



भारत सरकार के प्रधान वैज्ञानिक सलाहकार का कार्यालय
Office of the Principal Scientific Adviser
to the Government of India

MINIMUM STANDARD REQUIREMENTS

NATIONAL NETWORK OF BSL 3/4 LABORATORIES



Guidelines For High Containment Facilities Under National One Health Mission (NOHM)

2026

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FOREWORD

India's preparedness for emerging and re-emerging infectious disease threats requires not only adequate high-containment laboratory capacity, but also consistency in the standards, practices and operational readiness of laboratories that constitute the national network. The experience of recent public health emergencies has reinforced the importance of strong laboratory systems that can support timely detection, diagnosis, research and coordinated outbreak response across sectors. The National One Health Mission (NOHM) provides an important framework for strengthening such inter-sectoral preparedness by bringing together efforts across human, animal, plant and environmental health. Within this framework, the National Network of High-Risk Pathogen Laboratories is intended to strengthen connectivity, collaboration and coordinated response among high-containment laboratories operating across the country.

The Minimum Standard Requirements (MSR) for the National Network of BSL-3 Laboratories have been developed to provide a common operational framework for laboratories that are part of this network. The MSR brings together key requirements relating to containment integrity, engineering and essential equipment, biosafety and biosecurity, operational practices, personnel competency, validation, emergency preparedness and data management. By establishing common and verifiable expectations, the framework seeks to promote consistency and facilitate safe and effective collaboration across participating laboratories.

The MSR builds upon the mandatory compliance requirements established under the National Guidelines for the Establishment and Certification of BSL-3 Containment Facilities (2024) and the harmonized DBT–ICMR certification framework. It is important to emphasise that these existing regulatory and certification requirements remain applicable and are not superseded by the present document. Rather, the MSR provides an additional operational framework to support the integration of certified laboratories into a coordinated national network and to strengthen their mission-readiness. The development of these requirements has been a collective effort involving the members of the Advisory and Review Committee, the Sub-Working Group, and experts and institutions from relevant sectors. The deliberations have sought to balance the requirements of biosafety, operational practicality and national preparedness, while recognising the diversity of institutions and facilities that constitute the laboratory network.

I am confident that the implementation of these Minimum Standard Requirements will contribute to greater consistency in high-containment laboratory practices and strengthen the ability of laboratories across the network to work together safely and effectively during public health emergencies. I acknowledge with appreciation the contributions of all experts, institutions and colleagues who have supported the development of this document.

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EXECUTIVE SUMMARY

The National One Health Mission (NOHM) is a flagship Government of India initiative that seeks to operationalize the One Health approach through coordinated action across the human, animal, plant, and environmental health sectors. The Mission aims to strengthen national preparedness against endemic, emerging, and re-emerging diseases through integrated surveillance, early warning systems, and a collaborative framework for disease prevention, detection, and response.

As a key component of this effort, NOHM has established a National Network of High-Risk Pathogen Laboratories comprising Biosafety Level-3 (BSL-3) and Biosafety Level-4 (BSL-4) facilities across the country. The network is intended to enhance national capacity for diagnostics, research, surveillance, and emergency response while fostering collaboration among institutions operating in different sectors.

To support the effective functioning of this network, the present Minimum Standards Requirements (MSR) document has been developed as a harmonized framework outlining the essential requirements for laboratory infrastructure, containment systems, biosafety and biosecurity practices, operational governance, and quality assurance. The MSR seeks to promote uniformity, interoperability, and mission readiness across participating laboratories, thereby enabling coordinated action during public health emergencies.

The document recognizes that compliance with the National Guidelines for the Establishment and Certification of BSL-3 Containment Facilities (2024) and certification under the harmonized DBT–ICMR framework remains a mandatory prerequisite for laboratories handling Risk Group-3 pathogens. The MSR does not replace existing regulatory, certification, or accreditation mechanisms; rather, it builds upon them by providing an operational framework for integration into the National Network of Bio-Safety Level (BSL) Laboratories under NOHM.

By establishing common standards and operational expectations, the MSR aims to facilitate safe resource sharing, strengthen inter-institutional collaboration, and enhance the collective preparedness of the national laboratory ecosystem. Ultimately, it serves as a strategic instrument for aligning institutional capabilities with the broader objectives of the National One Health Mission, thereby contributing to a resilient, coordinated, and future-ready national response system for infectious disease threats.

The document has been developed through a collaborative drafting process involving the Sub-Working Group and the National One Health Mission team. The zero draft and subsequent revisions were drafted by Dr Sangeeta Agarwal, Dr Darshan Narayan, and Ms. Arundhati Mohanty, with inputs incorporated from all members.

1. INTRODUCTION

Biosafety Level 3 (BSL-3) laboratories are established to enable the secure handling of pathogens capable of causing serious or potentially fatal diseases. To ensure containment, protect laboratory personnel, and prevent environmental dissemination of these infectious agents, stringent requirements are being fulfilled in the design, operation, and validation of such facilities. The following outlines the essential requirements that are a part of the framework adopted by all the laboratories to ensure compliance with the BSL-3 standards and specifications.

To enhance national preparedness for emerging and re-emerging infectious diseases, under the “Program for Strengthening research and development (R and D) towards Integrated Disease Control and Pandemic Preparedness for the National One Health Mission (NOHM)” a major effort includes the establishment of a National Network of High-Risk Pathogen Laboratories (BSL-3/4) across various departments and research institutions to facilitate interconnectivity, resource sharing, and coordinated response during outbreaks. Only the BSL-3 containment facilities, certified in accordance with the National Guidelines for the Establishment and Certification of BSL-3 Containment Facility, 2024; may be considered for inclusion in the National Network of High-Risk Pathogen Laboratories (BSL-3/4).

As part of this effort, the development of Minimum Standard Requirements for BSL-3 Laboratories has been undertaken to provide a unified framework for laboratory design, containment, and operational practices, ensuring biosafety compliance and functional consistency across all participating facilities. A robust framework of minimum standards is not only essential for both routine functionality and regulatory compliance but also will enable networking of these facilities under a uniform framework of compliance to pandemic preparedness.

The primary consideration for the minimum standard requirements is to ensure and ascertain that all the laboratories in the network operate on a consistent platform with respect to their facility compliance to the critical parameters, thereby enabling safe and effective laboratory operations while protecting personnel, the community and the environment. In this connection, in compliance with the existing National Guidelines for the Establishment and Certification of BSL-3 Containment Facility, 2024, the following are the checklists of critical parameters that need to be appropriately addressed and validated to ascertain their adherence to the current guidelines:

- All BSL-3 laboratories are centered on structural design, containment of integrity, and operational controls with the access that are strictly regulated using self-closing, lockable doors and the incorporation of an anteroom or airlock for ingress and egress.
- Critical engineering controls include a dedicated Heating, Ventilation, and Air Conditioning (HVAC) system designed to maintain directional airflow from clean to potentially contaminated zones. The negative air pressure differential relative to adjacent



areas are also to be maintained for effective containment and prevent the accidental release of these pathogens.

- The airflow patterns are validated to confirm uniform flow toward exhaust points, and high-efficiency particulate air (HEPA) filters installed at both supply and exhaust terminals, and its integrity routinely verified and documented.
- The operational parameters such as air changes per hour, airflow velocity, and differential room pressures are continuously monitored and recorded to ensure containment efficacy.
- The laboratory environmental monitoring, including particle counts, is conducted to verify that a clean and controlled environment is sustained.
- All the critical equipment in such a facility are validated and maintained in accordance with established protocols. In addition to engineering measures, administrative and procedural controls are also made imperative.
- The personnel are trained in BSL-3 operational practices, including the correct use and disposal of personal protective equipment (PPE), decontamination protocols, and emergency response procedures.
- The standard operating procedures (SOPs) pertaining to each facility are comprehensively documented, encompassing sample handling, waste disposal, and incident management with respect to each facility in line with the pathogens that are being handled.

2. BACKGROUND

The development of the Minimum Standard Requirements (MSR) represents a strategic tool to harmonize high-containment operations. Driven by the NOHM, this initiative transitions the nation from a model of isolated departmental excellence to a unified, cross-sectoral defense system. While BSL-3 and BSL-4 facilities currently exist across various sectors, the NOHM identified a critical need for these labs to function as an interoperable, cohesive unit to ensure pandemic preparedness.

The foundation of this framework is the mandatory national certification for any facility handling Risk Group 3 (RG-3) agents. The DBT-ICMR harmonized certification remains the definitive benchmark for functional worthiness. Compliance with the 2024 National Guidelines¹ is a non-negotiable prerequisite, ensuring that structural design and annual validations meet rigorous safety benchmarks. For central government entities, certification is undertaken by the Department of Biotechnology (DBT) or respective line ministries.

While DBT or other certifications as per the National Guidelines confirm that a laboratory is safe for operation, the MSR introduces a strategic layer of Mission-Readiness. The MSR does not supersede existing authorities but acts as a bridge to ensure that facilities are technically and operationally compatible for national integration. This alignment enables safe resource sharing and a coordinated response that transcends traditional sectoral boundaries.

¹[National Guidelines for the Establishment and Certification of Biosafety Level3 \(BSL-3\) Containment Facility, 2024 \(https://ibkp.dbt.gov.in/Content/Rules\)](https://ibkp.dbt.gov.in/Content/Rules)

The National network of BSL 3/4 laboratories is operationalized through state/Union Territory (UT) departments, district administration and bodies such as the National Joint Outbreak Response Team (NJORT). By standardizing practices through the MSR, the network ensures that samples collected from infection hotspots can be seamlessly referred to the nearest certified BSL-3 laboratory for rapid, high-fidelity analysis. Ultimately, the MSR serves as a guidance document for laboratories to be included in the national network transforming individual high-containment laboratories into a synchronized national asset, harmonizing operations to safeguard public health during emergencies.

3. STRUCTURAL ENGINEERING AND EQUIPMENT REQUIREMENTS

All the validation data pertaining to the above critical checklists shall ensure traceability and compliance with ISO/IEC 17025:2017 standard. In addition, an annual re-validation is required to confirm that modifications in facility usage, maintenance activities, or environmental conditions do not compromise biosafety. The detailed descriptions on each of the critical parameters, its importance, the validation method and the acceptance criteria are listed below:

3.1. Structural/Environmental Parameters:

3.1.1. Temperature and Relative Humidity

The temperature and relative humidity are critical determinants of environmental stability and personnel comfort that are required for safe and compliant operation of a BSL-3 laboratory. Its effective control and systematic validation are essential and vital to maintain the operational integrity of BSL-3 laboratories, which include:

- **Containment integrity:** Needed for consistent air density, which directly influences the pressure differentials and directional airflow that are essential for primary and secondary containment (*The deviations can alter air balancing and compromise the negative pressure regime*)
- **Stability of biological agents and equipment:** Controlled environmental conditions ensure consistent equipment performance and biological process reproducibility
- **Personnel safety and comfort:** Maintaining a comfortable environment condition of the laboratory minimizes fatigue, ensuring compliance with biosafety protocols and enhancing the operational efficiency of the laboratory personnel
- **Validation to meet the compliance:**
 - ✓ Continuous monitoring is essential using the calibrated temperature and RH sensors placed at all representative locations and a part of annual routine validation
 - ✓ Validation is performed with a calibrated digital hygrometer / Temperature-RH data logger (ascertain whether this standard equipment used is within the calibration validity)



- ✓ *Acceptance criteria: (A temperature of $22\text{ }^{\circ}\text{C} \pm 2^{\circ}\text{C}$) Less than 60% RH (commonly targeted between 30% and 50% or max 60%) (Reference: DBT_ICMR Collaborative national guidance document).*

3.1.2. Light Intensity Level

The lighting system is designed to avoid heat generation or UV interference that could affect temperature regulation or integrity of light-sensitive reagents. Adequate and uniform illumination is essential to support

- Optimal visibility for laboratory personnel performing laboratory work
- Thorough cleaning, decontamination, and inspection of laboratory surfaces
- **Validation to meet the compliance:**
 - ✓ *There is no necessity for continuous monitoring of this criterion and to be a part of annual routine validation*
 - ✓ *Measurement of light intensity is performed using a calibrated lux meter at all the major work areas (e.g., biosafety cabinet work surface, bench top, and general laboratory space) as lux units (lumens per square meter)*
 - ✓ *Acceptance criteria: Lighting should be in the range of 300 - 500 lux in the laboratory and 700-800 lux in cleaning cycle area (Reference: DBT_ICMR Collaborative national guidance document).*

3.1.3. Noise Level Test

It is a critical environmental parameter and in a facility with full operations, background noise is expected from biosafety cabinets, HVAC blowers, exhaust fans and other laboratory equipment and meticulous attention is planned during the facility design to ascertain:

- A comfortable working environment (prolonged exposure to noise levels >70dB results in fatigue and hearing discomfort)
- For distinguishing the audible alarms that are set as critical warnings (e.g., differential pressure, door interlocks/airflow, etc.)
- **Validation to meet the compliance:**
 - ✓ *There is no necessity for continuous monitoring of this criterion and to be a part of annual routine validation*
 - ✓ *Measurement of noise level is performed at operator ear height i.e., typically 1.2 to 1.5 m from the floor under normal operation conditions (with the common equipment's functioning during the validation)*
 - ✓ *Measured using a calibrated sound level meter*
 - ✓ *The ambient level is $\leq 60\text{dB}$; the alarm audibility to be $\geq 10\text{dB}$ above the ambient noise level (Reference: DBT_ICMR Collaborative national guidance document).*



3.2. Containment Integrity:

3.2.1. Containment Leak Test

Since the laboratories operate under constant negative pressure to provide airtight containment, there might be a compromise in the seals, gaskets, ductwork and the filter systems over time resulting in compromised primary/secondary containment barriers.

- **Validation to meet the compliance:**

- ✓ *There is no necessity for continuous monitoring of this criterion and to be a part of annual routine validation*
- ✓ *The test is usually done by smoke visualization or tracer gas detection (e.g. Sulphur Hexafluoride (SF₆) or soap bubble test or pressure decay testing (not a common approach)*
- ✓ *Acceptance criteria:*
(I) The testing is performed at positive pressure of 1000 Pa and acceptance criteria is pressure drop less than 0.1% duct volume/min.
For containment space, the testing is performed at 125 Pa followed by 250 Pa, for 20 min. and reading is of pressure drop taken every minute. Acceptance criteria is pressure drop < 125 Pa in 20 min. (Reference: DBT_ICMR Collaborative national guidance document).

3.2.2. Air pressure and differential pressure test: This is one of the most critical elements to be constantly monitored and validated at appropriate intervals.

- The air pressure and pressure differential across the adjacent laboratory zones is critical for unidirectional air flow from lower to higher containment
- Requires support and monitoring by the engineering team and alarms are to be set for this parameter
- Typical requirement of the calibrated pressure sensors (preferably digital sensors) in all the different laboratory zones connected to the BMS
- The balancing is also done during the initial facility commissioning to ascertain that the pressure differential is maintained within the set limit – facility operation with or without all the critical equipment that require appropriate air circulation (e.g., the BSCs; all the BSCs in operation will require appropriate pressure balancing so that the differential pressure in the laboratory zone is always maintained within the set limits)
- Validation is done with a manometer or differential pressure meter
- *Acceptance criteria: (I) The laboratory needs to maintain a pressure cascade of at least -15 Pa between successive zones; (ii) Need to be correlated with the air flow visualization, air handling unit (AHU) performance, air velocity and air changes per hour (ACPH); (iii) Assessment critically requires the two values (a) baseline assessment (when no laboratory equipment is operational) and (b) dynamic condition assessment (when all equipment within the*



containment zone are in operation). (Reference: DBT_ICMR Collaborative national guidance document).

3.3. Air Handling and Filtration:

3.3.1. Air handling unit (AHU) performance: The performance of the air handling units validates the heating ventilation air conditioning (HVAC) system efficiency in maintaining controlled air quality, temperature, humidity and pressure in the laboratory.

- This is one of the critical parameters that needs continuous monitoring and is dependent on the other listed parameters, which are as follows;

Validation to meet the compliance:

- The validation includes checking the fan operation, damper positions, filter condition, particulate count and air flow rates (baseline and dynamic conditions)
- *Acceptance criteria: (i) Validation is usually performed with airflow meters and manometers; laboratory needs to maintain a pressure cascade of at least 15 Pa between successive zones; (ii) Need to be correlated with the air flow visualization, air velocity and ACPH, HEPA filter integrity and air particulate count; (iii) Acceptance criteria is indirect and the major parameter that are included as efficiency indicators are the air flow and negative pressure levels (the value to be within $\pm 10\%$ tolerance). (Reference: DBT_ICMR Collaborative national guidance document).*

3.3.2. Air flow pattern visualization: This is a qualitative assessment for confirming the movement of air within the laboratory (particularly confirming that the contaminated air is contained and does not flow into clean areas) and provides observable evidence on integrity of the containment system.

- Assessment ensures that the supply and exhaust systems are appropriate.
- Enables the identification of dead zones.

Validation to meet the compliance:

- Visualization at regular intervals is suggested to ensure the reliability of the containment system and supports safe operation
- *Acceptance criteria: (I) Smoke test/ fog generators/ tracer gas are routinely used; (ii) Uni-directional smoke pattern should be observed, from low contamination to high contamination area (iii) an indirect indicator of the HVAC and other containment parameter performance. (Reference: DBT_ICMR Collaborative national guidance document).*

3.3.3. Air velocity, Air changes per hour (ACPH) and Uniformity test: The air velocity, ACPH and its uniformity are interrelated features and measurements are conducted with calibrated anemometer



- *Air velocity* indicates the speed at which air passes through the ducts, inlets and workspace. The air velocity within supply and exhaust ducts ascertains proper ventilation and removal of contaminants.
- *Air changes per hour and uniformity tests* – It indicates the number of times the total air volume in the laboratory is replaced per hour. To assess the ACPH uniformity, measure the air velocity at multiple points (i.e., different laboratory locations). For its validation, the airflow measurement, room volume and the air velocity at multiple points are required

Validation to meet the compliance:

- In this connection, balancing the total air supply (by the AHU) and the total exhaust (air removed through exhaust ducts, biosafety cabinets etc.) ensures the maintenance of the negative pressure in the laboratory (Total exhaust offset)
- *Acceptance criteria: (I) Air velocity - requirement of 0.45 to 0.55 m/s with $\pm 10\%$ uniformity across measurement points; (ii) The ACPH is calculated by the formula $[Total\ supply / Room\ volume] \times 60$. An ACPH of 8 to 12 is recommended for BSL-3 laboratory and 10 to 12 for ABSL-3 facility (Reference: DBT_ICMR Collaborative national guidance document).*

3.3.4. HEPA filter integrity/Filter test: The efficiency and integrity of supply and exhaust air HEPA filters are a critical part of the validation.

- The performance needs to be validated for the facility as well as the BSCs and ventilation system that connects the individually ventilated cages (IVCs) (in ABSL-3 facility)
- Their performance directly determines the primary containment and secondary containment.

Validation to meet the compliance:

- Validation involves filtration efficiency, integrity and leak scanning using a known concentration of aerosolized agent upstream of the HEPA filter (Polyalphaolefin (PAO), Dioctyl Phthalate (DOP) testing) and evaluated using a calibrated photometer or aerosol detector
- The validation includes (i) *Efficiency of filtration*; (ii) *Leak scanning at the periphery of the filter, gasket and frame joints*
- The validation should be typically annually or after maintenance activities
- *Acceptance criteria: No detection exceeding 0.01% of upstream concentration of the HEPA filter is permissible (Reference: DBT_ICMR Collaborative national guidance document).*

3.3.5. Air particulate count: The measurement of airborne particles confirms the cleanliness of the environment

- Validated using calibrated laser particle counter at different locations and determination at appropriate intervals will confirm integrity and efficiency of the filtration system
- Calibrated particles sizes of ($\geq 0.3\mu\text{m}$, $\geq 0.5\mu\text{m}$, $\geq 1.0\mu\text{m}$ and $\geq 5.0\mu\text{m}$)
- The measurement – at base line and operational conditions
- Measurement also indirectly ensures the HEPA filter integrity
- The particulate count in terms of the filtration efficiency ascertains and confirming them to the ISO cleanroom standards (preferably ISO14644 Class 8)
- Acceptance criteria: The typical cleanroom classifications for a BSL-3 lab, along with the maximum allowed particles per m³ for different sizes, are shown in the following table (Reference: DBT_ICMR Collaborative national guidance document).*

ISO Class	Particle size and its respective count			
	At Rest		In Operation	
	$\geq 0.5\mu\text{m}$	$\geq 5.0\mu\text{m}$	$\geq 0.5\mu\text{m}$	$\geq 5.0\mu\text{m}$
ISO Class 7	3,52,000	2,930	3,52,000	2,900
ISO Class 8	35,20,000	29,300	35,20,000	29,000

3.4. Critical Equipment and Systems:

3.4.1. Calibration of critical Equipment: In addition to the validation of the facility requirements, the other equipment used in the facility (*biosafety and decontamination equipment, analytical and support equipment*) are critical to ensure safety of the personnel as well as of the environment. They need to be evaluated annually or after any maintenance activity and this includes the following;

For BSL-3 laboratory:

a. Biosafety and decontamination equipment

- Biological safety cabinets (BSCs) Class II:* They are the primary containment barriers, and the validation includes HEPA filter integrity, inflow/outflow air velocity, smoke test to assess the air flow, alarm function and efficiency of the sash height
- Autoclave:* An *in-situ*, double-door barrier autoclave for the decontamination of contaminated waste material should be available in the containment laboratory. Calibration of pressure gauge and temperature by thermal mapping should be ensured. If an automated timer is present, it also shall be calibrated. For testing of effective sterilization, chemical indicators can be used. However, Biological indicators are preferred.
- Effluent decontamination system (EDS):* Thermal profile in case of thermal decontamination system, leaks if any, microbial kill efficiency ($\geq 6\text{-log}$)



reduction; to be evaluated annually or as per SOP; in case of chemical decontamination the efficiency to be assessed as per SOP)

- *Fumigation systems:* Fumigator H₂O₂ or any other acceptable method used in the facility for gaseous decontamination of the equipment or the whole facility. *This should include concentration validation, exposure time and uniformity of distribution*
- *Incinerator/Waste disposal unit:* This is not a mandatory requirement and if available the validation should include temperature and emission control (flue/exhaust gas emissions as per CPCB norms).

b. Analytical and support equipment:

The validation documents of refrigerators/freezers are mandatory to be included as a part of the facility compliance. These and others are basic requirements of the laboratory which are as follows and contribute to the SOP

- *Refrigerators/Freezers:* should include temperature mapping and alarm test (at least quarterly)
- *CO₂ incubators:* should include the temperature and CO₂ concentration calibration (at least quarterly)
- *Centrifuges (with sealed rotors):* should include rotor seal integrity and speed calibration, if possible (annually)
- *Micropipettes:* should include volume accuracy and precision (as per requirement).

c. For an Animal facility (ABSL-3 specific): In addition to the biosafety and decontamination equipment, the following equipment used in the animal experiments needs appropriate validation:

- *Individually ventilated cage:* this should include the HEPA filter integrity, airflow rate and pressure differential (at least annually).
- *Rack blower unit/exhaust system:* this should include the flow and pressure validation.
- *Cage change stations/workstations* (usually like similar to BSCs and as indicated above).
- *Effluent and bedding decontamination units* (Biological Liquid Effluent Decontamination (BLED), the autoclaves as indicated above).

3.4.2. Water supply and drainage system validation:

The evaluation should include the following;

- The use of backflow prevention devices in supply and drains.
- In-facility distribution of piping (joints, isolation valves, etc.).
- The use of fixtures (sinks, eyewashes/ safety showers, equipment water inlets).
- Hot water systems in case of facility with exit showers.
- Drainage network – floor drains, traps (U-traps), drain piping, inspection ports.



3.5. Operational and Safety Systems:

3.5.1. Test report on alarm, safety and utility systems:

The validation is critical to the operational safety to alert the staff on any system failure that could compromise the biosafety. This includes the following;

- *Alarm and interlock systems (doors, AHU, pressure alarms):* this ensures biosafety interlocks and emergency control, and this should include functional testing of door interlocks, audible and visual alarms relating to the same (at least quarterly).
- *Uninterrupted Power Supply (UPS)/Emergency power supply system:* There should be a standby UPS of 100% capacity to support critical equipment. This ensures maintenance of the containment during power failure, and this should include load test, backup duration and automatic switch over (at least quarterly).
- *Fire detection and suppression systems:* this ensures protection against any unexpected fire hazard, and this should include sensor calibration, alarm testing, extinguishing agent pressure check (as per safety norms).
- *Gas supply and vacuum systems:* this ensures safe handling of compressed gases, and this should include leak tests, regulator calibration.
- *Alarm systems:* shall remain clearly audible at minimum of 10 dB above ambient background noise.

3.5.2. Waste management:

- The SOP pertaining to the following aspects;
 - Systematic handling, segregation, treatment and disposal of all materials that are supposed to be contaminated.
 - Use of color containers for disposal of the materials during operation to support appropriate segregation.
 - Internal transport of the waste before decontamination – this includes the double bagging, sealing, leak-proof trolleys and directional movement of the waste (clean to dirty zones).
 - Temporary storage of collected waste after decontamination if any.

3.5.3. High-Containment (RG3) Pathogen storage and management

3.5.3.1. Physical Isolation and Access Control

All Risk Group 3 (RG3) organisms shall be securely stored in dedicated, physically segregated units or rooms. These areas shall feature standardized biohazard signage and be protected by a secure perimeter.

- ***Validation to meet the compliance:***
- *Physical site inspection of storage segregation.*
- *Review of the Personnel Authorization Matrix.*

Acceptance Criteria:



- ✓ *All storage units shall bear visible, compliant hazard labels.*
- ✓ *Digital or physical access logs shall match the list of authorized personnel only. All staff with access shall possess valid, up-to-date biosafety training certifications. (Reference: DBT_ICMR Collaborative national guidance document).*

3.5.3.2. Inventory Management and Traceability

A centralized Master Inventory shall be maintained for all high-risk pathogens, ensuring a real-time chain of custody for every sub-sampling and transport event, as well as disposal.

- ***Validation to meet the compliance:***
- *Cross-reference of the Master Inventory against physical vial counts.*
- *Audit of inbound/outbound usage logs and timestamps.*

Acceptance Criteria:

- ✓ *Zero discrepancy between digital/paper records and physical stock. Completion of periodic integrity audits (weekly/monthly) with documented inspection logs. (Reference: DBT_ICMR Collaborative national guidance document).*

3.5.3.3. Biosecurity and Containment

All the materials used in the facility shall be disposed only after validated procedure of decontamination. The facility shall have pest control program.

- ***Validation to meet the compliance:***
- *Visual inspection of secondary containment systems.*
- *Review of Autoclave/Sterilization logs and biological indicator results.*
- *Verification of Integrated Pest Management (IPM) service records and bait station mapping.*

Acceptance Criteria:

- ✓ *Primary seals shall show no signs of degradation or compromise.*
- ✓ *Sterilization logs shall confirm that all retired samples were rendered non-viable per protocol.*
- ✓ *IPM records shall show no active infestations within the containment perimeter. (Reference: DBT_ICMR Collaborative national guidance document).*



4. OTHER IMPORTANT MSR

4.1. Facility Specific SOP for handling unknown pathogen samples:

The facility-specific SOP shall be developed by the Laboratory. These should include:

- 4.1.1. Operational SOP
- 4.1.2. Facility SOP
- 4.1.3. Test SOP
- 4.1.4. Equipment SOP
- 4.1.5. Handling SOP
- 4.1.6. Decontamination SOP
- 4.1.7. Disposal SOP.

Validation to meet the compliance: (Reference: DBT_ICMR Collaborative national guidance document).

4.2. Occupational health and safety programmes

- 4.2.1. Hazard communication
- 4.2.2. Respiratory protection
- 4.2.3. Hearing Protection
- 4.2.4. Fumigation, Fogging monitoring
- 4.2.5. Ethylene oxide monitoring
- 4.2.6. Anesthetic gas monitoring

Validation to meet the compliance: (Reference: DBT_ICMR Collaborative national guidance document).

4.3. Training and Competency of the staff

Validation to meet the compliance: Review of training records and practical competency test.

Acceptance Criterion: All personnel shall complete annual BSL-3 training and pass a written and/or practical exam with a score of >90%, covering sample handling, PPE donning/doffing and emergency response. Appropriate training of BSL-3 operational, servicing and maintenance staff of all engineering installations and handling of emergencies, should also be considered.

4.4. Medical Surveillance

Validation to meet the compliance: Review of Occupational Health Records.

Acceptance Criterion: All personnel shall have a current documented risk assessment and confirmation of required immunizations (as and if applicable), signed by the Occupational Health Physician, prior to independent entry into the BSL-3 for handling an unknown pathogen/sample.



4.5. Minimum Standard requirement for Lab Validation Process

- 4.5.1. Staffing Norms
- 4.5.2. People
- 4.5.3. Training
- 4.5.4. Orientation towards one health approach
- 4.5.5. Annual training plan
- 4.5.6. Scientific and Engineering and Maintenance

Validation to meet the compliance: (Reference: DBT_ICMR Collaborative national guidance document).

4.6. Regulatory Oversight and Emergency Response

The facility shall maintain an Emergency Action Plan (EAP) with immediate notification protocols for suspected releases. Regulatory officials shall be granted access to the facility for audits with prior information and logs should be maintained.

- ***Validation to meet the compliance:***
 - *Review of the EAP and emergency contact directory.*
 - *Evaluation of "Mock Drill" reports and incident templates.*
- ***Acceptance Criteria:***
 - ✓ *The EAP shall be accessible to all staff and contain current contact information for relevant facility, health and security authorities.*
 - ✓ *The facility shall provide documentation of at least one successfully completed mock drill or regulatory audit within the last 12 months.*

(Reference: DBT_ICMR Collaborative national guidance document).

4.7. Pre-Publication Review and Approval

All stakeholders, researchers, and project collaborators within the BSL Network shall obtain formal approval from the competent authority of the organization, institutional regulatory committee(s) and Scientific Steering Committee (SSC) of NOHM prior to the external submission or public dissemination of any article, manuscript, or data outcome derived from mission-related activities.

- ***Validation to meet the compliance:***
- *Ensure data fidelity, protect intellectual property, and maintain alignment with mission-wide strategic objectives.*
- ***Acceptance Criteria:***
 - ✓ *The manuscript should be submitted to the SSC at least 30 working days prior to the submission of the manuscript for publication.*
 - ✓ *A copy of the formal approval from the SSC Chairperson is documented in the project file. The article contains the standardized mission-related funding/support disclosure as prescribed by the SSC. No sensitive, proprietary, or unverified data should be released without specific SSC clearance.*

As per the DBT_ICMR Collaborative national guidance document.

4.8. Data Sharing and storage



Data sharing shall comply with Indian statutory requirements for sensitive data. Access is granted by Internal Governance Mechanism (IGM) of the laboratory through formal Data Sharing Agreements (DSAs). All data shall be de-identified before sharing or publication. Strict patient confidentiality is mandatory. Data and metadata shall be stored in machine-readable formats following standardized SOPs to ensure rapid retrieval and interoperability. Data shall be used for public health solutions. All outputs shall provide express credit to the source and adhere to "National Interest" public access requirements. Data custodian should be clearly defined.

- ***Validation to meet the compliance:*** *Technical data audit by experts.*
- ***Acceptance criteria:***
 - ✓ *A signed DSA on file and a documented approval/rejection, with remarks, as applicable.*
 - ✓ *Data shall be stored in structured, interoperable formats (SQL, JSON, CSV) with standardized metadata for rapid retrieval.*
 - ✓ *Physical or cloud storage shall be verified as located within Indian territory, as per statutory requirements.*
 - ✓ *Active encryption and un-editable audit logs for all access events shall be present.*
 - ✓ *Automated backup protocols and version control shall be in place to prevent data loss or accidental overwriting.*
 - ✓ *A verified "Anonymization Layer" shall be active to ensure no Personal Identifiable Information is stored in accessible research tiers.*

(Reference: DBT_ICMR Collaborative national guidance document).

5. BSL-3 LABORATORY RISK ASSESSMENT CHECKLIST

5.1. Pre-Assessment Preparation:

Checklist Item	Yes	No	NA	Remarks
Risk assessment schedule prepared and approved				
Risk assessment team identified (Lab In-charge, Team Leader, Team Members) Line of Response, in case of emergency, defined				
Roles and responsibilities clearly assigned				
Previous risk assessments reviewed				
Required documents gathered (design, SOPs, validation, training logs)				

5.2. Facility Design and Infrastructure:

Checklist Item	Yes	No	NA	Remarks
Containment zones clearly demarcated				
Airflow direction verified; negative pressure maintained				
Anterooms functional and interlocks working				
HEPA filters certified and documented				
HVAC performance validated; alarms functional				
Emergency exits accessible and visible Emergency exit door/its handle/name shall be red/green illumination/photoluminescent glow-in-the-dark green				
ETP system functional and validated				

5.3. Critical Utilities:

Checklist Item	Yes	No	NA	Remarks
Uninterrupted power supply available with backup				
Essential equipment connected to UPS				
Plumbing and gas supply systems functioning safely				
Drains free of leakage or backflow				
Ventilation and exhaust fans operational				

5.4. Laboratory Practices and Biosafety:

Checklist Item	Yes	No	NA	Remarks
SOPs for all workflows updated and available				
Staff follow PPE requirements consistently				

Incident response and spill management SOPs accessible				
Biosecurity practices implemented (access control, logs)				
Decontamination procedures (surface, equipment, room) followed				
Biological waste segregation ensured				
Sharps handling procedures followed				

5.5. Personnel-Related Requirements:

Checklist Item	Yes	No	NA	Remarks
Staff entry/exit procedures followed				
Personnel vaccination status verified (as and if applicable)				
Competency training completed and documented				
Annual refresher training conducted				
Fit testing for respiratory PPE completed				

5.6. Material, Sample and Waste Movement:

Checklist Item	Yes	No	NA	Remarks
Material entry/exit documented				
Sample movement tracked properly				
Transport containers leak-proof and labeled				
Waste treatment/disposal SOP followed				
Autoclave cycles validated with indicators				
Dedicated waste routes maintained				

5.7. Equipment and Operational Risks:

Checklist Item	Yes	No	NA	Remarks
Biosafety cabinets validated (annual/after relocation)				
Centrifuges used with sealed buckets				
Incubators, freezers and CO ₂ systems functional				
Calibration records up to date				
Maintenance logs available				
Alarms, sensors, and monitors working				

5.8. Risk Identification:

Checklist Item	Yes	No	NA	Remarks
Hazards identified in all operational areas				
Facility-level hazards identified				

Equipment-related failure modes documented				
Personnel and procedural risks reviewed				
Biosecurity risks assessed				

5.9. Risk Analysis:

Checklist Item	Yes	No	NA	Remarks
Severity assigned for each hazard				
Occurrence/frequency evaluated				
Risk detectability assessed				
Risk ranking (H/M/L) documented				

5.10. Risk Evaluation:

Checklist Item	Yes	No	NA	Remarks
Existing control measures evaluated				
Gaps identified in engineering controls				
Gaps identified in administrative practices				
Additional control measures recommended				
Action items assigned with timelines				

5.11. Risk Summary and Reporting:

Checklist Item	Yes	No	NA	Remarks
Risk assessment report prepared				
Summary of risks and mitigations completed				
Corrective and preventive action (CAPA) plan developed				
Documentation reviewed and approved by Lab In-charge				
Report archived and shared with relevant authorities				

5.12. Review & Follow-Up:

Checklist Item	Yes	No	NA	Remarks
Follow-up inspection scheduled				
Previous CAPA items closed				
Updated risk register maintained				

6. BSL-3 ANNUAL VALIDATION CHECKLIST

(Each item should be marked: **Completed/Not Completed/Not Applicable**, with space for observations)



6.1. Structural, Environmental and Facility Integrity:

6.1.1. Temperature and Relative Humidity

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Continuous monitoring system functional				
Calibration validity of sensors verified				
Temperature – values observed to be recorded				
RH – values observed to be recorded				
Annual validation with calibrated data logger performed				

6.1.2. Light Intensity

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Lux levels measured at:				
BSC work zone				
Lab bench tops				
Corridors/work areas				
Lux meter calibration verified				

6.1.3. Noise Level

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Noise measured at operator ear height				
Ambient noise level				



Audible alarms ≥10 dB above ambient				
Sound meter calibrated				

6.1.4. Containment Integrity

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Annual containment leak test performed				
Smoke test/tracer test performed				
No visible smoke/tracer escape				
Seal integrity verified (ducts, gaskets, joints)				
Negative pressure maintained under test conditions				

6.2. Pressure Cascade and HVAC Performance

6.2.1. Air Pressure and Differential Pressure

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Pressure cascade (10–15 Pa) verified between zones				
Base condition (equipment OFF) tested				
Dynamic condition (equipment ON) tested				



Pressure alarms tested and functional				
Digital pressure sensors calibrated				

6.2.2. AHU Performance Validation

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Fan operation, damper positions, belt condition checked				
Filter condition recorded				
Airflow rate measured and within acceptable limits				
AHU correlated with ACPH, air velocity, HEPA integrity				
Pass if within $\pm 10\%$ tolerance				

6.2.3. Airflow Pattern Visualization

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Smoke visualization conducted across all zones				
Airflow direction: clean → contaminated confirmed				
No reverse or stagnant air zones				
Documentation with photographs/videos				



6.2.4. Air Velocity, ACPH and Uniformity

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Air velocity at supply/exhaust (0.45–0.55 m/s $\pm 10\%$)				
Air changes per hour (ACPH) calculated				
BSL-3: 8–12 ACPH				
ABSL-3: 10–12 ACPH				
Uniformity of airflow validated across locations				

6.2.5. Air Particulate Count

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Particle counts ($>0.3 \mu\text{m}$, $>0.5 \mu\text{m}$, $>1 \mu\text{m}$, $>5 \mu\text{m}$) measured “At rest” and “In operation” counts recorded				
Meets ISO 14644 Class 8 criteria				
Particle counter calibration validated				



6.3. Filtration and HEPA Systems

6.3.1. HEPA Filter Integrity Testing

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Supply and exhaust HEPA filters tested				
PAO/DOP upstream challenge test performed				
Leak scan performed at filter gasket and perimeter				
Downstream penetration $\leq 0.01\%$				
Annual or post-maintenance test completed				

6.4. Critical Equipment Validation

6.4.1. Biological Safety Cabinets (BSCs)

Checklist item	Completed	Not Completed	Not Applicable	Remarks
HEPA integrity test				
Inflow/outflow velocity test				
Sash height alarm test				
Smoke test				
Certification stickers updated				

6.4.2. Autoclaves

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Biological indicator validation				
Bowie–Dick test (quarterly)				
Temperature mapping				



Cycle record verification				
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6.4.3. Effluent Decontamination System (EDS)

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Thermal profile validated				
Leak check performed				
≥6-log microbial kill confirmed				

6.4.4. CO₂ Incubators

Checklist item	Completed	Not Completed	Not Applicable	Remarks
CO ₂ calibration				
Temperature validation				
Quarterly verification				

6.4.5. Refrigerators/Freezers

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Temperature mapping				
High/low alarm checks				
Calibration record review				

6.4.6. Centrifuges

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Sealed rotor integrity check				
Speed calibration (if applicable)				
Preventive maintenance records				



6.4.7. Micropipettes

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Volume accuracy/precision calibration				

6.4.8 (For ABSL-3, if applicable):

IVC racks: HEPA integrity, airflow rate, rack pressure				
Blower units: flow and pressure test				
Cage change stations: BSC validation				

6.5. Operational and Safety Systems

6.5.1. Alarm, Safety and Utility Systems

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Door interlock alarms functional				
Pressure alarms functional				
Exhaust/supply failure alarms functional				
Fire alarm and suppression systems tested				
UPS load test and automatic switchover tested				
Gas supply and vacuum leak tests				

6.5.2. Waste Management Systems

Checklist item	Completed	Not Completed	Not Applicable	Remarks
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Waste segregation SOP followed				
Autoclave logs verified				
Internal transport procedures validated				
Temporary waste storage area inspected				

6.5.3. Water Supply and Drainage System

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Backflow prevention devices tested				
Drain traps functional				
No leakage in drainage network				
Eyewash/safety shower functional				

6.6. Administrative, SOP and Personnel Validation

6.6.1. SOP Review and Validation

Checklist item	Completed	Not Completed	Not Applicable	Remarks
All SOPs valid and facility-specific				
SOPs for unknown pathogen handling available				
Decontamination SOP validated				
Transport and waste SOPs validated				



6.6.2. Staff Training Competency

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Annual BSL-3 training completed				
Practical PPE donning/doffing test				
Emergency response drill				
Score $\geq 90\%$ in written/practical competency test				
Training records verified				

6.6.3. Occupational Health and Medical Surveillance

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Annual medical clearance documented				
Vaccination status updated				
Risk assessment signed by Occupational Health Physician				
Fitness-to-work certificate available				

6.6.4. Hazard Communication and Safety Programs

Checklist item	Completed	Not Completed	Not Applicable	Remarks
Respiratory protection program updated				
Hearing conservation program (if noise >70 dB)				
Chemical exposure monitoring (Formaldehyde, EtO, anesthetic gases)				



MSDS and chemical inventory verified				
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6.7. Documentation and Certification

6.7.1. Annual Validation Documentation

Checklist item	Completed	Not Completed	Not Applicable	Remarks
All validation reports compiled				
Calibration certificates updated				
Deviations recorded with corrective actions				
Facility compliance certificate issued				

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