- 4.1. The scope of this procedure includes all trained personnel, SSF Staff or personnel who are trained per SSF SOPs, who have access to the SSF Liquid Nitrogen Freezer Room. Low O₂ alarm off-site notifications are addressed in the Facility Alarm SOP (SF-2-4) and/or other validated alarm system and the SOP(s) corresponding to that alarm system.
- 4.2. Alarm Management Definitions:
 - 4.2.1. Local Alarm: Alarm issued from equipment (audible, visual, or both)
 - 4.2.2. Remote Alarm: Alarm recorded and communicated from validated alarm system
 - 4.2.3. Alarm System: the SSF's validated alarm system
- 4.3. The SOP defines routine maintenance and monitoring of the SSF hard-plumbed LN₂ system and the room in which the system is installed (C156).

5. MATERIALS

- 5.1. Praxair/Linde bulk liquid nitrogen supply tank Model Number: TM6000
- 5.2. Emergency stop system by Chart Model Number: 11665631
- 5.3. Low O₂ Monitor(s) / Sensor
 - 5.3.1. Dräger Pointgard II, Model Number: LP3M
 - 5.3.2. Dräger Pointgard 2000 Series, Model Number: 2100
 - 5.3.3. DragerSensor O2 LS, part 6809630
- 5.4. Personal Low O₂ Monitoring Devices Manufactured by Dräger
- 5.5. Emergency escape breathing apparatus units Dräger QuickAir, 5 Minute, 4054953
- 5.6. Cryo approved gloves
- 5.7. Face Shield or Safety Goggles
- 5.8. Cryo approved apron
- 5.9. Protective gloves from cold temperatures
- 5.10. E-Stop 3-way solenoid valve (Make: Braas Company, item #: 11785288 or equivalent)
- 5.11. E-Stop regulator (.125 FPT 7-125 psi filter and gauge (Make: Braas Company, item #: 11668410 or equivalent)
- 5.12. E-Stop / Mixed air tank regulator (Make: Praxair ProStar Platinum, Model: PRS1003 or equivalent)
- 5.13. Praxair/Linde UN1002 compressed mixed air tanks (Size medical K) for E-Stop
- 6. PROCEDURE (Components of the Liquid Nitrogen System are described in Appendix A)

6.1. Maintenance

6.1.1. Quarterly

NOTE: Quarterly does not represent a calendar quarter and is defined in SF-1-1 SOP for Writing, Reviewing, and Maintaining SOPs.

- 6.1.1.1. SSF Staff checks the pipes and transfer hoses for all in service, functional units as follows:
 - 6.1.1.1. Check status of connections to determine if they are secure and tightened. Press "Start Fill" button on unit monitor and look for LN₂ leaks in the connections. If no leaks present, stop fill (if unit does not need to be filled based on digital reading). If there are leaks present, use a wrench to tighten connection. If the leak is not corrected notify SSF Management.
 - 6.1.1.1.2. Check integrity of hoses to determine if they are intact.

- 6.1.1.1.3. Record observations and actions on the LN₂ SYSTEM MAINTENANCE LOG: QUARTERLY ASSESSMENT (Appendix B1).
- 6.1.1.1.4. File records as described in Section 8.
- 6.1.1.2. If a functional unit is not present at a given location (i.e. the unit is either out of service or there is no unit at that location), mark as "N/A" for the connection status on Appendix B1. The integrity of the hoses, if present, must still be evaluated.
- 6.1.1.3. Calibrate Low O₂ Alarms
 - 6.1.1.3.1. Notify IUPUI Environmental Health and Safety to calibrate the wall-mounted Low O₂ monitors.
 - 6.1.1.3.2. Record their actions and results on the Appendix B1. File records as described in Section 8.
 - 6.1.1.3.3. After IUPUI Environmental Health and Safety completes calibration, SSF personnel must verify parameters have been returned to SSF defined settings per Appendix A. Refer to the Dräger PointGard II and Dräger PointGard 2000 Series Operating Manuals as needed.

6.1.2. Annually

- 6.1.2.1. Schedule Campus Facilities Services (CFS) to assist with Step 6.1.2.4.
- 6.1.2.2. Confirm that Praxair/Linde has scheduled and completed the annual maintenance prior to due date.
 - 6.1.2.2.1. Once completed, obtain documentation of performance from Praxair/Linde.
 - 6.1.2.2.2. Document completed or not completed on the LN₂ SYSTEM MAINTENANCE LOG: ANNUAL ASSESSMENT (Appendix B2).
 - 6.1.2.2.3. File records (to include Praxair/Linde-supplied documentation) as described in Section 8.
 - **NOTE**: Allow approximately 4 hours for completion of Steps 6.1.2.3 6.1.2.6.
- 6.1.2.3. Check external emergency shut-off valve as follows:
 - 6.1.2.3.1. Check that the valve can be turned by hand.
 - 6.1.2.3.2. Check that the flow of LN₂ liquid is stopped when the valve is in the off position.
 - 6.1.2.3.2.1. Turn valve off.
 - 6.1.2.3.2.2. Open dewar fill outlet and dispense residual LN_2 into a dewar.
 - 6.1.2.3.2.3. LN₂ will stop dispensing when lines are empty, confirming that the valve is functional.
 - 6.1.2.3.2.4. Close the dewar fill outlet.
 - 6.1.2.3.2.5. Re-open the external emergency shut-off valve.
 - 6.1.2.3.3. Record results and actions on the Appendix B2. File records as described in Section 8.
- 6.1.2.4. Check three internal vacuum piping valves as follows:
 - 6.1.2.4.1. Supervise CFS personnel verifying that valves can be turned by hand.
 - 6.1.2.4.2. Check that the flow of LN₂ liquid is stopped when the valve is in the off position.
 - 6.1.2.4.2.1. Request that CFS personnel turn valve off.
 - 6.1.2.4.2.2. SSF personnel open dewar fill outlet and dispense residual LN₂ into a dewar.

- 6.1.2.4.2.3. LN₂ will stop dispensing when lines are empty, confirming that the valve is functional.
- 6.1.2.4.2.4. SSF personnel close the dewar fill outlet.
- 6.1.2.4.2.5. Request that CFS personnel re-open the internal piping valve.
- 6.1.2.4.2.6. Repeat for other two valves.
- 6.1.2.4.3. Record name of assisting CFS personnel on Appendix B2.
- 6.1.2.4.4. Record results and actions on Appendix B2. File records as described in Section 8.
- 6.1.2.5. Check all seventeen valves connecting to the transfer hoses.
 - 6.1.2.5.1. Check that valves can be turned by hand.
 - 6.1.2.5.2. Check that the flow of LN₂ liquid is stopped when the valve is in the off position. Note: If a functional unit is not present at the location, connect a transfer hose to perform the assessment and dispense the LN₂ into a Dewar.
 - 6.1.2.5.2.1. Turn valve off.
 - 6.1.2.5.2.2. Disconnect transfer hose from LN₂ freezer at the freezer end.
 - 6.1.2.5.2.3. Allow any remaining LN₂ from the transfer hose to empty into a dewar.
 - 6.1.2.5.2.4. The LN₂ will stop draining from the transfer hose once the residual LN₂ is cleared making it possible to confirm that the valve is functional.
 - 6.1.2.5.2.5. Reconnect the transfer hose to the freezer (if functional unit is present).
 - 6.1.2.5.2.6. Re-open valve and check for leaks.
 - 6.1.2.5.2.7. Repeat for other sixteen valves.
 - 6.1.2.5.2.8. Remove transfer hose as applicable.
 - 6.1.2.5.3. Record results and actions on Appendix B2. File records as described in Section 8.
- 6.1.2.6. Check E-Stop function.
 - 6.1.2.6.1. Press red plunger button to activate E-Stop and note the time.
 - 6.1.2.6.2. Using an empty dewar at the LN₂ fill station, dispense residual LN₂ into a dewar from the transfer hose at the filling station.
 - 6.1.2.6.3. Observe that LN₂ liquid stops dispensing within a maximum of 10 minutes after activation of the E-Stop.
 - 6.1.2.6.4. Reset the E-Stop by pulling out the mushroom button and press the reset button on the Chart Controller inside C156. After resetting the E-Stop ensure that the associated compressed air cylinder continues to exhibit a delivery air pressure between 500-680 kPa and supply pressure between 300-2200 psi.
 - 6.1.2.6.5. If results meet acceptance criteria, record on Appendix B2. File records as described in Section 8.
 - 6.1.2.6.6. If results fail to meet acceptance criteria, proceed as follows:
 - 6.1.2.6.6.1. Anytime C156 is accessed by anyone, assign trained personnel to be posted outside the facility near the manual LN₂ valve on the bulk LN₂ tank to turn the valve manually should the need arise. Personal cell phones or the SSF's non-SOP-driven walkie talkies are used for communication

- between personnel watching staff working in C156 and personnel stationed at the bulk tank.
- 6.1.2.6.6.2. Inform the SSF Director (or designee) and wait approximately 4 hours to repeat the steps defined above.
- 6.1.2.6.6.3. Document the results of the second test.
 - 6.1.2.6.6.3.1. If the second test met acceptance criteria, the final assessment may be considered as a passing result.
 - 6.1.2.6.6.3.2. If the second test failed to meet acceptance criteria, contact the SSF Director and Praxair/Linde.
- 6.1.2.6.7. Repeat testing for other red plunger button and document results as defined above.

6.1.3. As Needed

- 6.1.3.1. E-Stop Replacement Parts
 - 6.1.3.1.1. Per vendor, Cryogenic Support Systems, and equipment manual recommendation, one 3-way solenoid valve and one regulator / filter should be available on site in the event of an e-stop failure.
 - 6.1.3.1.2. Replacement parts will be maintained in or near the E-Stop system control box, located in C156, or in the supply closet in C156 (C156A).
 - 6.1.3.1.3. A failed solenoid valve or regulator / filter may be replaced by CFS personnel per Cryogenic Support Systems.

6.1.3.2. Wall-Mounted O₂ Monitor Troubleshooting

- 6.1.3.2.1. The first troubleshooting step for a malfunctioning PointGard II or 2000 Series unit is replacement of the monitor's sensor (per the equipment manual). Best practice is keeping one DragerSensor O₂ LS sensor on hand.
- 6.1.3.2.2. Note: PointGard IIs are obsolete, cannot be repaired, and must be replaced if replacing the sensor did not rectify malfunction. Replace unit with the Drager PointGard 2000 series monitor.
- 6.1.3.2.3. Contact Drager technical support and provide the monitor's make (PointGard 2000 Series) and the sensor's serial number (format is likely 4 letters 4 numbers (XXXX-####))
- 6.1.3.2.4. Post signage at the impacted monitor denoting that repair is underway.
- 6.1.3.2.5. Access to the LN₂ freezer room(s) and C158A during monitor malfunction is per Section 6.3.3.

6.2. Routine Monitoring

- 6.2.1. Initial/Annual Re-Evaluation
 - 6.2.1.1. Establish LN₂ use expectations as defined on LN₂ System Expected Use Range Calculation Instructions (Appendix D):
 - 6.2.1.1.1. Record value obtained as the "expected use standard" and use as comparison.
 - 6.2.1.1.2. Re-evaluate the standard annually (or sooner if an unexplainable/unexpected change occurs). The specific weeks chosen are not defined.
 - 6.2.1.1.3. Record results and actions on the Excel spreadsheets as described in Appendix D. A template for the Excel spreadsheet is provided in Appendix D. File records as described in Section 8.

- 6.2.2. Daily
 - 6.2.2.1. Check the LN₂ level in the bulk supply tank by reading the analog gauge located near the bulk tank or via the alarm system per alarm system SOP.
 - **NOTE:** It is only permissible to obtain bulk supply tank level from the alarm system IF bulk tank level reconciliation has been completed per Section 6.2.4 within the past 6 weeks, verifying that differential between level reported by the alarm system and level from the bulk tank's analog gauge meets defined acceptance criteria.
 - 6.2.2.1.1. If the level is above 45 inches, no action is required.
 - 6.2.2.1.2. If the level is below 45 inches, contact Praxair/Linde to request replenishment and anticipated delivery date.
 - 6.2.2.1.3. NOTE: Per Praxair/Linde personnel, Praxair/Linde considers low LN₂ level acceptance criteria to equal 20". Therefore, while Praxair/Linde does monitor level, they will not automatically schedule an LN₂ fill at 45".
 - 6.2.2.1.4. If the level is below 35 inches proceed to SF-1-10 SOP for Out of Specification Condition and Notification Management.
 - 6.2.2.1.5. Record the current level and the difference from the previous scheduled workday. Indicate if the amount of LN₂ usage calculated is within the applicable expected use range and document any actions on the monitoring log (See LN₂ System Daily Use / Weekly and Monthly System Check Log (Appendix C). File records as described in Section 8.
 - 6.2.2.1.5.1. Mark "NA" if the day before was:
 - 6.2.2.1.5.1.1. Not a scheduled workday
 - 6.2.2.1.5.1.2. A day off for weather conditions
 - 6.2.2.1.5.1.3. The day following a holiday
 - 6.2.2.1.5.1.4. The first day of the work week
 - 6.2.2.1.5.2. Or, the bulk tank was filled since the last daily level check
 - 6.2.2.1.6. Notify the SSF Director if an unexplainable increase in usage (>2SD from that calculated in Step 6.2.1.1.3) is noted.
 - 6.2.2.1.7. Praxair/Linde monitors the bulk supply tank level via a cellular modem connection.
 - 6.2.2.2. Check for negative pressure differential as indicated by the Ball in Wall (BIW) comparing C156 to C158A and C156 to C199.
 - 6.2.2.2.1. Observe whether BIW in C156 to C158A is visible in C156 (yes = negative pressure = expected result). Record on Appendix C.
 - 6.2.2.2.2. Observe whether BIW in C156 to C199 is visible in C156 (yes = negative pressure = expected result). Record on Appendix C.
 - 6.2.2.2.3. If either of above observations are "no":
 - 6.2.2.3.1. Notify SSF Management.
 - 6.2.2.3.2. Notify Campus Facility Services.
 - 6.2.2.3.3. Post sign indicating that repairs are underway.
 - 6.2.2.3.4. Record actions on Appendix C.
 - 6.2.2.2.4. When resolved, remove signage.
 - 6.2.2.3. Check the LN₂ pressure in the bulk supply tank by reading the analog gauge located near the bulk tank or via the alarm system (per alarm system SOP).

NOTE: It is only permissible to obtain bulk supply tank pressure from the alarm system IF bulk tank *level* reconciliation has been completed as directed in Step 6.2.2.1 and per Section 6.2.4 within the past 6 weeks.

6.2.2.3.1. Record the pressure reading on Appendix C.

6.2.2.3.1.1. If outside of expected range (25-40 psig), proceed to Section 6.2.4.2 and record actions on Appendix C.

6.2.3. Weekly

- 6.2.3.1. Check piping in LN₂ room for any visible leaks.
 - 6.2.3.1.1. Look for frost rings which indicate a LN₂ leak.
 - 6.2.3.1.2. Record on LN₂ Weekly System Check (Appendix G).
 - 6.2.3.1.2.1. If frost rings are present, notify SSF Management.
- 6.2.3.2. Check delivery pressure and supply pressure of the E-Stop system air cylinder tank.



- 6.2.3.2.1. **Delivery Pressure:** Read the left-hand gauge of the compressed air tank connected to the E-Stop system.
 - 6.2.3.2.1.1. The compressed air tanks are located next to the Chart Controller in C156.
- 6.2.3.2.2. If the reading is between 500-680 kPa, no action needs to be taken.
- 6.2.3.2.3. If the reading is <500 kPa:
 - 6.2.3.2.3.1. Change out the spent compressed air tank with the backup compressed air tank.
 - 6.2.3.2.3.2. Contact Praxair/Linde to order an additional cylinder to replace the backup cylinder.
- 6.2.3.2.4. Record results and actions on Appendix G.
- 6.2.3.2.5. A tank regulator ensures that pressure does not exceed 680 kPa. Should the regulator malfunction and pressure exceed 680 kPa:
 - 6.2.3.2.5.1. Post a sign to not enter C156.
 - 6.2.3.2.5.2. Notify SSF Management and proceed to SF-1-10.
 - 6.2.3.2.5.3. Contact Choice Mechanical or Cryogenic Support Systems to evaluate and/or repair e-stop system.
- 6.2.3.2.6. **Supply Pressure:** Read the right-hand gauge of the air tank connected to the E-Stop system.
- 6.2.3.2.7. If the reading is between 300-2200 psi, no action needs to be taken.
- 6.2.3.2.8. If the reading is <300 psi:
 - 6.2.3.2.8.1. Change out the spent mixed air tank with the backup mixed air tank.
 - 6.2.3.2.8.2. Contact Praxair/Linde to order an additional cylinder to replace the backup cylinder.

- 6.2.3.2.9. Low supply pressure in the air tank will cause the E-Stop system to shut down, discontinuing LN₂ supply to C156.
- 6.2.3.2.10. If reading is >2200 psi:
 - 6.2.3.2.10.1. Close supply valve on the air tank.
 - 6.2.3.2.10.2. Change out the air tank with the backup mixed air tank.
 - 6.2.3.2.10.3. Notify SSF Management and proceed to SF-1-10.
 - 6.2.3.2.10.4. Contact Praxair/Linde to replace tank.
- 6.2.3.2.11. Record results and actions on Appendix G.
- 6.2.4. After each bulk supply tank fill or as needed
 - 6.2.4.1. Check the gauge on the bulk supply tank as follows:
 - 6.2.4.1.1. Check the LN₂ supply level, obtained from the analog gauge at the bulk tank, measuring in inches of water.
 - 6.2.4.1.1.1. LN₂ Supply level values are recorded in inches of water column, with approximation of pressure recorded the nearest whole number.
 - 6.2.4.1.2. Compare the level from the tank to the alarm system level, if applicable.
 - 6.2.4.1.2.1. If the difference is < 7.0 inches, no action is required.
 - 6.2.4.1.2.2. If the difference is > 7.0 inches, notify the SSF Director.
 - 6.2.4.1.3. Record results (rounding to the nearest whole number) and actions on Appendix E. File records as described in Section 8.
 - 6.2.4.2. Pressure level
 - 6.2.4.2.1. Check the bulk supply tank pressure level, measured in psig.
 - 6.2.4.2.1.1. Normal pressure range is 25-40 psig. No action is required if the pressure level on the tank is within this range.
 - 6.2.4.2.1.2. If pressure is above 40 psig:
 - 6.2.4.2.1.2.1. Activate E-Stop
 - 6.2.4.2.1.2.2. Post signage indicating that LN₂ freezer room entry is prohibited and notify impacted personnel.
 - 6.2.4.2.1.2.3. Turn off LN₂ supply at LN₂ bulk tank.
 - 6.2.4.2.1.2.4. Notify Praxair/Linde to release pressure and resolve issue.
 - 6.2.4.2.1.2.5. Re-entry prohibited until pressure meets acceptance criteria. Re-entry may occur prior to Praxair/Linde inspection IF pressure meets acceptance criteria.
 - 6.2.4.2.1.2.6. Document OOS condition per SF-1-10.
 - 6.2.4.2.1.3. If pressure is below 25 psig, contact Praxair/Linde, notify SSF Management, and document OOS condition per SF-1-10.
 - 6.2.4.2.2. Record results and actions on LN₂ Systems Post-Fill Observation Log (Appendix E). File records as described in Section 8.

6.3. Entry Into LN₂ Freezer Room

6.3.1. An authorization for access is obtained as described in the Controlled Access Operations SOP SF-2-3.

- 6.3.2. During operations in which *all Wall-Mounted O₂ Monitors are functioning as expected*, before staff enter the LN₂ Freezer Room, perform the following:
 - 6.3.2.1. Post an observer at the C158A/C156 door to monitor personnel who have entered the LN₂ freezer room. The observer must be stationed directly outside the LN₂ freezer room.
 - 6.3.2.1.1. Prior to serving as an observer, the individual must receive verbal instruction on emergency response procedures per Section 6.4 and Appendix F of this SOP, unless the observer has documented training on the current required version of SF-2-2.
 - 6.3.2.1.2. CFS personnel may serve as observers after hours. Record name of observer on Visitor Log, SF-2-3 Appendix D.
 - 6.3.2.2. Obtain a personal O₂ monitor for each personnel entering the room according to the following priorities and exceptions:
 - 6.3.2.2.1. If the number of personnel entering the LN₂ Freezer Room at a given time exceeds the number of functional O₂ monitors, the personal O₂ monitors will be utilized by personnel who will:
 - 6.3.2.2.1.1. Be in the LN₂ freezer room for the longest duration AND
 - 6.3.2.2.1.2. Have the closest proximity to an open liquid nitrogen source.
 - 6.3.2.2.2. If the number of functional O₂ monitors fall below 2, entry into the LN₂ freezer room will be limited to 1 personnel unless a deviation is approved by SSF Management and IUPUI Environmental Health and Safety.
 - 6.3.2.3. If there are available additional unused personal O₂ monitors, personnel entering the LN₂ Freezer Room will wear a low O₂ monitor until either (a) all personal O₂ monitors are in use or (b) each person in the LN₂ Freezer Room is wearing a personal O₂ monitor. Priorities are as described in Step 6.3.2.2.1.
 - 6.3.2.4. Verify that the oxygen level on the low O₂ alarm registers above 19.5%,
 - 6.3.2.5. Verify that the bulk liquid nitrogen tank is not being filled (DO NOT ENTER WHILE THE BULK TANK IS FILLING DUE TO POTENTIAL FOR WHITE OUT CONDITION IF THE SYSTEM MALFUNCTIONS).
 - 6.3.2.6. Verify negative pressure differential by observing that the Ball in Wall (BIW) indicator is visible on the C156 side of the wall and NOT IN C158A.
 6.3.2.6.1. If negative pressure is inadequate, follow Section 6.2.2.2 directives.
- 6.3.3. Entry is limited per the following directives when <u>Wall-Mounted O₂ Monitor(s) Malfunction</u>: 6.3.3.1. Malfunction of C158A Wall-Mounted O₂ Monitor:
 - 6.3.3.1.1. Personnel may continue working in C158A, since the personal O₂ monitors housed below the monitor will continue measuring O₂ level in its vicinity.
 - 6.3.3.1.1.1. Station a personal O₂ monitor as close as possible to the malfunctioning monitor to measure O₂ level in its vicinity.
 - 6.3.3.1.1.2. Personnel access to C156 is thereby limited based on the remaining number of available personal O₂ monitors per Section 6.3.2.2.
 - 6.3.3.1.2. Personnel may continue accessing C156.
 - 6.3.3.1.3. Alarm response is per Appendix F, Emergency Contacts, Wall-Mounted Low O₂ Monitors Immediate Actions & Response, and Collaborating Biobank Personnel (CBP) Alarm Response and Malfunction Response Directives.

- 6.3.3.2. Malfunction of LN₂ Freezer Room Wall-Mounted O₂ Monitor(s):
 - 6.3.3.2.1. Post signage at LN₂ Freezer Room entry doors stating that "LN₂ Freezer Room access is restricted See SSF personnel," or similar.
 - 6.3.3.2.2. Rescind CBP access to the LN₂ Freezer Room per SF-2-3 until both wall-mounted O₂ monitors are functioning as expected.
 - 6.3.3.2.3. Notify CBP of safety issue and that CBP access is only permitted with SSF assistance and SSF watcher until both wall-mounted O₂ monitors are functioning as expected. Provide CBP with safe access directives via notification email and in person when accessing facility.
 - 6.3.3.2.4. Place a personal O₂ monitor as close as possible to the malfunctioning monitor to measure O₂ level in its vicinity.
 - 6.3.3.2.5. Personnel access is limited based on the remaining number of available personal O₂ monitors.
 - 6.3.3.2.6. Only SSF personnel can serve as safety watchers.
 - 6.3.3.2.7. Alarm response is per Appendix I.
- 6.3.4. Personnel entering the cell repository must comply with the following directives:
 - 6.3.4.1. Gown and glove per universal precautions when accessing samples or performing freezer monitoring or maintenance.
 - 6.3.4.2. Wear a personal O₂ monitor. (See Drager Pac 5500 Personal O₂ Monitor SOP SF-3-15)
 - 6.3.4.3. When working with liquid nitrogen in the liquid phase, wear chemical splash goggles or a full face shield; wear approved cryogenic gloves and an apron.
 6.3.4.3.1. Since splashing can occur while filling dewars with liquid nitrogen, donning a full face shield while filling dewars is preferred.
 - 6.3.4.4. When working with liquid nitrogen in the vapor phase, wear appropriate heavy gloves (i.e. nitri-flex lite gloves, cryogloves, or similar).
 - 6.3.4.5. Personnel accessing samples should minimize sample exposure to room temperature by returning racks to the LN₂ freezers as soon as possible.
- 6.3.5. Instruct visitors to don lab coat, gloves, face or eye protection, and personal low oxygen monitor if indicated in Section 6.3.2.2. Provide information regarding alarm locations, alarm responses, LN₂ safety precautions, and emergency escape breathing apparatus units before entering the LN₂ freezer room.

6.4. SSF Alarm Actions and Response

- 6.4.1. Emergency Contacts, Wall-Mounted Low O₂ Monitors Alarm Immediate Actions & Response, and Collaborating Biobank Personnel (CBP) Alarm Response and Malfunction Discovery Directives are defined in Appendix F.
 - 6.4.1.1. While the Table of Contents and SOP SF-1-6 for Controlled Document Management define that Appendix F is posted at SSF LN₂ freezer rooms and adjacent spaces, the following table defines locations for posting each page of Appendix F. NOTE: Appendix F Pages 1, 2, and 7 are not required postings.

Page	Title	Posting Requirement
1	Appendix F Table of Contents	Not posted
2	Collaborating Biobank Personnel Alarm Response and Malfunction Response Directives	Not posted
3	Immediate Alarm Response for Personnel Working in LN ₂ Freezer Room	At each wall-mounted O ₂ monitor in an LN ₂ freezer room
4	Alarm Actions & Response for Staff Monitoring Personnel Working in LN ₂ Freezer Room	At LN ₂ freezer room entrances
5	Immediate Alarm Response for C158A Wall-Mounted O ₂ Monitor	At the wall-mounted O ₂ monitor in R3-C158A
6	Alarm Actions & Response following alarm on C158 Wall-Mounted O ₂ Monitor	At each entrance to R3-C158A
7	SSF Personnel Response to C158A Wall- Mounted Low O ₂ Monitor - SSF Personnel Follow-Up Actions	Not posted
8	Emergency Contacts	At LN ₂ freezer room entrances

- 6.4.2. Appendix I defines SSF Wall-Mounted Low O₂ Monitors Alarm Monitoring / Notification and Alarm Malfunction Response Actions, and Appendix I directives are followed only once Appendix F directives have been carried out.
 - 6.4.2.1. Response to wall-mounted low O₂ monitor alarms (whether functional or malfunctioning) is per Appendix I. Directives are applicable only <u>once all personnel</u> have vacated the low O₂ area.
- 6.4.3. <u>Personal low O₂ monitor alarm response</u>
 - 6.4.3.1. If the audible and/or vibrating alarm activates, stop all activity and move away from the nitrogen source.
 - 6.4.3.2. Check O₂ monitor on wall to verify that reading is greater than or equal to 19.5%.
 - 6.4.3.3. Once confirmed that the room O₂ is within acceptable range above, press the green OK button to silence the audible alarm.
 6.4.6.3.1. Personal O₂ monitor alarm will not clear until it's reading ≥19.5% O₂.
 - 6.4.6.4. Once the personal O₂ monitor is within range and is not in alarm, resume activities.
 - 6.4.6.5. If alarm recurs more than two times successively, vacate the room. Verify the O₂ level in room is within range.
 - 6.4.6.5.1. If O₂ level is demonstrated to be adequate by the wall mounted oxygen monitors, obtain an alternate O₂ monitor and return to room.
 - 6.4.6.6. CLARIFICATION: Best practice is for O₂ monitors to be worn near the user's head. On occasion, personal O₂ monitors will alarm because the monitor's air intake has been obstructed or a monitor suspended from around the neck descends into an LN₂ freezer. O₂ monitors should NOT be placed in the user's pocket and should NOT be worn suspended by the lanyard from around the user's neck. Personal O₂ monitor alarm response is per Steps 6.4.4.1 6.4.4.5.
 - 6.4.6.7. Operation of the low oxygen monitors is described in the Sensit P100 Personal O₂ Monitors SOP, SF-3-3, and the Drager Pac 5500 Personal O₂ Monitors SOP, SF-3-15.
- 6.4.7. **CBP Alarm Actions and Response** Refer to Appendix F and Appendix H, Collaborating Biorepository Personnel Training.

6.5. Liquid Nitrogen Dispensation

- 6.5.1. Requests to fill LN₂ Dewars are completed as follows:
 - 6.5.1.1. PI staff submit LN₂ requests to SSF staff (which may be hard copy or web-based) to include the following:
 - 6.5.1.1.1. Number of dewars to be filled.
 - 6.5.1.1.2. The size of the dewar(s) to be filled.
 - 6.5.1.1.3. Date and time requested for fill.
 - 6.5.1.1.4. Contact person's email.
 - 6.5.1.2. Weigh the dewar on the scale before dispensing LN₂.
 - 6.5.1.3. Weigh the dewar after the dispensing is completed.
 - 6.5.1.4. Calculate the pounds (lbs) of LN₂ used.
 - 6.5.1.5. Invoice per the IUSOM recharge policy for the LN₂ rate.
 - 6.5.1.6. Note: Invoicing policies and rates are determined by the Operations Oversight Committee and approved by Indiana University.

6.5.2. If LN₂ fails to be dispensed

- 6.5.2.1. Check that the E-stop is not activated.
- 6.5.2.2. Confirm adequate delivery and supply air pressure in the E-stop compressed air cylinder (refer to Section 6.2.3.2 for instructions on how to confirm and corrective actions).
- 6.5.2.3. Confirm adequate LN₂ pressure in the supply tank (refer to Section 6.2.4.2 for instructions on how to confirm and corrective actions).
- 6.5.3. If LN₂ still fails to be dispensed, notify SSF Management.

6.6. Non-Routine Monitoring

6.6.1. Documentation of OOS conditions that occur at times other than routine monitoring which DO NOT generate an alarm is optional.

7. REFERENCES

- 7.1. ISBER Best Practices (current version)
- 7.2. Campus Facilities Services (CFS) Switchboard: 317-278-1900
- 7.3. Choice Mechanical: 317-885-0200, Toll Free 1-877-885-9200 or www.choicemechanical.net
- 7.4. Cryogenic Support Systems: 765-884-0031; 5790 N 100 E Fowler, Indiana 47944
- 7.5. IUPUI Police: 317-274-7911
- 7.6. Dräger technical support at 800-437-2437
- 7.7. Dräger PointGard II Operating Manual
- 7.8. Dräger PointGard 2000 Series Operating Manual
- 7.9. Praxair/Linde Local: 317-554-2200; National: 1-800-772-9247

8. DOCUMENTATION

- 8.1. LN₂ System and LN₂ Freezer monitoring logs are reviewed by SSF Management and are maintained per SF-1-6 Controlled Document Management SOP.
- 8.2. LN₂ System and LN₂ Freezer maintenance logs (including Praxair/Linde supplied documentation) are reviewed by SSF Management and are maintained for two years in the SSF Operations Office per SF-1-6 Controlled Document Management SOP.
- 8.3. Deviations are managed and documented per SF 1-9 Deviation Management SOP
- 8.4. Storage space in the SSF LN₂ Room or Liquid Nitrogen freezers is managed as defined per SOP SF-1-4 Managing Storage Space.

9. APPENDICES

9.1. The current version of each of the following appendices is used to guide and/or implement this SOP:

Appendix A: Components of the Liquid Nitrogen System (2 Pages)

Appendix B1: LN₂ System Maintenance Log - Quarterly Assessment (1 Page) Appendix B2: LN₂ System Maintenance Log - Annual Assessment (2 Pages)

Appendix C: LN₂ System Daily Use/Weekly and Monthly System Check Log (1 Page)
Appendix D: LN₂ System Expected Use Range Calculation Instructions (2 Pages)

Appendix E: LN2 Systems Post-Fill Observation Log (1 Page)

Appendix F: Emergency Contacts, Wall-Mounted Low O2 Monitors Immediate Actions &

Response, and Collaborating Biobank Personnel (CBP) Alarm Response and

Malfunction Response Directives (8 Pages)

Appendix G: LN₂ Weekly System Check (1 Page)

Appendix H: Collaborating Biorepository Personnel Training (2 Pages)

Appendix I: SSF Wall-Mounted Low O₂ Monitors Alarm Monitoring / Notification and Alarm

Malfunction Response Actions (2 Pages)

10. COLLABORATING BIOBANK TRAINING DIRECTIVES

10.1. CBP comply with Read and Understand training on SOP SF-2-2, SOP for LN₂ System and LN₂ Freezer Room Operations per SF-2-3, SOP for Controlled Access Operations, by continuing to and reading Collaborating Biorepository Personnel Training, Appendix H, of this SOP.

10.2. CBP comply with directives defined in Appendix H.

Appendix A Page 1 of 2

Components of the Liquid Nitrogen System

1.0 Bulk Supply Tank

- 1.1 The liquid nitrogen (LN₂) is stored in a bulk supply tank to the exterior and adjacent to the LN₂ Freezer Room (C156).
 - 1.1.1 The key to access the LN_2 bulk tank is located in the SSF Management Office.
 - 1.1.2 The manual shut off is located outside of the building to the eastside of room C156 in the LN₂ Bulk Supply Gate on the left, bottom side of the tank and is labeled as such.

1.2 Bulk Supply Source

- 1.2.1 Liquid nitrogen is supplied by Praxair/Linde Healthcare Services at 144 Polco Street, Indianapolis, IN 46222; 1-800-PRAXAIR.
 - 1.2.1.1 Praxair/Linde is contracted to monitor and replenish the supply when the bulk tank reaches a defined level.
- 1.2.2 Remote access for monitoring the LN₂ level of the bulk tank is provided via the alarm system.

2.0 LN₂ Piping

- 2.1 The LN₂ is piped into the building via vacuum-insulated piping.
 - 2.1.1 The piping runs through the room with the venting point being at the highest level.
 - 2.1.2 The piping has separately controlled access valves.
- 2.2 The LN₂ is dispersed into transfer hoses which connect to the liquid nitrogen freezers.

3.0 E-Stop System

- 3.1 The E-Stop System is located inside the liquid nitrogen freezer room on the south wall.
 - 3.1.1 The E-Stop system is connected to the bulk tank. A solenoid valve stops the supply of LN₂ to the vacuum-insulated piping when there is a loss of pressure. The valve opens and closes based on pneumatic activation of pressure supplied by a compressed air tank.
 - 3.1.1.1 The E-Stop activates automatically when pressure in the LN₂ piping is too low or high and/or the Low O₂ alarm is activated.
 - 3.1.1.2 The E-stop may be activated manually by depressing the red plunger button located inside the LN₂ freezer room (C156) or inside the processing lab (C158A).
 - 3.1.1.3 A regulator monitors delivery and supply pressure in the compressed air tank.
 - 3.1.1.3.1 Delivery pressure indicates pressure to the E-Stop and is monitored by the left-hand gauge on the regulator (see picture in Step 3.1.1.3.3.
 - 3.1.1.3.2 Supply pressure indicates air remaining in air tank.



3.1.2 Press the E-Stop red reset button to reset the E-Stop system.

Appendix A Page 2 of 2

4.0 Low Oxygen Alarms (Wall-Mounted)

- 4.1 There are three Low oxygen alarms: two are located inside the LN₂ Freezer room, and the third is located in the adjacent processing lab. The three alarms are **not** networked.
- 4.2 Alarms activate when oxygen levels in the immediate area of the alarm location reaches a defined critical level.
 - 4.2.1 When oxygen level reaches a critical level (<19.5%) on the unit connected to the alarm system, a remote notification is dispatched.
 - 4.2.2 The unit connected to the alarm system is on the south wall of C156.
- 4.3 These units must maintain 4 critical parameters to ensure safety for all personnel.
 - 4.3.1 Latching: Unit will remain in alarm even after the alarm condition has returned to normal until the alarm is reset manually.
 - 4.3.2 Non-acknowledgeable / Not Acknowledgeable: The alarm cannot be reset while the O₂ level is below or at alarm conditions.
 - 4.3.3 Alarm point 1 (A1) is triggered by a drop in O₂ at or below 19.5% concentration.
 - 4.3.4 Alarm point 2 (A2) is triggered by a drop in O₂ at or below 19.0% concentration.
 - 4.3.5 Programming for the Drager PointGard II and Drager PointGard 2100 Parameters:
 - 4.3.6 PointGard II Parameters:
 - 4.3.6.1 A1 Lat = Latching
 - 4.3.6.2 A2 Lat = Latching
 - 4.3.6.3 A1 Acn = Non-acknowledgeable
 - 4.3.6.4 A2 Acn = Non-acknowledgeable
 - 4.3.6.5 A1-Adj = 19.5
 - 4.3.6.6 A2-Adj = 19.0
 - 4.3.7 PointGard 2100 Parameters:
 - 4.3.7.1 A1 Alarm = Enabled
 - 4.3.7.2 A2 Alarm = Enabled
 - 4.3.7.3 A1 Alarm Setpoint = 19.5
 - 4.3.7.4 A2 Alarm Setpoint = 19.0
 - 4.3.7.5 A1 Latch Mode Setpoint = Latching
 - 4.3.7.6 A2 Latch Mode Setpoint = Latching
 - 4.3.7.7 A1 Acknowledgement Mode = Not Acknowledgeable
 - 4.3.7.8 A2 Acknowledgement Mode = Not Acknowledgeable
- 4.4 Refer to Dräger PointGard II and Dräger PointGard 2000 Series Operating Manuals.

5.0 Emergency Escape Breathing Apparatus Units are located within C156.

6.0 System features for dispensing LN₂ manually

- 6.1 One hose is available for dispensing liquid nitrogen manually.
- 6.2 A scale featuring an attached meter with a digital readout is used to weigh the amount of LN₂ dispensed.

Appendix B1 Page 1 of 1

LN ₂ System Maintenance Log: QUARTERLY ASSESSMENT Quarter Year:											
QUARTERLY ASSESSMEN	T	is	nnection to Secure & tig ocument Res	ght*	Integrity of Hose is intact** Document Result $()$		als	Date	Comments / Corrective Actions		
Acceptance C	riteria:	M			Mo						
Valve for Freez	zer A1		Met Not Met	□ N/A			Met				
Valve for Freez	zer A2		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer A3		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer B1		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer B2		Met Not Met	N/A		Met Not	Met				
Valve for Freez	zer B3		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer B4		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer B5		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer C1		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer C2		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer C3		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer C4		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer C5		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer D1		Met Not Met	□ N/A		Met Not	Met				
Valve for Freez	zer D2		Met Not Met	□ N/A		Met Not	Met				
Valve for Freezer D3			Met Not Met	□ N/A			Met				
Valve for Freez	zer D4		Met Not Met	□ N/A		Met Not	Met				
Low Oxygen Alarm Calibration	Co	mpl	bration eted Date)	Confirm Settings R to As Foun (Tech/I	etur d Va	ned lues	EHS Re Rec'd/atta (Tech/D	ached		S Report ceptable	
C156 North									□ Y	es 🗖 No	
C156 South									□ Y	es 🗆 No	
C158A									□ Y	es 🗆 No	
Reviewed By /	Date:						•				

^{*}For locations where units are out of service or absent, document "N/A."

^{**}For locations where units are out of service or absent, integrity of hose, if applicable, must still be evaluated.

Appendix B2 Page 1 of 2

				۰
v	69	r	•	
	$\mathbf{L}a$		•	

LN2 SYSTEM MAINTENANCE LOG: ANNUAL VALVE ASSESSMENT								
ANNUAL VALVE ASSESSMENT	Document Result ($$): Note: Expected Result = Yes	Initials	Date	Comments/Corrective Actions				
External valve Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
*								
Internal Valve #1 Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
Internal Valve #2	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Internal Valve #3	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer A1	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer A2	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer A3 Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
Valve for Freezer B1	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer B2	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer B3	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer B4	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer B5 Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
Valve for Freezer C1	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer C2	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer C3	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer C4 Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
Valve for Freezer C5	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer D1 Turned by Hand	☐ Yes ☐ No☐ Not Applicable							
Valve for Freezer D2	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer D3	☐ Yes ☐ No							
Turned by Hand	☐ Not Applicable							
Valve for Freezer D4								
Turned by Hand								
Name of CFS Personnel Assisting with Internal Valve #1-3 Testing Initials / Date								
Reviewed By / Date:								

Appendix B2 Page 2 of 2

Year:		
ı caı.		

National	LN ₂ SYSTEM MAINTENANCE LOG: ANNUAL VALVE ASSESSMENT, cont.							
External valve LAN Flow Stopped in Off Position Not Applicable Not Applicable LAN Flow Stopped in Off Position Not Applicable No			AL VALV	L ASSE				
Valve Passessment			Initials	Date				
Internal Valve # 1		1			Actions			
Internal Valve # 1								
INZ Flow* Stopped in Off Position	**	**						
Internal Valve #3								
Internal Valve # 3								
Internal Valve #3								
Not Applicable Not								
Valve for Freezer A1								
Not Applicable Not Applicable Not Applicable	LN2 Flow* Stopped in Off Position	**						
Valve for Freezer A2		☐ Yes ☐ No						
Not Applicable Not								
Valve for Freezer B2	Valve for Freezer A2	☐ Yes ☐ No						
Not Applicable Not	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer B1	Valve for Freezer A3	☐ Yes ☐ No						
Not Applicable	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer B2	Valve for Freezer B1	☐ Yes ☐ No						
LN2 Flow Stopped in Off Position Valve for Freezer B3 LN2 Flow Stopped in Off Position Valve for Freezer B4 LN2 Flow Stopped in Off Position Valve for Freezer B5 LN2 Flow Stopped in Off Position Valve for Freezer B5 LN2 Flow Stopped in Off Position Valve for Freezer B5 LN2 Flow Stopped in Off Position Valve for Freezer C1 LN2 Flow Stopped in Off Position Valve for Freezer C2 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D2 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D5 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer B3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer B4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer B5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Not Applicable LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4	Valve for Freezer B2	☐ Yes ☐ No						
Valve for Freezer B3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer B4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer B5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Not Applicable LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Not Applicable LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 </td <td>LN2 Flow Stopped in Off Position</td> <td>☐ Not Applicable</td> <td></td> <td></td> <td></td>	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer B4 LN2 Flow Stopped in Off Position Valve for Freezer B5 LN2 Flow Stopped in Off Position Valve for Freezer C1 LN2 Flow Stopped in Off Position Valve for Freezer C2 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D2 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Document Result (√) Note: Expected Result = Yes No C156 E-Stop stops flow of LN2* within 10 minutes of activation C158A E-Stop stops flow of LN2* within 10 minutes of activation Valve Or Freezer D3 C158A E-Stop stops flow of LN2* within 10 minutes of activation	Valve for Freezer B3							
Valve for Freezer B4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer B5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Not Applicable LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL □ Yes	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer B5	Valve for Freezer B4							
Valve for Freezer B5	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer C1 LN2 Flow Stopped in Off Position Valve for Freezer C2 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D2 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position ANNUAL Monitoring and PM Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN₂* within 10 minutes of activation □ Yes □ No Within 10 minutes of activation □ Yes □ No Within 10 minutes of activation □ Yes □ No	Valve for Freezer B5							
LN2 Flow Stopped in Off Position	LN2 Flow Stopped in Off Position	☐ Not Applicable						
University of the property of	Valve for Freezer C1	☐ Yes ☐ No						
Valve for Freezer C2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Not Applicable LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No C156 E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No □ Yes □ No □ Yes □ No <td></td> <td></td> <td></td> <td></td> <td></td>								
LN2 Flow Stopped in Off Position Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D2 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position ANNUAL Monitoring and PM Praxair PM Acceptable C156 E-Stop stops flow of LN₂* within 10 minutes of activation C158A E-Stop stops flow of LN₂* within 10 minutes of activation C158A E-Stop stops flow of LN₂* within 10 minutes of activation								
Valve for Freezer C3 LN2 Flow Stopped in Off Position Valve for Freezer C4 LN2 Flow Stopped in Off Position Valve for Freezer C5 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D1 LN2 Flow Stopped in Off Position Valve for Freezer D2 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D3 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position Valve for Freezer D4 LN2 Flow Stopped in Off Position ANNUAL Monitoring and PM Praxair PM Acceptable C156 E-Stop stops flow of LN2* within 10 minutes of activation C158A E-Stop stops flow of LN2* within 10 minutes of activation Pyes □ No Not Applicable □ Yes □ No □ Not Applicable □ Y		☐ Not Applicable						
Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL □ Not Applicable Monitoring and PM □ Document Result (√) Note: Expected Result = Yes Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN₂* within 10 minutes of activation □ Yes □ No C158A E-Stop stops flow of LN₂* within 10 minutes of activation □ Yes □ No Yes □ No								
Valve for Freezer C4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL □ Not Applicable Monitoring and PM □ Document Result (√) Note: Expected Result = Yes Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN₂* within 10 minutes of activation □ Yes □ No C158A E-Stop stops flow of LN₂* within 10 minutes of activation □ Yes □ No Yes □ No	LN2 Flow Stopped in Off Position	☐ Not Applicable						
Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL Document Result (1) Note:								
Valve for Freezer C5 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL Document Result (1) Note:								
United to the stopped in Off Position								
Valve for Freezer D1 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL Document Result (1) Note: Expected Result = Yes Initials Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No C158A E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No Yes □ No								
	11	□ Yes □ No						
Valve for Freezer D2 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D3 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable Valve for Freezer D4 □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable ANNUAL Document Result ($$) Note: Monitoring and PM Expected Result = Yes Praxair PM Acceptable □ Yes □ No C156 E-Stop stops flow of LN2* □ Yes □ No within 10 minutes of activation □ Yes □ No Within 10 minutes of activation □ Yes □ No								
		i						
Valve for Freezer D3 LN2 Flow Stopped in Off Position \square Yes \square No \square Not Applicable \square Yes \square No \square Not ApplicableValve for Freezer D4 LN2 Flow Stopped in Off Position \square Yes \square No \square Not Applicable \square InitialsDateComments/Corrective ActionsANNUAL Monitoring and PM \square Document Result ($\sqrt{\square}$) Note: \square Expected Result = YesInitialsDateComments/Corrective ActionsPraxair PM Acceptable C156 E-Stop stops flow of LN2* within 10 minutes of activation \square Yes \square No \square Yes \square No								
LN2 Flow Stopped in Off Position □ Not Applicable □ Yes □ No LN2 Flow Stopped in Off Position □ Not Applicable □ Not Applicable ANNUAL Monitoring and PM Document Result ($$) Note: Expected Result = Yes Initials Date Comments/Corrective Actions Praxair PM Acceptable □ Yes □ No □ Yes □ No □ Yes □ No C156 E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No □ Yes □ No C158A E-Stop stops flow of LN2* within 10 minutes of activation □ Yes □ No	**							
Valve for Freezer D4 LN2 Flow Stopped in Off Position \square Yes \square No \square Not Applicable \square Not Applicable \square Comments/Corrective ActionsANNUAL Monitoring and PM \square Document Result $(\sqrt{)}$ Note: Expected Result = YesInitials \square DateComments/Corrective ActionsPraxair PM Acceptable C156 E-Stop stops flow of LN2* within 10 minutes of activation \square Yes \square No \square Yes \square NoC158A E-Stop stops flow of LN2* within 10 minutes of activation \square Yes \square No								
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		i						
ANNUAL Monitoring and PM Praxair PM Acceptable C156 E-Stop stops flow of LN_2^* within 10 minutes of activation C158A E-Stop stops flow of LN_2^* within 10 minutes of activation Wes \square No Yes \square No Yes \square No Yes \square No								
Monitoring and PMExpected Result = YesInitialsDateActionsPraxair PM Acceptable \square Yes \square No \square Yes \square NoC156 E-Stop stops flow of LN2* within 10 minutes of activation \square Yes \square No \square Yes \square NoC158A E-Stop stops flow of LN2* within 10 minutes of activation \square Yes \square No	• •	,			Comments/Corrective			
Praxair PM Acceptable \square Yes \square No C156 E-Stop stops flow of LN ₂ * within 10 minutes of activation \square Yes \square No C158A E-Stop stops flow of LN ₂ * within 10 minutes of activation \square Yes \square No		` '	Initials	Date				
C156 E-Stop stops flow of LN_2^* within 10 minutes of activation C158A E-Stop stops flow of LN_2^* within 10 minutes of activation Yes \square No Wes \square No		*			ACHORS			
within 10 minutes of activation C158A E-Stop stops flow of LN ₂ * within 10 minutes of activation Yes \(\text{No} \) Yes \(\text{No} \) Yes \(\text{No} \)		LI res LI NO						
within 10 minutes of activation C158A E-Stop stops flow of LN_2^* within 10 minutes of activation Yes \square No		☐ Yes ☐ No						
within 10 minutes of activation		_ 100 _ 110						
within 10 minutes of activation		□ Ves □ No						
Reviewed By / Date:	within 10 minutes of activation	— 1 CS — 1 NO						
Reviewed By / Date:								
	Reviewed By / Date:							

^{*} In order to meet acceptance criteria, flow of *liquid* LN₂ must stop.

Appendix C Page 1 of 1

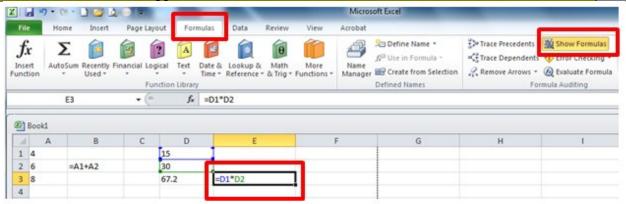
	Month: Year:									
				I	N ₂ SYST	EM DAILY LN ₂ US	AGE L	OG		
Note	Note: Liquid Nitrogen Level Expected Result ≥ 45.0 inches									
	Liquid Nitrogen Pressure Expected Range = 25-40 psig									
Ente	er Curr	ent Exp	ected U	se Rang	$ge (Avg. \pm i)$	2 SD):		Note: Pressure and level values are recorded in whole numbers.		
			1		DAILY					
		n Wall ble in	Curre	nt LN ₂						
		156			Level					
	158A	199 -	Pres-		Change	LN ₂ use is within the	Tech	Comments/ Corrective Actions		
Day	-156	156 Z/N)	sure	Level (inches)	from Previous	Expected Use Range:	Initials			
	If "No"	, refer to	(psig)	(inenes)	Day					
		document ons in								
1	commo	ent field				□Yes □No □NA				
2						☐Yes ☐No ☐NA				
3						☐Yes ☐No ☐NA				
4						☐Yes ☐No ☐NA				
5						☐Yes ☐No ☐NA				
6						☐Yes ☐No ☐NA				
7						☐Yes ☐No ☐NA				
8						☐Yes ☐No ☐NA				
9						☐Yes ☐No ☐NA				
10						☐Yes ☐No ☐NA				
11						☐Yes ☐No ☐NA				
12						□Yes □No □NA				
13						□Yes □No □NA				
14						□Yes □No □NA				
15						□Yes □No □NA				
16						□Yes □No □NA				
17						□Yes □No □NA				
18						□Yes □No □NA				
19						□Yes □No □NA				
20						□Yes □No □NA				
21						□Yes □No □NA				
22						□Yes □No □NA				
23						□Yes □No □NA				
24						□Yes □No □NA				
25						□Yes □No □NA				
26						□Yes □No □NA				
27						□Yes □No □NA				
28						☐Yes ☐No ☐NA				
29						☐Yes ☐No ☐NA				
30						□Yes □No □NA				
31						□Yes □No □NA				
Rev	iewed	By / D	ate:							

Appendix D Page 1 of 2

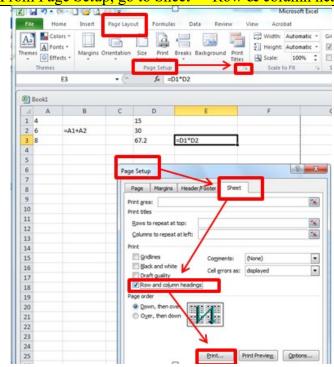
LN₂ Expected Use Range Calculation Instructions

- 1. Determine the amount of LN₂ used by subtracting the LN₂ level of the previous day from the current LN₂ level.
- 2. Enter this data as day 1 usage in an Excel spreadsheet. Include a column in which to document the actual date (a spreadsheet template is provided on page two).
- 3. Perform the same calculation for the next 27 consecutive days
 - 3.1. Omit the following days:
 - 3.1.1. All days marked "N/A" per Steps 6.2.2.1.4.1 6.2.2.1.4.2.
 - 3.1.2. Days greyed out on SF-2-2, Appendix C by SSF personnel (i.e. weekends, holidays)
 - 3.2. Since the days above are omitted, to obtain 27 more days it is acceptable to continue into the next month omitting the same days listed above.
- 4. After 28 days of data have been obtained, use Microsoft Excel to determine the average amount used over 28 days
- 5. Use Excel to calculate the standard deviation
- 6. Multiply the standard deviation by 2.
- 7. Calculate the upper limit of the expected use range as "= average + (2*stdev)" (rounding up to the nearest whole number)
- 8. Calculate the lower limit of the expected use range as "= average (2*stdev)" (rounding down to the nearest whole number)
- 9. Print the numerical spreadsheet data.
- 10. Print the same spreadsheet, but displaying the formulas instead of the data.
- 11. Initial and date both spreadsheets. Enter comments as applicable and submit for review and approval.
- 12. To print the spreadsheet formulas (using MS Excel 2010), perform the following steps:

1. Go to Formulas Tab -> Toggle "Show Formulas" Button on



2. Go to Page Layout Tab, From Page Setup, go to Sheet -> Row & column headings (enable) -> Print



Appendix D Page 2 of 2

	LN ₂ Expected Use Range Calculation Instructions (cont.)										
I	Examp	ole Sprea	adsheet showin					ving Formulas V			
(4)	Book1										
	Α	В	С	D							
1			nnual Verification (_		A	В	С	D		
2	Day#	Date	Amt of LN2 Used		1	SSF LN2 Levels	В	C	U		
3	1	04.01.13	8		2	Day#	Date	Amt of LN2 Used			
4	2	04.02.13	9			1	04.01.13	8			
5	3	04.03.13	7		4	2	04.02.13	9			
6	4	04.04.13	7		5	3	04.03.13	7			
7	5	04.05.13	6		6	4	04.04.13	7			
8	6	04.08.13	9			5	04.05.13	6			
9	7	04.10.13	8			6	04.08.13	9			
10	8	04.11.13	8			7	04.10.13	8			
11	9	04.12.13	9		10		04.11.13	9			
12	10	04.15.13	6		11	10	04.12.13 04.15.13	6			
13	11	04.16.13	4			11	04.15.13	4			
14	12	04.17.13	9			12	04.17.13	9			
15	13	04.18.13	7			13	04.18.13	7			
16	14	04.19.13	6			14	04.19.13	6			
17	15	04.22.13	6			15	04.22.13	6			
18	16	04.23.13	6		18	16	04.23.13	6			
19	17	04.24.13	5		19	17	04.24.13	5			
20	18	04.25.13	9		20	18	04.25.13	9			
21	19	04.25.13	9			19	04.26.13	9			
			7			20	04.29.13	7			
22	20	04.29.13				21	04.30.13	8			
23	21	04.30.13	8			22	05.01.13	8			
24	22	05.01.13				23	05.02.13	7			
25	23	05.02.13	7			24	05.03.13 05.06.13	5			
26	24	05.03.13	6			26	05.08.13	9			
27	25	05.06.13	5			27	05.09.13	5			
28	26	05.08.13	9			28	05.10.13	7			
29	27	05.09.13	5		31			Average	=AVERAGE(C3:C30)		
30	28	05.10.13	7	7.44005	32			Std Dev	=STDEV.P(C3:C30)		
31			Average	7.14286	33			2 Std Dev	=2*D32		
32			Std Dev	1.45686	34			Avg + (2 Std Dev)	=D31+D33		
33			2 Std Dev	2.91373	35			Avg - (2 Std Dev)	=D31-D33		
34			Avg + (2 Std Dev)	10.0566	36			Range	4-11		
35			Avg - (2 Std Dev)	4.22913							
36			Range	4-11							
37											

Appendix E Page 1 of 1

			Date	e of Fill:				
LN ₂ Systems Post-Fill Observation Log								
AFTER EACH FILL	Note: Expected Result = Completed and/or Acceptable, Not Applicable	Initials	Date	Comments/ Corrective Actions				
LN ₂ Level from Gauge	Completed inches Not Completed							
LN ₂ Level from Alarm System, if applicable	☐ Completedinches ☐ Not Completed ☐ Not Applicable							
Calculate LN ₂ Level Difference, if applicable: Note: Expected Result is ≤ 7 Inches	inches □ Acceptable □ Not Acceptable □ Not Completed □ Not Applicable							
Pressure Level Note: Expected Result: 25-40 psig Reviewed By / Date:	□ Completed psig □ Acceptable □ Not Acceptable □ Not Completed							

Appendix F Page 1 of 8

Emergency Contacts,

Wall-Mounted Low O₂ Monitors Alarm Immediate Actions & Response, and Collaborating Biobank Personnel (CBP) Alarm Response and Malfunction Response Directives

Contents:

•	Table of Contents	Page 1
•	Collaborating Biobank Personnel Alarm Response and Malfunction	_
	Response Directives	Page 2
•	Immediate Alarm Response for Personnel Working in LN ₂ Freezer Room	Page 3
•	Alarm Actions & Response for Staff Monitoring Personnel Working in LN ₂	
	Freezer Room	Page 4
•	Immediate Alarm Response for C158A Wall-Mounted O ₂ Monitor	Page 5
•	Alarm Actions & Response following alarm on C158 Wall-Mounted O ₂	
	Monitor	Page 6
•	Response to C158A Wall-Mounted Low O ₂ Monitor - SSF Personnel	
	Follow-Up Actions	Page 7
•	Emergency Contacts	Page 8

Appendix F Page 2 of 8

Collaborating Biorepository Personnel Alarm Response & Malfunction Response Directives:

LN₂ Freezer Room Wall-Mounted O₂ Monitor Audible Alarm and Flashing Lights

Watcher – Safety Observer

Personnel Working in LN₂ Room

- Must observe personnel working in LN₂ Freezer Room from position next to wall-mounted red plunger, labeled "LN2" EMERGENCY STOP" (E-Stop)
- Refer to Page 4 of this Appendix F for Alarm response
- Directives are posted at:
 - o Entrance to LN₂ Freezer Room

- Refer to Page 3 of this Appendix F for Alarm response
- Directives are posted at:
 - Each Wall-Mounted O₂ Monitor
- NOTE: LN₂ Freezer Room entry PROHIBITED unless a trained observer is stationed next to wall-mounted red plunger, labeled "LN2 EMERGENCY STOP" (E-Stop), observing your LN₂ Freezer Room access for safety purposes.

Personnel must NOT RE-ENTER the LN₂ Freezer Room until the alarm has been resolved

Contact SSF Personnel for assistance per Page 8 of this Appendix F.

R3-C158A Lab Wall-Mounted O₂ Monitor Audible Alarm and Flashing Lights

- Refer to Pages 5 & 6 of this Appendix F for Alarm response directives
 - o Page 5 directives are posted at the C158A Wall-Mounted O₂ Monitor
- Directives applicable only after all personnel have vacated C158A are per Page 6 of this Appendix F, and Page 6 is posted at C158A entries. Re-entering the Lab is prohibited until SSF personnel direct that re-entry is permitted.

Discovery of Malfunctioning Wall-Mounted O₂ Monitor

- Evacuate space
- Report malfunction to SSF Personnel
- DO NOT re-enter space
- Further directives will be provided by SSF personnel

Personal O₂ Monitor Alarm Response

Watcher – Safety Observer

Monitor that personnel working in the LN₂ Room follow all safety procedures

Personnel Working in LN₂ Room

- Immediately move away from LN₂ source
- Verify Wall-Mounted O₂ Monitor reads ≥ 19.5%
 - If the O_2 level is \leq **19.5%**, immediately vacate the room
- Verify Personal O₂ Monitor reads ≥ 19.5%
- Press green OK button on Drager Pac 5500 personal O₂ monitor to silence alarm.
 - o For Sensit P100 personal O₂ monitors used at the TK facility, audible alarm automatically silences when O_2 level reads \geq 19.5%.
- If alarm recurs more than two times successively:
 - Vacate the room
- Verify Wall-Mounted O₂ Monitor reads ≥ 19.5%
 - If O_2 level in the room is \geq 19.5% per the wall-mounted O_2 monitors, you may obtain an alternate O₂ monitor & return to the LN₂ room.
- Contact SSF Personnel

Appendix F Page 3 of 8

Flashing Lights & Audible Alarm?

VACATE THE ROM

Use an Emergency Escape Breathing Apparatus (EEBA) Only As Necessary

Inside LN₂ Freezer Room Posting

Appendix F Page 4 of 8

Watcher (Safety Observer) Directives:

LN₂ Freezer Room Low O₂ Alarm Actions & Response

- 1. DO NOT ENTER LN₂ Room
- 2. Press E-STOP
- 3. Ensure ALL PERSONNEL HAVE VACATED Room
- 4. CALL 911 or IUPUI Police at 317-274-7911 if Personnel Rescue Is Needed
- 5. Ensure ALL DOORS ARE CLOSED
- 6. Manually CLOSE VALVE AT BULK TANK

(May be done by any SSF Staff, Campus Facilities Services Personnel, Collaborating Biobank Personnel, IUPUI Campus Police)

- 7. Contact SSF Personnel for Assistance:
 - a. ictsissf@iupui.edu
 - **b.** 317-274-2213 Management Office
 - c. 317-946-6477 Manager Cell Phone
 - d. 317-274-3569 Technician Office
 - e. 317-696-9925 Director Cell Phone
 - f. Page SSF at 317-541-5481
- 8. Report any wall-mounted O₂ monitor malfunction to SSF personnel.

Appendix F Page 5 of 8

Flashing Lights & Audible Alarm?

VACATE THE ROM

Contact SSF Personnel for Assistance

Activate E-Stop ONLY if Immediately Accessible

R3-C158A Posting Appendix F Page 6 of 8

Alarm Actions & Response to C158A Wall-Mounted Low O₂ Monitor Directives applicable only from spaces adjacent to C158A

- 1. DO NOT ENTER C158A
- 2. Ensure ALL PERSONNEL HAVE VACATED ROOM
- 3. CALL 911 or IUPUI Police at 317-274-7911 If Personnel Rescue Is Needed
- 4. Ensure ALL DOORS ARE CLOSED
- 5. Obtain bulk tank key from C158A main entrance (SSF administrative space) or SSF Key lockbox
 - 5.1. DO NOT ENTER C158A to obtain bulk tank key
- 6. Manually CLOSE VALVE AT BULK TANK

(May be done by any SSF Staff, Campus Facilities Services Personnel, Collaborating Biobank Personnel, IUPUI Campus Police)

- 7. Notification:
 - 7.1. Contact SSF Personnel for Assistance:
 - 7.1.1. ictsissf@iupui.edu
 - 7.1.2. 317-274-2213 Management Office
 - 7.1.3. 317-946-6477 Manager Cell Phone
 - 7.1.4. 317-696-9925 Director Cell Phone
 - 7.1.5. 317-274-3569 Technician Office
 - 7.1.6. Page SSF at 317-541-5481
 - **7.2.** SSF Personnel Notify Emergency Contacts per Page 8 of Appendix F
- 8. Report any wall-mounted O₂ monitor malfunction to SSF personnel

Posted at R3-C158A entrances & at C158A O₂ Monitor

Appendix F Page 7 of 8

SSF Personnel Response to C158A Wall-Mounted Low O₂ Monitor Alarm— SSF Personnel Follow-Up Actions

- 1. SSF personnel may return to C158A and investigate Wall-Mounted Low O₂ Monitor alarm and equipment malfunction **ONLY IF** both conditions 1.1 and 1.2 can be confirmed *FROM SPACES ADJACENT TO C158A* and permission is obtained per condition 1.3:
 - 1.1. If no nitrogen vapor or nitrogen liquid are visible in C158A
 - 1.2. If personal O₂ monitors beneath the wall-mounted O₂ monitor are NOT alarming
 - 1.3. With SSF Management permission
- 2. Investigation of malfunction and facility re-entry is per the Wall-Mounted O₂ Monitor Troubleshooting section of this SOP.
- 3. If (a) nitrogen vapor or nitrogen liquid ARE visible in C158A and/or (b) personal O₂ monitors beneath the wall-mounted O₂ monitor ARE alarming:
 - 3.1. **C158A RE-ENTRY PROHIBITED** until LN₂ system is evaluated by safety personnel (EHS, Praxair/Linde, other qualified vendor) and all safety concerns are addressed.
 - 3.2. Manually CLOSE VALVE AT BULK TANK
 (May be done by any SSF Staff, Campus Facilities Services Personnel, Collaborating Biobank Personnel, IUPUI Campus Police)
 - 3.3. Notify Emergency Contacts per Appendix F, page 8

Appendix F Page 8 of 8

EMERGENCY CONTACTS

SSF Operations Manager: Name

Pager:

Office:

Phone:

SSF Facilities Manager: Name

Pager:

Office:

Phone:

SSF Director Name

Phone:

Campus Facilities: 317-278-1900

Environmental Health and

Safety: 317-274-2005

IUPUI Police: 4-7911 (from an IU phone)

317-274-7911

Police: 911

Date Effective:
Obsolete Date:

Appendix G Page 1 of 1

			Month:	Y	ear:						
LN₂ WEEKLY SYSTEM CHECK											
	System Observation										
v	VEEK (Observable Frost Rings Present? (Yes / No)	Tech Initials	s / Date							
	1	☐ Yes ☐ No									
	2	☐ Yes ☐ No									
	3	☐ Yes ☐ No									
	4	☐ Yes ☐ No									
	5	☐ Yes ☐ No ☐ NA									
		Coi	mments								
		Mixed Air Cylinder G	auge Readings for	r E-Stop							
WEEK	Delivery Pressure (kPa) – Acceptable Range 500-680 kPa	Acceptable	Supply Pressure (psi) – Acceptable Range: 300-2200 psi	Acceptable (Yes / No)	Tech Initials / Date						
1		☐ Yes ☐ No	•	☐ Yes ☐ No							
2		☐ Yes ☐ No		☐ Yes ☐ No							
3		☐ Yes ☐ No		☐ Yes ☐ No							
4		☐ Yes ☐ No		☐ Yes ☐ No							
5		☐ Yes ☐ No ☐ NA		☐ Yes ☐ No ☐ NA							
Comments											
_											
Review	ved By / Date:										

Appendix H Page 1 of 2

Collaborating Biorepository Personnel (CBP) Training

The SSF has a hard-plumbed, vacuum-insulated, liquid nitrogen (LN₂) system that connects a 6000-gallon bulk tank to freezers, providing liquid & vapor phase nitrogen storage to users. Routine maintenance and monitoring of the SSF LN₂ system is completed by SSF personnel. This SOP defines safety procedures and entry directives for access to R3-C156, the SSF LN₂ room.

LN₂ Room safety directives are critical, because in an atmosphere filled with nitrogen gas, oxygen is easily displaced and can result is asphyxiation. Therefore, there are several safety procedures in place to assure adequate oxygen supply when working in the LN₂ room.

Responsibility	Watcher – Safety Observer	Personnel Entering the LN ₂ Room		
Room Entry	 Must be positioned next to wall-mounted red plunger, labeled "LN₂ EMERGENCY STOP" (E-Stop) Verify personnel entering LN₂ room is 	 DO NOT ENTER unless trained observer is stationed next to wall-mounted red plunger, labeled "LN2 EMERGENCY STOP" (E-Stop), observing your LN2 room access for safety purposes. Don a personal O₂ monitor prior to entering the LN₂ room 		
	wearing a personal O ₂ monitor. 3. Verify O ₂ level is above 19.5% per the	per directives in "Don a Personal O ₂ Monitor" section below. 3. Verify O ₂ level is above 19.5% per the wall-mounted O ₂		
	wall-mounted O ₂ monitor. 4. Verify LN ₂ bulk tank is not being filled	monitor 4. Verify the LN₂ bulk tank is not being filled		
	 Verify Negative pressure differential by observing that the Ball in Wall indicator is not visible inside C158A 	 5. Verify Negative pressure differential by observing that the Ball in Wall indicator is not visible inside C158A 		
	If negative pressure differential indicator, ball-in-wall, is visible in C158A and repair signage is not already posted, notify SSF personnel.			
Entry into C156 IS NOT prohibited.				
Don a Personal O₂ Monitor	 Verify personnel entering LN₂ room is wearing a personal O₂ monitor. 	 Obtain a personal O₂ monitor for each personnel entering the room according to the following priorities and exceptions: Personal O₂ monitor must display O₂ level above 19.5% AND must not display low battery symbol or [!] (indicating an error), per SF-3-15 SOP for Drager Pac 5500 Personal O₂ Monitors		
Personal O ₂ Monitor DOs and DON'Ts	 Personal O₂ monitors alarm because personnel are surrounded by LN₂ vapor. DO wear the monitor near your head. 			
	 DON'T hang the monitor from your neck by the attached lanyard. The monitor will descend into an LN₂ freezer and alarm. DON'T place the monitor in your pocket, where its air intake will be obstructed, and the monitor will 			
	alarm.	, meneral an internet minute observation, and the monitor will		

Appendix H Page 2 of 2

Responsibility		Watcher – Safety Observer		Personnel Entering the LN ₂ Room	
Sample Access	1.	Keep a constant eye on personnel working in the \ensuremath{LN}_2 room	1.	Personnel must be wearing approved and proper PPE per universal precautions	
	2.	Wall mounted mirrors facilitate tracking of personnel out of direct line of sight	2.	Don cryogenic gloves when working with liquid nitrogen in the vapor phase	
			3.	 When working with liquid nitrogen in the liquid phase, don: a. chemical splash goggles or full face shield i. Since splashing can occur, a full face shield is preferred for filling dewars b. cryogenic gloves 	
				c. cryogenic apron	
LN ₂ Dispensing and	1.	Monitor that personnel in C156 adhere to safety procedures defined at right	1.	LN_2 may only be dispensed by personnel trained to fill dewars by trained-CBP or SSF staff	
Dewar Fills			2.	 Before dispensing LN₂, personnel must don: a. chemical splash goggles or full face shield (Full face shield recommended) b. cryogenic gloves c. cryogenic apron 	
			3.	Report all dewar fills in iLab for billing purposes	
Wall-Mounted O₂ Monitors Measure and					

Wall-Mounted O₂ Monitors Measure and Report O₂ Levels in the Vicinity of the Wall-Mounted O₂ Monitor

Refer to Appendix F for directives for responding to:

- Wall-Mounted O₂ monitor Alarms
- Discovery of Wall-Mounted O₂ monitor Malfunction is per Appendix F

Personal O₂ Monitors Measure and Report O₂ Levels Around the User

Refer to Appendix F for response to Personal O₂ Monitor Alarm

LN₂ Freezer Room Access when a Wall-Mounted O₂ Monitor is Malfunctioning:

LN₂ Freezer Room access will be limited per SSF Directives – SSF personnel will contact CBP

Additional Appendices Requiring Review:

Appendix A: Components of the Liquid Nitrogen System defines critical components of the LN₂ room, also covered during onsite technical training with SSF personnel.

Appendix F: Emergency Contacts and Wall-Mounted Low O2 Monitors Alarm Actions Template

Appendix I Page 1 of 2

SSF Wall-Mounted Low O₂ Monitors Alarm Monitoring / Notification & Alarm Malfunction Response

SSF Directives - Low O₂ Alarm Resolution during Normal Function of <u>Both</u> Wall-Mounted O₂ Monitors in the LN₂ Freezer Room

Directives applicable only following appropriate alarm response per Appendix F

1. SSF Monitoring Following Alarm

- 1.1. Alarm Lights Flashing & Intermittent Audible
 - 1.1.1. Re-Entry Prohibited Until O₂ Level Rises To ≥ 19.5%

1.2. Alarm Lights Flashing & Continuous Audible

- 1.2.1. Re-Entry Prohibited Until O_2 Level Rises $To \ge 19.5\%$
- 1.2.2. Manually Close Valve at LN₂ Bulk Tank If Not Already Done
- 1.2.3. Notify Emergency Contacts Per Appendix F If Reading Decreases Below 18.5% (Appendix F, Page 8)

1.3. White-Out Conditions

- 1.3.1. Defined as vapor density that blocks view of room or sections of the room.
- 1.3.2. Re-Entry Prohibited until LN₂ system is evaluated by safety personnel (EHS, Praxair/Linde, other qualified vendor) and all safety concerns are addressed
- 1.3.3. Following White-Out Conditions, Notify Emergency Contacts per Appendix F, page 8

Appendix I Page 2 of 2

SSF Directives - Low O₂ Alarm Resolution With One or Both LN₂ Freezer Room Wall-Mounted O₂ Monitor(s) Malfunctioning

Directives applicable only following appropriate alarm response per Appendix F

Directives Applicable to:

- Alarm Lights Flashing & Audible Alarm
- White-Out Conditions
- Alarm From Personal O2 Monitor Stationed At Malfunctioning Wall-Mounted O2 Monitor

Monitoring Following Alarm on:

1. MALFUNCTIONING Wall-Mounted O₂ Monitor

- 1.1. Alarm Lights Flashing & Audible Alarm: Re-Entry Prohibited Until O₂ Level Rises To ≥ 19.5%
- 1.2. White-Out Conditions
 - 1.2.1. Defined as vapor density that blocks view of room or sections of the room.
 - 1.2.2. Re-Entry Prohibited until LN₂ system is evaluated by safety personnel (EHS, Praxair/Linde, other qualified vendor) and all safety concerns are addressed
 - 1.2.3. Following White-Out Conditions, Notify Emergency Contacts per Appendix F, page 8

2. Personal O₂ Monitor Stationed At Malfunctioning Wall-Mounted O₂ Monitor

- 2.1. Alarm Lights Flashing & Audible Alarm: Re-Entry Prohibited Until O₂ Level Rises To ≥ 19.5%
- 2.2. White-Out Conditions
 - 2.2.1. Re-Entry Prohibited until LN₂ system is evaluated by safety personnel (EHS, Praxair/Linde, other qualified vendor) and all safety concerns are addressed
 - 2.2.2. Following White-Out Conditions, Notify Emergency Contacts per Appendix F, page 8