INSTITUTIONAL
BIOSAFETY MANUAL

The purpose of this manual is to define the biological safety policies and procedures pertaining to research operations at Boston University (BU) and Boston Medical Center (BMC). The policies and procedures located within this document are designed to safeguard personnel and the environment from biologically hazardous materials and to comply with federal, state, and local regulatory requirements. All BU and BMC Principal Investigators (PIs) and laboratory workers must adhere to the biological safety policies and procedures in the conduct of their research and the management of their laboratories.

For information about specific biological safety programs for operations not covered in this manual, contact the Institutional Biosafety Committee (IBC) office or the appropriate Biological Safety Officer (BSO).

Chair, Institutional Biosafety Committee Associate Vice President, Research Compliance

Version/Year of Review	Summary of changes	Effective Date
2.2/2025	Contact updates, updates to regulatory information, policies, processes throughout document; addition of sections on plant work and nanomaterials; removal and revision of information for consistency; updated appendices.	12/16/2025
2.1/2024	Administrative changes: all hyperlinks updated and minor clarification provided to the required decontamination process for biosafety cabinet relocation or disposal.	11/16/2024
2.0/2022	Contact updates, formatting; regulatory updates to information on prion research, select agents, RAC review, references to BSL3 and 4, updates to roles and responsibilities, approval processes, biosafety procedures, PPE practices; updated appendices.	12/13/2022

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Biological Safety Program:
Purpose, Scope, and Responsibilities

Purpose

The purpose of this Biosafety Manual (BSM) is to define policies and procedures pertaining to use of biological materials in research at Boston University (BU) and under the jurisdiction of Boston Medical Center (BMC), as well as tenants of BU and BMC laboratory space. The policies and procedures within this document are designed to safeguard personnel and the environment from biologically hazardous materials without unduly limiting academic research. This manual also offers guidelines to comply with federal and state regulatory requirements.

The work practices, procedures, and policies specified in this manual are based on current regulatory requirements and best biosafety practices. Implementation of these measures will reduce the likelihood that an incident involving a biological agent will occur and will fulfill regulatory biosafety expectations. Laboratory microbiological work usually involves potential exposure to biological hazards, as well as to chemical and radiological hazards. Consequently, this manual should be used in conjunction with the BU Chemical Hygiene Plan and Radiation Safety Manual, respectively. This manual will be reviewed tri-annually by Environmental Health and Safety, in coordination with Research Compliance, and updated as needed in accordance with any change in local, state, or federal biosafety regulations.

For information about specific biological safety programs for operations not covered in this manual, contact the EHS Biological Safety Officer (BSO), the Institutional Biosafety Committee (IBC) office.

Scope

This manual applies to all individuals engaged in research at or under the jurisdiction of BU or BMC working with biological materials.

Definition of Biological Materials

Biological materials include:

- All infectious microorganisms (i.e., virus, bacteria, fungi, parasite, prion, rickettsia) and the toxins derived from such organisms (biological toxins) that can cause disease in humans or pose significant environmental or agricultural impact;
- Recombinant or synthetic nucleic acid molecules;
- Human or non-human primate materials including blood, plasma, serum, body fluids, unfixed tissues, organs, and cells;
- Field studies involving wild animals and vectors including samples that may be inherently infected or would be experimentally infected with Biosafety Level 2 (BSL2) or higher agents;
- Transgenic plants, animals, or vectors.

Note: *Please see Appendix E for further information on biological toxins.*

Biological Safety Program

The goals of the Biological Safety Program, referenced in this manual as the Biosafety Program, are to protect laboratory workers, the public, and the environment from potentially-hazardous biological agents. The Institutional Biosafety Committee (IBC) and the Biological Safety Officer (BSO) advocate the use of biosafety precautions that effectively reduce or eliminate the risk of exposure to potentially hazardous agents used in research. In developing its guidelines, the IBC and the BSO are ensuring that all policies and procedures are in accordance with institutional standards, regulatory frameworks governing the use of biological materials and best practices adopted nationally.

Roles and Responsibilities

The success of the Biosafety Program, like any other safety program, requires a team effort involving the IBC, Research Compliance staff, Principal Investigators (PIs), laboratory workers, the Research Occupational Health Program (ROHP), and Environmental Health and Safety (EHS). Principal Investigators are responsible for ensuring compliance with this policy as it applies to laboratory areas they may occupy, oversee or share, and ensuring that laboratory personnel who work under their supervision and occupy their laboratory space are informed of this policy and follow it. The BU administration, the IBC, and EHS endorse this manual and encourage active participation in maintaining high standards.

Associate Vice President, Research Compliance (AVPRC)

The AVPRC has overall responsibility for:

- Oversight for the control of hazards in the research laboratories and for ensuring that comprehensive, enterprise-wide programs are in place for the safe handling of all hazardous materials (e.g., biological, chemical, radiological);
- All non-financial research compliance at BU;
- Direct functional responsibility for the IBC, Biosafety Program, EHS, Laboratory Safety Committee, Laboratory animal use and care programs (Institutional Animal Care and Use Committees (IACUC), Institutional Review Board (IRB), BU animal care program, and other research-related oversight committees;
- Acts as the Institutional Official (IO) with responsibility for appointing officers required by regulation including the Biosafety Officer, the Federal Select Agent Program (CDC and USDA) Responsible Official among others.
- Acts as the Responsible Official (RO) for the City of Boston's Public Health Commission (BPHC) laboratory regulations;
- In consultation with provosts and deans at the Medical Campus and the Charles River campus, as well as the BMC leadership, appoints various committee members. The IBC, Research Compliance staff, the Research Safety Director, the BSO, ROHP, and EHS, have been charged with planning and implementing the Biosafety Program, the purpose of which is to protect all personnel, the public, and the environment from biohazardous or infectious agents.

Institutional Biosafety Committee (IBC)

The IBC is responsible for the oversight of biological research within the Biosafety Program at BU. The IBC carries out these functions pursuant to requirements set forth by the National Institutes of Health

(NIH), the Centers for Disease Control and Prevention (CDC), Occupational Safety and Health Administration (OSHA), the City of Boston Public Health Commission (BPHC), the Massachusetts Department of Public Health (DPH), and Boston University.

Further information on the IBC's responsibilities are located in the IBC charter.

IBC Program

The IBC Program staff (housed in Research Compliance) responsibilities include, but are not limited to:

- Initiating the review process of the Biosafety Manual in consultation with EHS, ROHP, and other BU stakeholders;
- Development and implementation of programmatic needs and policy related to biosafety in research in accordance with institutional, city, state, and federal regulatory requirements;
- Managing submission of IBC applications (also known as BUAs) and their reviews via the RIMS submission system;
- Monitoring and communicating with stakeholders in consultation with EHS, BSO, ROHP, and other offices as needed;
- Providing ancillary review of IACUC and IRB protocols that use biohazardous materials;
- Advising PIs on IBC applications in the RIMS system and in completing their IBC applications;
- Assigning review of IBC applications for scheduled IBC meetings and designated member review;
- Documenting IBC meeting discussion and votes;
- Submitting applicable protocols and tracking any approval requirements;
- Submitting reportable incidents and annual IBC registration updates to NIH.

Biological Safety Officer (BSO)

The BSO is responsible for providing guidance on the safe handling of biological agents and contributing to the overall management of the Biosafety Program. The BSO is a voting *ex-officio* member of the IBC. The BSO's responsibilities include, but are not limited to:

- Providing technical advice to the IBC, PI and researchers on laboratory containment, security, and safety procedures;
- Working with EHS on oversight of periodic and unscheduled inspections to ensure that laboratory standards are rigorously followed and maintained;
- Developing emergency plans for handling spills and personnel contamination;
- Developing guidelines and procedures for safe laboratory practices and overseeing their implementation;
- Developing design specifications and criteria for laboratory containment facilities;
- Developing and implementing training programs in consultation with the IBC;
- Preparing and providing reports on the Biosafety Program to the IBC. The BSO's report includes routine operational updates, any significant problems or regulatory violations and any research-related accidents or illnesses;
- Working with the appropriate Institutional Official of BU, BMC, and BU tenant laboratories to prepare, renew, and submit the recombinant DNA permit application to BPHC;
- Preparing response to BPHC on inquiries concerning laboratory related incidents reported by ROHP;

- Acting as a liaison and working with regulatory agencies during visits and inspections of laboratories;
- Overseeing follow-up investigation and response to laboratory incidents involving transgenic animals and biohazardous materials.

Research Safety Director

The Research Safety Director (RSD) works with the different safety committees to ensure that laboratory research is conducted in appropriate facilities, using safety equipment, and implementing safety processes. The RSD's responsibilities include, but are not limited to:

- Overseeing Research Safety activities and response during regulatory inspections and visits to laboratories;
- Providing update as necessary to institutional leadership;
- Working with the AVPRC, Research Compliance, IBC, and other institutional officials to coordinate responses to any regulatory findings;
- Reviewing new and proposed regulations and other requirements and summarizing the impact to the institution;
- Determining programs and processes necessary for effective and safe process to conduct laboratory research.

Environmental Health and Safety

Various programs within EHS work closely with the BSO to ensure that operations for which EHS has responsibility are conducted in accordance with the criteria and guidelines established by the BSO. These include, but are not limited to:

- Storage and disposal of biological and medical waste;
- Selection of appropriate Personal Protective Equipment (PPE) for individuals working with hazards, including biohazardous materials;
- Performing fit testing of research and support laboratory personnel required to use respiratory protection based on the hazard assessment of the agent/s handled and procedures performed;
- Development and implementation of emergency response and preparedness plans;
- As necessary, assess and monitor work areas, including the presence of allergens;
- Performing job hazard analysis upon request when reproductive hazards are used in a laboratory.

Department Chairperson

The Chairperson of the department is responsible for overseeing the overall implementation of safe practices and procedures of laboratories within their department.

- Ensuring that the IBC and EHS are informed of new in-coming or out-going Principal Investigators (PIs) to assist in their transitions including close out of their IBC protocol;
- Informing EHS of planned laboratory relocations;

- Assisting in addressing laboratory inspection findings for laboratories that have continuously failed to address those findings as requested by EHS.

Principal Investigators (PIs)

Principal Investigators (PIs) or Laboratory Directors are responsible for ensuring compliance with this manual as it applies to laboratory areas they may occupy or oversee; and ensuring all laboratory personnel who work under their supervision and occupy their laboratory space are aware of this manual and understand how it applies to their areas. PI responsibilities include, but are not limited to, the following:

- Conducting an appropriate risk assessment of the planned research project which includes, without being limited to the following: hazard associated with the agent/s under study (e.g., assessment of risk associated with research on BSL2 agent/s), procedures to be performed, appropriate primary containment that may be necessary (e.g., use of a biosafety cabinet), personal protective equipment (PPE) use, etc. Risk assessments shall be completed and shall be a part of the protocol submitted for review and approval by the IBC:
- Ensuring that specific laboratory hazards are effectively communicated to laboratory personnel, personnel have received appropriate training and are competent to conduct procedures performed in the laboratory, and that appropriate controls are in place to minimize risks associated with these hazards;
- Ensuring that specific laboratory hazards are effectively communicated to laboratory visitors prior to entering the laboratory, including posting of signs at the entrance to the laboratory to inform individuals of laboratory hazards, and that visitors have met the specific requirements prior to entering or exiting the laboratory;
- Developing standard operating procedures (SOPs), when warranted, to address the hazards so that work can be performed safely;
- Ensuring that engineering controls are available, in good working order, and are used appropriately to prevent or minimize exposure to biohazardous materials;
- Ensuring that appropriate PPE are available and used by laboratory personnel;
- Ensuring that all laboratory personnel receive appropriate training including biosafety training that is conducted as part of the Biosafety Program, as well as specific training on the hazards, procedures, and practices relevant to the laboratory in which they are working; documenting and maintaining all training records;
- Ensuring that research personnel have completed their required medical clearance from ROHP before starting work on an approved IBC protocol;
- Notifying the IBC and obtaining IBC approval prior to starting work with recombinant or synthetic nucleic acid molecules and/or biohazardous material, and making sure that the lab conforms to all terms and conditions of the IBC approval;
- Ensuring that the approved IBC protocol is up to date and in active status;
- Ensuring that laboratory workers are provided access to immunizations and medical surveillance prior to, and in the event of, exposure to biohazardous agents as appropriate (based on current CDC and ROHP recommendations), immunizations are provided through BU's ROHP;
- Notifying the BUMC Control Center at (617) 358-4144 or EHS emergency telephone, (617) 353-2105 at the Charles River Campus, of any spills or incidents involving biological agents or recombinant and synthetic nucleic acids. The Control Center will notify the BSO;

- Ensuring that all biological wastes including biological agents are disposed of according to regulations, as outlined in this manual;
- Ensuring that biohazardous materials to be transported are packaged, transferred, or shipped in accordance with regulations and BU policies;
- Ensuring that periodic self-inspections of the laboratory are conducted by the PI or designee;
- Maintaining liaison with the EHS Department Safety Specialist (DSA) and the BSO.
- Ensuring that all changes to work conditions including materials, equipment and space being used are reported to the BSO and IBC so that risk assessments, including those from EHS and ROHP, can be updated in consideration of those changes. Providing designated Lab Safety Coordinators with the mechanisms to fulfill their roles as described below or assuming responsibility directly.

Laboratory Safety Coordinators

The Laboratory Safety Coordinator's responsibilities include, but are not limited to:

- Supporting the PI and ensuring that safety practices are implemented and followed in the lab's daily operations;
- Work with ROHP to ensure researchers are medically cleared for the job activities that require medical surveillance as identified by the PI;
- Representing the PI in matters related to the implementation of laboratory and worker safety;
- Serving as the primary laboratory contact with EHS for issues related to safety (e.g., biological, chemical, fire, general safety, controlled substances, etc.);
- Taking positive actions to help reduce the potential for accidents and incidents associated with laboratory operations;
- Instructing all laboratory personnel and students in safe work methods;
- Informing laboratory personnel and/or students of the safety hazards associated with their work;
- Reporting all accidents or safety concerns to the PI and EHS;
- Ensuring that appropriate SOPs are established and that lab personnel and students are appropriately trained to follow them;
- Working with EHS to determine best safe practices and procedures;
- Working with EHS to ensure that lab personnel and students complete all required safety trainings in a timely manner;
- Ensuring that all deficiencies identified by EHS or outside regulatory inspectors are addressed and corrected within the time required;
- Participating in the incident review process;
- Stopping operations that are in clear violation of the safety requirements, approved SOPs, or may potentially result in injuries or potential exposures;
- Serving as liaison and informing EHS when new PI will set up a new laboratory or when departing and closing out the laboratory.

Research Occupational Health Program (ROHP) and Occupational Health Officer (OHO)

The OHO has overall responsibility for the ROHP and is responsible for reporting BU exposure incidents involving select agents (see Chapter 9 CDC/USDA Select Agents). ROHP is responsible for establishing

and performing appropriate medical surveillance for all personnel performing or supporting research, such as animal care workers, facilities, EHS, IT, Police, and Public Safety. Medical surveillance is required at the time of hire, or transfer into the research environment, prior to beginning work on an IBC-approved protocol, and periodically depending on changes in the work environment, occupational exposure, and risk for each position or job category. The ROHP and OHO's responsibilities include, but are not limited to:

- Reviewing and providing medical surveillance based on the job activities in the work environment assigned by the supervisor or PI;
- Developing exposure treatment and management plans for laboratory incidents and animal exposures or incidents, including post-exposure management and monitoring;
- Participating in IBC proceedings, including application reviews for quality improvement and risk mitigation;
- Participating in Laboratory Safety Committee, IBC, IACUC, and other committee meetings, as required, to ensure appropriate medical surveillance is in place;
- Creating and issuing wallet-sized medical surveillance agent cards for researchers or research support staff who may potentially be exposed to laboratory areas, animals, biological or other hazardous agents or materials. This card contains ROHP contact information; the card must be presented to a health care provider if the card holder has an unexplained illness;
- Developing Agent Information Sheets which are available on the ROHP website;
- Developing emergency medical response protocols for support staff and personnel approved by the IBC to work with biological agents with potential to cause laboratory acquired infection;
- Assisting PIs and EHS on request in the preparation and presentation of biosafety and agent specific training;
- Providing medical support coverage 24 hours a day, 7 days a week, for researchers and other personnel to call for triage, risk assessment, evaluation of laboratory incidents or exposure concerns, and medical care. referral based on severity, location and time of laboratory exposure or incident;
- Working with EHS on request to develop SOPs and appropriate health and safety practices;
- Performing return-to-work assessments in conjunction with BPHC for laboratory exposures involving "high risk" biological agents.

Laboratory Workers

Laboratory workers are an important element in developing and maintaining a safe laboratory environment. An incident caused by one laboratory worker can have a widespread effect on others. Laboratory worker responsibilities include, but are not limited to:

- Following policies, procedures and practices established by BU, EHS, IBC, and the laboratory;
- Using best biosafety laboratory practices to minimize exposures to biological agents and to prevent or avoid other incidents (such as personal injuries, chemical and radiation spills, laboratory fires, explosion, etc.);
- Complete medical tasks or services that are triggered from the Job Activities assigned by their PI or supervisor so that they are medically-cleared for their laboratory work within the assigned deadline. Lab workers who are required to obtain medical clearance and do not follow-through

on the requirements are in non-compliance with university policies and potentially at risk should there be exposure in the lab;

- Completing the required Laboratory Safety Training and other appropriate safety trainings thereafter; Following established procedures and report spills, accidents, and unsafe laboratory conditions to the PI, EHS, and other responsible parties;
- Utilizing appropriate control measures, such as biological safety cabinets and PPE, to prevent or minimize exposure to biological agents and contamination of personnel and facilities;
- Immediately contact ROHP at (617) 358-ROHP (7647) in the event of exposure or injury so that medical triage and evaluation, documentation and notification can be performed.

Chapter 2
Requirements and Approval of Research Projects

IBC Approval

Principal Investigators (PIs) planning to conduct research using recombinant or synthetic nucleic acid molecules and/or hazardous biological materials that pose a potential risk to the health of humans, animals, or plants, either directly through infection or indirectly through damage to the environment, must submit an IBC application to the IBC program for review and approval by the IBC. Researchers using any of the following must complete and submit an IBC application for review and approval:

- Recombinant and synthetic nucleic acid molecules;
- Hazardous biological agents (e.g., virus, bacteria, fungi, parasite, prion, rickettsia etc.);
- Other potentially infectious materials (e.g., human or non-human primate blood, serum, plasma, body fluids, unfixed tissue, organ and cells);
- Inactivated Biological Samples derived from BSL3 or higher agents;
- Select agents and biological toxins;
- Transgenic animals or plants;
- Human gene transfer;
- Sheep and any tissues derived from them (these tissues can transmit *Coxiella burnetii*, the causative agent of Q-fever), and;
- Field studies with wild animals and vectors and their tissues potentially infected or would be experimentally infected with BSL2 or higher agents.

For work with potentially infectious agents on human or animal subjects, IBC review and approval is necessary in addition to review and approval by the appropriate IRB or IACUC. For more information on the IBC review process, please refer to the IBC website.

Training and Medical Clearances

All individuals listed on an IBC application must complete specific set of biosafety trainings and ROHP medical clearance requirements before engaging in any approved protocol related activities as detailed in the IBC Training Requirements website. Additionally, laboratory specific training may also be required depending on the type of biohazardous agents used in a protocol.

Materials that May Require Further Clarification During IBC Review

Biohazardous and Potentially Infectious Materials

It is important that the PI clarifies the intended use of the agents, how risks associated with its handling are mitigated, and the management of the wastes generated from such work. If a PI intends to use hazardous biological agents that are not mentioned in the list of biological agents with potential to cause a laboratory acquired infection, they should contact the IBC Program or BSO for advice regarding proper completion of the IBC application. In addition, ROHP has developed "Agent Information Sheets" that are available on ROHP's website. When applicable, agent specific vaccines will be offered at no charge to all persons who are approved by the IBC to work with or could potentially get exposed to these materials.

Recombinant or synthetic nucleic acid Materials
Specific guidelines on the facility containment and regulatory requirements should be followed depending on the exact detail of the proposed work. A PI must follow the defined containment directive. There may be experiments not covered by the NIH Guidelines that require further review and approval by external, state or federal agencies before initiation. If the experimental protocol is not covered by the NIH's guidelines, contact the BSO at (617) 358-7840 at BUMC and (617) 353-4094 at CRC to determine further review requirements.

Human Gene Transfer
After review by the IBC office, in consultation with the IBC Chair, any protocols involving human gene transfer will be sent to the BU Human Gene Transfer subcommittee for review prior to the IBC, as well as the appropriate BU Institutional Review Board (IRB). These studies remain subject to Federal Drug Administration (FDA) and other clinical trial regulations, and only after FDA, IRB, IBC, and other relevant approvals are in place can these protocols proceed. *Note:* the deliberate transfer of recombinant or synthetic nucleic acids into a human research participant conducted under a FDA regulated individual patient expanded access Investigational New Drug (IND), including for emergency use, is *not* research subject to the *NIH Guidelines* and thus does not need to be submitted to the IBC for review and approval.

For more information about IBC approval of human gene therapy protocols, contact the IBC Office at (617) 358-7910 or ibc@bu.edu. For information about IRB submissions, contact the BUMC IRB at (617) 358-5372 or medirb@bu.edu or the CRC IRB at (617) 358-6115, or irb@bu.edu.

Dual Use Research of Concern
Although biological research produces useful knowledge for health, and creates beneficial products and technologies, some research may present biosafety and biosecurity risks or provide knowledge, information, products, or technologies that could be misapplied. The misapplication of such research has the potential to do harm with no, or only minor, modification that can pose a significant threat with potential consequences to public health and safety, agricultural crops and other plants, animals, the environment, materiel, or national security. Any research that may fall into the categories stated above is defined as Dual Use Research of Concern (DURC). Academic research institutions are expected to comply with the federal requirements for institutional Dual Use Research of Concern (DURC) oversight. This federal requirement applies to all institutions that receive federal funding to perform life science research that meets DURC criteria. BU's DURC Committee (DURCCom) is responsible for the oversight and implementation of the standards in the research community at BU. For further information, please see the BU Dual Use Research of Concern Policy.

Use of Animals
All PIs planning to use recombinant or synthetic nucleic acid molecules and/or biohazardous materials in their laboratory or in research animals must receive IBC approval prior to commencing the work. The use of animals in research also requires compliance with the "Animal Welfare Act," administered by the USDA's Animal and Plant Health Inspection Service (APHIS), the "Public Health Service Policy on Humane Care and Use of Laboratory Animals," administered by NIH's Office of Laboratory Animal Welfare (OLAW), and all applicable state, local or University regulations covering the care and use of animals. All protocols involving the use of live animals must be reviewed and approved by the IACUC before implementation.

Transgenic Animals
PIs who create transgenic animals, either in the PI's lab or through the BU Transgenic Core facility, as well as PIs who use transgenic animals at ABSL2, or at ABSL1 if not considered exempt under the NIH Guidelines (Section III-E, Appendix C-VIII) must submit an IBC application and receive IBC approval prior to initiation of experimentation. In addition, the IACUC must approve the protocol. The IACUC can be reached at IACUC@bu.edu.

Transgenic Plants
The development of transgenic plants must be reviewed and approved by the IBC. These plants are considered recombinant organisms and subject to the NIH rDNA Guidelines. Transgenic plants are typically created by introduction of foreign DNA into the plant's genome. The IBC will recommend the appropriate level of Biosafety Containment for the housing and growing of plants based on the nature of the plant materials, upon its review (See Appendix F).

Tissue Culture/Cell Lines
All tissue and cell cultures derived from humans are considered potentially infectious under the OSHA Bloodborne Pathogens Standard, Title 29 Code of Federal Regulations (CFR) Part 1910.1030 and must be handled in a laboratory using BSL2 principles, practices and facilities (the concept of "universal precautions"). Nonhuman primate tissues and cell cultures are also considered potentially infectious and must be handled in the BSL2 laboratory. Individuals working with these materials in the laboratory are considered to have potential exposure to bloodborne pathogens, such as human immunodeficiency virus (HIV) and hepatitis B virus (HBV), and must be included in the Bloodborne Pathogens program. These individuals must complete bloodborne pathogens training and must be offered the HBV vaccination. When cell cultures are used in the context of Risk Group 3 or 4 agents, the cell lines will be classified at the same higher containment level and must be handled in BSL3 or BSL4 containment.

Select Agent Biological Toxins
The use of select agent biological toxins generally needs approval by the IBC (see Appendix E for more details). Please check with IBC staff directly if you have questions regarding a particular biological toxin. A current and updated list of these CDC and USDA Select Agent Toxins may be found here.

Testing and Certification of Containment Equipment Required
Handling infectious or potentially infectious materials could potentially generate infectious aerosols. Such work must be performed in a Biological Safety Cabinet (BSC) while wearing appropriate PPE. All BSCs are required to be tested and certified when they are first installed. They are also required to be tested and recertified annually. BU works with an experienced outside vendor certified to test and repair this equipment. Laboratories must ensure that their BSC certifications are current before their use in protocols. EHS will verify their status during BUA inspection of protocols under review by the IBC.

Nanomaterials use with Biological Materials
Research protocols involving the use of nanomaterials with microbiological agents, biological samples, recombinant or synthetic nucleic acid molecules, shall be reviewed and approved by the IBC. This may include adding nanomaterials to cells, tissues *in vitro*, into animals *in vivo*, or using the nanomaterial as a delivery tool to carry nucleic acids and other molecules. The IBC works in conjunction with EHS in assessing the risks posed by the combination of these materials and provides appropriate guidance for safe handling, storage and disposal.

Completion of Laboratory Safety Trainings

All PIs and laboratory personnel that are listed on the protocol must complete their required lab safety trainings, available online via SciSure (formerly known as SciShield). These trainings must be completed prior to initiation of lab work and need to be refreshed annually. EHS will verify that the PI and personnel that are listed on the protocol have completed the required training during BUA inspection of protocols under review by the IBC.

Chapter 3
Biosafety Principles

The risk of exposure to biological agents in a research environment depends on a number of factors (., the agent, its virulence, subject's susceptibility, route of transmission, etc.) In general, the biosafety procedures followed are designed to prevent such exposures by containing the agents being handled and using the appropriate types of PPE. To properly design the containment, it is important to recognize the potential routes of transmission for the given agent.

Routes of Transmission

Skin and Mucous Membrane Contact

Procedures, such as decanting of liquids, pipetting, opening of screw caps, vortexing, streaking agar plates, and inoculation of animals, can potentially result in the generation of splatters of infectious droplets, as well as direct contact with infectious material. Direct contact to the mucous membranes of the eyes, nose, and mouth is considered a route of exposure. Wearing of contact lenses in the laboratory is prohibited. Appropriate eye and face protection should be used when performing work that generate splashes.

Ingestion

Performing mouth pipetting is prohibited in the laboratory. It presents a risk of ingesting infectious material. Touching the face and mouth when working can cause an oral exposure to the infectious material being handled. Storage of food or drinks and their consumption is prohibited. Laboratory personnel should refrain from the use of personal electronic mobile devices such as cell phones, headphones, and earbuds when working in the laboratory. They can cause distractions and could potentially become contaminated.

Percutaneous Inoculation

Use of syringes, needles, and other sharps presents the greatest risk of exposure through inoculation. Accidental inoculation can also occur as a result of cuts and scratches from contaminated items including syringes used for animal inoculations, as well as animal bites.

Inhalation

Certain procedures have the potential for generation of respirable aerosols, including sonication, centrifugation, "blowing out" of pipettes, heating inoculating loops, and changing bedding from the cages of infected animals. Aerosol refers to a droplet of 5 microns or less in diameter that can potentially be suspended in air for some amount of time. Aerosols may contain infectious particles.

Containment

The term "containment" is used to describe safe methods for handling infectious agents in the laboratory environment. The purpose of containment is to reduce or eliminate exposure of laboratory workers, other people, and the outside environment to potentially hazardous agents. The four elements of containment include administrative controls, work practices, personal protective equipment, and facility design.

Primary Containment
The protection of personnel and the immediate laboratory environment from exposure to infectious agents is provided by good microbiological techniques, use of appropriate PPE, and the use of appropriate safety equipment, such as the Biological Safety Cabinets (BSCs).

Secondary Containment
Protecting the laboratory's external environment from exposure to infectious materials is accomplished by a combination of the facility design and safe operational practices. The risk evaluation of the work to be done with a specific agent will determine the appropriate combination of these elements

Safety Equipment

Safety equipment includes biological safety equipment, sealed containers, safety centrifuge cups, down-draft tables, and other engineered controls designed to minimize exposure to biological agents. Biological safety cabinets are among the most important safety equipment for protection of personnel and the laboratory environment, and most also provide product protection. Safety equipment is most effective at minimizing exposure when workers are trained in the proper use of such equipment and the equipment is regularly inspected and maintained.

Biological Safety Cabinet (BSC)

Because of its importance in providing containment and safety protection in the laboratory, a BSC is considered one of the most critical pieces of safety equipment in biological laboratories. When used appropriately, BSCs are designed to capture aerosols that may be generated during work with biological materials. The BSC is equipped with a high efficiency particulate air (HEPA) filtration that filters the air in BSC before being recirculated or exhausted. There are three types of BSCs (Class I, II, and III) used in laboratories. Open-fronted Class I and Class II BSCs are containment devices that provide a primary barrier offering significant levels of protection to laboratory personnel and to the environment when used in combination with good laboratory technique.

The *Class I BSC* is suitable for work involving low-to-moderate risk agents where there is a need for containment, but not for product protection. It provides protection to personnel and the environment from contaminants within the cabinet but does not protect the work within the cabinet from "dirty" room air.

The *Class II BSC* protects the material being manipulated inside the cabinet (e.g., cell cultures, microbiological stocks) from external contamination. It meets requirements to protect personnel, the environment, and the product. The two basic types of Class II BSCs are Type A and Type B. The primary difference between the two types may be found in the ratio of air that is exhausted or recirculated and the manner in which exhaust air is removed from the work area.

The *Class III BSC* is typically used to work with highly infectious microbiological agents including those classified as RISK Group IV or agents requiring a BSL4 laboratory. It provides the maximum protection to personnel, the environment, and the product.

Another type of device that is typically referred to as a "laminar flow" or a "clean bench" can sometimes be seen used in the lab. It is important to note that this device *must not* be utilized for work with

biohazardous or chemically hazardous agents. These units provide *product protection* by ensuring the product is exposed only to clean HEPA-filtered air. They do not provide protection to personnel working or the environment.

PIs are responsible for ensuring the proper maintenance of lab equipment. BSCs used as primary barriers must be certified annually by a qualified vendor. Contact the Biosafety Officer or EHS for information about testing and certification vendors or other BSC-related information. Proper operation and maintenance of a BSC requires knowledge of how the system operates, as well as training and experience in effective techniques for working within the cabinet volume without compromising its functions. Additional details concerning the design and use of BSCs are provided in Appendix B.

Two specialized forms of quality control are strongly recommended for all BSCs and are required for cabinets used to contain Risk Group 2 or higher agents:

- At least daily, or each time the BSC is operated, the operator or user should observe the magnahelic gauge and note its relative position. Magnahelic gauges measure the pressure drop across the outlet HEPA filter and are important indicators of filter integrity and loading. The gauge will typically indicate the same measurement over a long period of time. A significant change in the reading over a short period of time may indicate an issue including clogging or a leaking HEPA filter. In such cases, the BSC should not be used until the problem is identified and resolved. ***If the BSC located within a laboratory does not have a magnahelic gauge, users must understand the operation of the airflow monitor, controls, and alarm settings.***

- The BSC must be tested and recertified annually or after it is moved to a different location. Testing and recertification is performed by a contractor certified to test the BSC. EHS can provide information on qualified vendors. The certification ensures that the BSC is meeting its operating specifications and providing maximum protection. In addition, certifiers provide service and preventive maintenance for BSC and can often forecast expensive requirements like HEPA filter replacements, allowing PIs to budget for the event.

  If BSC recertification is required, the recertification must be completed before the current certification expires. If the certification lapses, the BSC may not be used for BSL2 or higher procedures until it is recertified. The lab will report the lapsed recertification to EHS immediately. EHS will inform the PI and lab workers not to use the BSC and affix an "OUT OF SERVICE" notice and assist the lab to get the BSC recertified. Unless a good reason exists for more frequent certification, a one-year certificate life is appropriate. The certificate will generally expire on the last day of the month in which the certification was performed, one year later (i.e., a certificate issued on June 2, 2021 will expire on June 30, 2022).

Personal Protective Equipment (PPE)
PPE is designed to protect the wearer against chemical, biological, radiological, or mechanical irritants. Different types and levels of PPE may be required for individuals, depending upon each person's role within a laboratory, the materials handled, and the work being performed.

In order to ensure that all laboratory faculty, staff, students, affiliates and visitors are sufficiently protected from the hazards present in the workplace, the Personal Protection Equipment in Laboratories Policy has established the minimum PPE requirements, based on best laboratory practices.

Facility Design

The design of a facility is important in providing a barrier to protect people working inside and outside the laboratory, as well as to protect people or animals in the community from infectious agents that may be accidentally released from the laboratory. Facility design must be commensurate with the laboratory's function and the recommended biosafety level for the agent being used or stored.

The recommended secondary barrier(s) will depend on the risk of transmission of specific agents. For example, the exposure risks for most laboratory work in BSL1 and BSL2 facilities will be direct contact with the agents or inadvertent contact exposures through contaminated work environments. Secondary barriers in these laboratories may include separation of the laboratory work area from public access, availability of decontamination equipment (e.g., autoclave), and handwashing facilities. In BSL3 facilities, additional safeguards, such as directional airflow airlock-controlled entry and exiting, a shower used for personnel to shower out may be required.

As the risk for aerosol transmission increases, higher levels of primary containment and multiple secondary barriers may become necessary to prevent infectious agents from escaping into the environment. Such design features could include specialized ventilation systems to ensure directional airflow, air treatment systems to decontaminate or remove agents from exhaust air, controlled access zones, an airlock at the laboratory entrance, or separate buildings or modules for physical isolation of the laboratory building itself.

Note: *It is EHS policy that autoclaves used to sterilize biohazardous materials be validated quarterly using a sporulation test and that validation records be kept (See Appendix C). Unless sterilization of the waste is otherwise specified by the IBC, solid biohazardous waste should be directly disposed into red bags as medical waste without autoclaving.*

Biosafety Levels

Four (4) biosafety levels (BSLs) represent combinations of laboratory practices and techniques, safety equipment, and laboratory facilities. Each combination is specifically appropriate for the operations performed and the documented or suspected routes of transmission of the infectious agents, as well as for the laboratory function or activity. The recommended biosafety level for an organism represents the conditions under which the agent can be ordinarily handled safely.

As a general rule for selecting the appropriate laboratory containment, the biosafety safety level (BSL) selected should correspond with the highest risk group (RG) category of the organisms involved. For example, work with vaccinia virus which is a Risk Group 2 (RG2) agent, should be conducted at BSL2 or higher and *Mycobacterium tuberculosis* which is a Risk Group 3 (RG3) should be conducted at BSL3.

Descriptions of biosafety levels, as well as assigned biosafety levels for specific organisms, are contained in the CDC/NIH document, *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) 6th edition, and in the below table:

Biosafety Level (BSL)	Agents	Practices	Primary Barriers and Safety Equipment	Facilities (Secondary Barriers)
1	Not known to cause disease in healthy adults; RG1	Standard microbiological practices	• No primary barriers required. • PPE: laboratory coats and gloves; eye, face protection, as needed	Open bench top, sink required
2	Associated with human disease, which is rarely serious and for which preventive or therapeutic interventions are *often* available; RG2	BSL1 practice plus: • Limited access • Biohazard warning signs • Sharps precautions • Biosafety manual • BMBL BSL2 practices and procedures	• *Primary barriers:* BSCs or other physical containment devices used for manipulations of agents that cause splashes or aerosols of infectious materials. • *PPE:* lab coats; gloves; eye/face protection as needed.	BSL1 plus: • Autoclave available • BMBL BSL2 facility design
3	Associated with human disease for which preventive or therapeutic interventions *may be* available; RG3	BSL2 practice plus: • Controlled access • Decontamination of all waste • Decontamination of lab clothing before laundering • Baseline serum as recommended by Occupational Health • BSL3 Biosafety Manual • BSL3 SOPs • BSL3 Emergency Response Plans (ERPs) • BMBL BSL3 practices and procedures	• *Primary barriers:* BSCs or other physical containment devices used for all open manipulations of agents. • *PPE:* protective lab clothing; gloves; face, eye and respiratory protection as needed.	BSL2 plus: • Physical separation from access corridors • Self-closing, double-door access • Exhausted air not recirculated • Negative airflow into laboratory • BMBL facility design and verification
4	Agents are likely to cause serious or lethal human diseases for which preventive or therapeutic interventions *are not usually* available; RG4	BSL3 practices plus: • BMBL BSL4 practices and procedures • BSL4 Biosafety Manual • BSL4 CDC/BPHC Plans • BSL4 SOPs • BSL4 ERPs	*Primary barriers:* All procedures conducted in the BSC <u>in combination with</u> full-body, air-supplied, positive-pressure personnel suit.	BSL3 plus: • BMBL BSL4 facility design

Note: *Consult the BMBL 6th edition for a more complete description of the four biosafety levels, as well as recommended biosafety levels for specific organisms.*

Biosafety Level 2+ ("BSL2+")
"BSL2+" is a hybrid designation for laboratories performing biohazardous research protocols that require more stringent laboratory practices than those outlined for BSL2. Generally, BSL3 practices are followed during such work to provide additional level of safety to the workers such as the use of a solid front and splash resistant lab coat, double gloves and use of full-face protection, The BSL2+ practices are outlined by the PI in their protocol and reviewed by the IBC and the BSO as part of the protocol approval.

In addition to the four (4) biosafety levels described above, there are also four (4) biosafety levels for work with infectious agents in vertebrate animals, referred to as the Animal Biosafety Level (ABSL):

Animal BSL/ ABSL	Agents	Practices	Primary Barriers and Safety Equipment	Facilities (Secondary Barriers)
1	Not known to consistently cause disease in healthy human adults; RG1	Standard animal care, use, and management practices, including appropriate medical surveillance programs.	• As required under the Program of Veterinary Care and Use of Animals at BU for normal care, use, and handling of each species. • *PPE*: laboratory coats and gloves; eye, face protection, as needed	• Standard animal facility, remote housing facility, or laboratory • No recirculation of exhaust air • Directional air flow recommended • Hand washing sink recommended
2	• Agents associated with human disease; • Hazard: percutaneous injury, ingestion, mucous membrane exposure RG2	ABSL1 practices plus: • Limited access Biohazard warning signs • Sharps precautions Biosafety manual • Decontamination of all infectious wastes and of animal cages prior to washing	ABSL1 equipment plus primary barriers: • Containment equipment appropriate for animal species. • *PPE*: laboratory coats, gloves, face, eye and respiratory protection as needed.	ABSL1 facility plus: • Autoclave available • Hand washing sink available in the animal room. • Mechanical cage washer recommended. • Negative airflow into animal and procedure rooms recommended

3	Indigenous or exotic agents that may cause serious or potentially lethal disease through the inhalation route of exposure; RG3	ABSL2 practices plus: • Controlled access • Decontamination of clothing before laundering • Cages decontaminated before bedding removed • Disinfectant foot bath as needed	ABSL2 equipment plus: • Containment equipment for housing animals and cage dumping activities • Class I, II or III BSCs available for manipulative procedures (inoculation, necropsy) that may create infectious aerosols. • *PPE*: appropriate respiratory protection	ABSL2 facility plus: Physical separation from access corridors Self-closing, double-door access Sealed penetrations Sealed windows Autoclave available in facility Entry through ante-room or airlock Negative airflow into animal and procedure rooms Hand washing sink near exit of animal or procedure room
4	• Dangerous/exotic agents which post high risk of aerosol transmitted laboratory infections that are frequently fatal, for which there are no vaccines or treatment. • Agent with a close or identical antigenic relationship to an agent requiring BSL4 until data are available to redesignate the level. • Related agents with unknown risk of transmission life-threatening disease; RG4	ABSL3 practices plus: • Entrance through change room where personal clothing is removed and laboratory clothing is put on; shower upon exiting • All wastes are decontaminated before removal from the facility	ABSL3 equipment plus: • Maximum containment equipment (e.g., Class III BSC or partial containment equipment in combination with full body, air-supplied, positive-pressure personnel suit) used for all procedures and activities	ABSL3 facility plus: • Separate building or isolated zone • Dedicated supply and exhaust, vacuum, and decontamination systems • Other requirements outlined in the text

Note: *Consult the BMBL 6th edition for a more complete description of the four (4) animal biosafety levels, as well as recommended containment requirements for biosafety specific type of animals.*

Chapter 4
Laboratory Biosafety Practices

The foundations of protective practices in a laboratory lie in an individual's laboratory experience, technical knowledge, personal work habits, and attitude toward laboratory safety. Unlike administrative controls, which are behaviors dictated by regulation or laboratory policy, the term "protective behavior" is used to define an innate part of each individual worker's personal approach to the laboratory environment. As such, "protective behaviors" form the first and most important line of defense against injury or exposure in the biomedical workplace.

Basic Laboratory Practices

Prudent practices and good techniques are of primary importance in laboratory safety. Both are based on sound technical knowledge, experience, common sense, and an attitude of courtesy and consideration for others.

Techniques and practices are spelled out in detail as "Standard Microbiological Practices" in the CDC/NIH's Biosafety in Microbiological and Biomedical Laboratories, 6th edition and the NIH's Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules, as well as in the National Research Council's Biosafety in the Laboratory - Prudent Practices for the Handling and Disposal of Infectious Materials (National Academy Press, Washington, D.C., 1989). At a minimum, the seven basic rules of biosafety, based on the National Research Council's Prudent Practices document, should be the basis of any personal laboratory work ethic; some are noted in Table 1.

Table 1: Biosafety practices and blocked routes exposure

Biosafety Practice	Routes of Exposure Blocked
1. Do not mouth pipette.	Inhalation, ingestion, skin, and mucous membrane contact
2. Handle infectious fluids carefully to avoid spills and the production of splatters or aerosols.	Inhalation, skin, and mucous membrane contact
3. Restrict use of needles, syringes, and other sharps to those procedures for which there are no alternatives; dispose of sharps immediately after use in leak- and puncture-proof containers.	Percutaneous, inhalation
4. Use lab coats, gloves, safety eyewear, and other personal protective equipment.	Skin and mucous membrane contact
5. Wash hands with disinfectant soap and running water after all laboratory activities, following the removal of gloves, and immediately following contact with infectious agents.	Skin and mucous membrane contact

6. Decontaminate work surfaces before and after use, and immediately after spills.	Skin and mucous membrane contact
7. Do not eat, drink, store food, smoke, or wear contact lenses in the laboratory.	Ingestion, skin, and mucous membrane contact

Laboratory Practice and Technique

The most important element of containment is the strict adherence to standard microbiological practices and techniques. Persons working with infectious agents or infected materials must be aware of potential hazards and be trained and proficient in the practices and techniques required for handling such material safely. The PI is responsible for ensuring that laboratory personnel are properly trained; the PI may delegate the provision of training to the laboratory supervisor or a designee, but the overall responsibility remains with the PI.

Each laboratory is expected to adopt this Biosafety Manual and develop written procedures if there are lab-specific hazards not addressed in the Manual or specific procedures needed in order to conduct the work safely. Personnel should be advised of special hazards and should be required to read and to follow the required practices and procedures. A scientist trained and knowledgeable in appropriate laboratory techniques, safety procedures, and hazards associated with the handling of infectious agents must direct laboratory activities.

When standard laboratory practices are not sufficient to control the hazard associated with a particular agent or laboratory procedure, additional measures may be needed. The PI is responsible for selecting additional safety practices, which must be appropriate to address the hazard associated with the agent or procedure. The BSO or EHS may be consulted for more information.

Laboratory personnel safety practices and techniques must be supplemented by appropriate facility design and engineering features, safety equipment, and management practices. Each laboratory will designate a person as the Laboratory Safety Coordinator (LSC).

Note: *Although each individual is responsible for their own safety, the PI has ultimate responsibility for ensuring that personnel working in the laboratory are adequately trained, follow the prescribed safety measures and wear appropriate PPE.*

Laboratory Housekeeping and Personal Hygiene
Personal safety is greatly enhanced by keeping workspaces neat, clean, and orderly. Injuries and exposures are more likely to occur in poorly maintained, disorderly areas.

If workspace is shared, the importance of maintaining a neat, clean area increases significantly. Individuals sharing workspace must understand their responsibility to keep the work area and equipment clean and properly maintained. Coworkers must rely on one another to maximize efficiency and safety. Personal materials should be properly labeled, waste discarded, and the shared space disinfected or cleaned prior to leaving for the next user.

The following guidelines should be observed in the laboratory:

- Routine housekeeping ensures work areas are free of sources of contamination and hazards;
- Establish and follow housekeeping procedures in order to maintain a clutter free and well organized work environment. Laboratory personnel are responsible for the routine cleaning of laboratory benches, equipment, and areas that are used during research work;
- Access to exits, sinks, eyewashes, emergency showers, and fire extinguishers must not be blocked;
- The workplace should be free of physical hazards;
- Follow electrical safety. Use of extension cords are temporary measures and placement must avoid heavily traffic areas. Equipment should be properly grounded. Overloaded electrical circuits and the creation of electrical hazards in wet areas are to be avoided;
- Work benches should be free of infrequently used chemicals, glassware, and equipment;
- Do not store unnecessary items on floors, under benches, or in corners;
- All compressed gas cylinders should be properly secured.

Personal hygiene, including proper handwashing techniques, is also a means by which to enhance personal protection in the laboratory. Washing of hands immediately after de-gloving ensures that contamination of the hand by glove micropuncture or prior exposure is cleaned.

The laboratory is also an inappropriate place to perform personal hygiene including cosmetic tasks, such as applying makeup, cleaning or trimming fingernails, or brushing hair including wearing or removal of contact lenses. These activities provide new opportunities for exposure and contribute to retrograde contamination of the laboratory environment.

Universal Precautions

Prudent practices often overlap with a set of practices known as “universal precautions.” The overarching universal precaution espoused by the Bloodborne Pathogens (BBP) Standard (see Appendix J for a list and more detailed discussion of universal precautions) should be adopted by all laboratory personnel. Universal precautions require that all human blood and tissues be handled as though they are infectious. Adopting and applying universal precautions to all laboratory reagents clearly creates a heightened awareness of potential risk and adds another level of caution to activities involving reagents.

Administrative Controls

Administrative controls are policies and procedures designed to assist with the safe handling of potentially hazardous biological materials. They include training, medical surveillance, vaccinations, access control, etc.

Biological Hazard Information

Laboratory workers must be knowledgeable about the hazards associated with the biological agents present in the laboratory and have hazard information available to them. The following are sources of hazard information for biological agents:

Microbial Agents
The CDC/NIH's *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) 6th edition has descriptions of biosafety levels and recommended biosafety practices for specific biological agents. The Canadian Laboratory Centre for Disease Control maintains Material Safety Data Sheets for microbial agents.

Toxins
Isolated biological toxins are chemical hazards, although many such toxins produce adverse effects at doses significantly below that of "traditional" laboratory chemicals. Laboratory use of isolated toxins falls under the BU Lab Safety Manual and Chemical Hygiene Plans, and Safety Data Sheets (SDSs) must be maintained and available to personnel. SDSs for a specific toxin should be obtained from the vendor upon receipt of the toxin. Toxicology textbooks, such as *Casarett & Doull's Toxicology,* are also good sources of hazard information for toxins. Biological toxins that are listed under the select agent category and are over the permitted quantities are regulated by CDC/APHIS. The IBC also requires laboratories to register these toxins with their office regardless of the amount.

Each laboratory is expected to adopt this Biosafety Manual, the CDC/NIH publications *Biosafety in Microbiological and Biomedical Laboratories, 6th edition*, and *Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules*. However, because these publications cover relatively general topics, individual laboratories should develop written procedures if there are lab-specific hazards not addressed in the Manual or CDC/NIH documents that cover the biosafety concerns and laboratory procedures for that particular laboratory.

For example, laboratory-specific SOPs should address safe manipulation of specific organisms, specific exposure control methods, and specific decontamination and waste-handling requirements. EHS provides a recommended standard format for SOPs, and laboratories are encouraged to use this format to effectively convey the biosafety information (including use of pictures and illustrations). The laboratory-specific SOPs do not need to duplicate the more general SOPs contained in this manual or the CDC/NIH documents, but should serve as supplements. For an SOP template, please contact EHS.

Security and Inventory of Biological Agents

Each PI must develop site-specific criteria that safeguard all biological materials, regardless of their risk group, from unauthorized removal. It is the PI's responsibility to ensure their laboratory implements sufficient security measures and procedures to prevent unauthorized access to biological agents.

Select agents (*See Chapter 9*) and other higher-risk microorganisms and toxins must be stored in a locked container, and a detailed inventory must be maintained per CDC requirements. In many instances, during the application review process, the IBC will review the proposed acceptable safeguards and either approve or recommend enhancements to the proposed plans.

Prevention of Aerosols and Droplets

Handling of liquids or dry powders may generate aerosols or droplets. In practice, high-energy procedures, such as centrifuging, vortexing, and mixing, tend to produce respirable aerosols that stay airborne for extended periods and are small enough to be inhaled, while low-energy procedures,

including opening containers and streaking plates, produce droplets that settle quickly on surfaces, skin, and mucous membranes.

Utilization of Biological Safety Cabinets

In general, the following guidelines are recommended when using biological safety cabinets (BSCs):

- The BSC should be tested and certified when it is initially installed and recertified after it is moved to new place, and annually thereafter. For information on cabinet certification, call 617-358-3914 or email EHS at safety@bu.edu.
- The magnahelic gauge should be checked after the BSC is turned ON before it is used. This gauge will normally run at a relatively fixed value. When it deviates significantly, the cabinet should not be used until the cause of the deviation has been identified and fixed.
- Personnel should know how the BSC works and perform work in it appropriately.
- Personnel should be familiar with the safe and effective use of any UV lamps inside the BSC (if equipped) and use appropriate precautions to avoid any UV exposure. **Note***: EHS has discouraged the use of UV lamps because they have a short half-life and are difficult to maintain.*
- The BSC's front laminar airflow curtain should not be disrupted. The user should avoid quick arm movement when working in the cabinet, and no heavy foot-traffic in front of the BSC while in use. Also avoid opening and closing nearby laboratory doors, as those activities may disrupt the airflow pattern and reduce the cabinet's effectiveness.
- Preplanning of the work and organization of necessary materials minimizes the need to exit and reenter the BSC workspace.
- Avoid the accumulation of materials inside the BSC and keep only the materials needed to conduct the work to reduce the disruption of airflow.
- The BSC should be left running whenever the cabinet is in use.
- Use of only appropriate disinfectants to avoid damaging the cabinet's interior.
- Work surface should be disinfected with an appropriate chemical disinfectant. If 10% bleach solution is used, it must be followed by 70% ethanol to prevent surface damage from the bleach solution. Each item needed for the planned procedures should be wiped off with 70% ethanol and placed in the cabinet.
- After the work volume is set up, the BSC should run for at least five (5) minutes to allow for stabilization of air flow before starting any procedure.
- If equipment such as a centrifuge or blender could create air turbulence in the BSC, it should be placed in the back one-third of the cabinet. All other work should be stopped while this equipment is operating.
- Open flames should not be used because they create air flow turbulence that may compromise sterility. In addition, the heat buildup may damage the HEPA filters or cause fire. If possible, substitute disposable items such as loops as there is no need to sterilize. If a flame is necessary, a burner with a pilot light should be used. Electric devices, such as loop sterilizers, are often satisfactory alternatives to open flames.
- A pan with disinfectant and/or a sharps container should be placed inside the BSC for pipette/sharps disposal. Vertical pipette discard canisters on the floor outside the cabinet should be avoided.
- Contaminated and clean items should be segregated, and personnel should work from "clean to dirty." The biohazardous waste collection bag should be in a rigid container. Do not block air flow into the front and rear exhaust grilles.
- Move arms slowly when removing items from or introducing items into the cabinet work volume.

- Protect the facility vacuum system from biohazards by using dual aspirator flasks in series (A and B) and placing an in-line hydrophobic HEPA filter (C) between the vacuum trap and the source valve (D) in the cabinet. **Note***: EHS requires that the flasks are placed in a secondary container, such as a Nalgene tub*.
- All spills in the cabinet should be cleaned immediately. Upon clean up, the BSC should be allowed to filter and purge its air and work should not resume for ten (10) minutes.
- When work is complete, all materials and wastes should be removed from the BSC and all interior surfaces should be wiped with an appropriate disinfectant.
- Gloves must be removed before exiting the BSC, and after touching or handling contaminated materials. Discuss the appropriateness of other alternatives, such as use of double gloves, with the BSO before deploying as part of the protocol.
- Laboratory coats must be removed and hands thoroughly washed with disinfectant soap and running water before leaving the laboratory.

Utilization of Pipettes

Pipettes are used for volumetric measurements and the transfer of fluids that may contain infectious, toxic, corrosive, or radioactive agents. Laboratory-associated infections have occurred from oral aspiration of infectious materials, mouth transfer via a contaminated finger, touching face (eyes, nose, etc.) and inhalation of aerosols. Exposure to aerosols may occur when liquid from a pipette is dropped onto the work surface, when cultures are mixed by pipetting, or when the last drop of an inoculum is blown out.

The following outlines safe pipetting techniques to minimize the potential for exposure to hazardous materials:

- Do not mouth pipette. Always use a pipetting aid.
- If working with biohazardous or toxic fluid, confine pipetting operations to a biological safety cabinet.
- Always use cotton-plugged pipettes when pipetting biohazardous or toxic materials, even when safety pipetting aids are used.
- Do not prepare biohazardous materials by bubbling expiratory air through a liquid with a pipette.
- Do not forcibly expel biohazardous material out of a pipette.
- Never mix biohazardous or toxic material by suction and expulsion through a pipette.
- When pipetting, avoid accidental release of infectious droplets.
- Use “to deliver” pipettes rather than “to contain” pipettes, which require “blowout.” Be careful not to dislodge the residual liquid.
- Do not discharge material from a pipette at a height. Whenever possible, allow the discharge to run down the container wall.
- Place contaminated, reusable pipettes horizontally in a pan containing enough liquid disinfectant to completely cover them. Autoclave the pan and pipettes as a unit before processing them for reuse.
- Discard contaminated, broken, or intact Pasteur pipettes and broken glass in a sharps container. Dispose of the container properly when it is, at most, three-fourths full.
- Pans or sharps containers for contaminated pipettes should be placed inside the BSC, if possible.
- Proper procedures for disposal of plastic pipettes are presented in Chapter 8**.**

Utilization of Centrifugation

Hazards associated with centrifuging include mechanical failure and the creation of aerosols. To minimize the risk of mechanical failure, centrifuges must be maintained and used according to the manufacturer's instructions. Users should be properly trained and operating instructions that include safety precautions should be prominently posted on or near the unit.

Aerosols are created by activities such as filling centrifuge tubes, removing plugs or caps from tubes after centrifugation, removing supernatant, and re-suspending sediment pellets. A significant aerosol hazard can be created if a tube breaks during centrifugation.

To minimize the generation of aerosols when centrifuging biohazardous material, the following procedures are recommended:

- Use sealed tubes and safety buckets that seal with O-rings. Before use, inspect tubes, O-rings, and buckets for cracks, chips, erosions, bits of broken glass, etc. Do not use aluminum foil to cap centrifuge tubes because it may detach or rupture during centrifugation;
- Only use appropriate centrifuge tubes. Open and fill the centrifuge tubes, rotors, and accessories in a biological safety cabinet. Avoid overfilling centrifuge tubes to prevent closures from becoming wet. After tubes are filled and sealed, wipe them down with disinfectant;
- In the event of a tube breakage during centrifugation or an abnormal stoppage that could result in a spill in the centrifuge bucket, the bucket should be brought into the BSC and opened for inspection of potential breakage or spills. The bucket containing the spilled sample should be decontaminated inside the BSC and debris removed prior to reuse;
- Always follow the manufacturer's instructions when operating the centrifuge and balance the buckets, tubes, and rotors properly before centrifugation;
- Avoid decanting or pouring off supernatant; unless the supernatant must be retained, use a vacuum aspirator with appropriate in-line reservoirs and filters;
- Work in a BSC when re-suspending pelleted material. Use a swirling rotary motion rather than shaking. If shaking is necessary, wait a few minutes to permit the aerosol to settle before opening the tube;
- Small, low-speed centrifuges may be placed in the BSC during use to reduce the aerosol escape. High-speed centrifuges pose additional hazards. Precautions should be taken to filter the exhaust air from vacuum lines, to avoid metal fatigue resulting in disintegration of rotors, and to use proper cleaning techniques and centrifuge components. Manufacturers' recommendations must be meticulously followed to avoid metal fatigue, distortion, and corrosion;
- Avoid the use of celluloid (cellulose nitrate) tubes with biohazardous materials. Celluloid centrifuge tubes are highly flammable and prone to shrinkage with age. They distort on boiling and can be highly explosive in an autoclave. If celluloid tubes must be used, an appropriate chemical disinfectant must be used.

Utilization of Cryostats

Use of cryostats is very common in many research laboratories. Anyone using the device must be properly trained, know and have demonstrated that they can operate the device safely before allowing independent use. These devices may pose potential hazards associated with sharp cutting edges and cold environments and should be handled with extra care.

The following guidelines should be followed when using cryostats:

- Frozen sections of unfixed human tissue or animal tissue infected with an etiologic agent pose a risk because freezing tissue does not necessarily inactivate infectious agents. Use of freezing propellants under pressure is not recommended with frozen sections because they may cause spattering of droplets of potentially infectious material.
- Appropriate gloves should be worn during preparation of frozen sections.
- When working with human or infected animal tissue, consider the contents of the cryostat to be contaminated and decontaminate it frequently with 70% alcohol.
- Ribbons and trimmings from the frozen tissue samples can accumulate in the cryostat during its use. The machine should be cleaned up and decontaminated routinely.
- Defrost and decontaminate the cryostat with a tuberculocidal hospital disinfectant once a week and immediately after use with tissue known to contain bloodborne pathogens, *M. tuberculosis,* or other infectious agents.
- Use disposable knife blades. Handle microtome knives or blades with extreme care. Cut-resistant gloves with stainless steel mesh or similar gloves should be worn when changing knife blades.
- Solutions used for staining potentially infected frozen sections should be considered contaminated.
- Always use the safety lock of the equipment before mounting or unmounting tissues or replacing the blade.

Utilization of Inoculating Loops

Flaming inoculating loops can result in spatter and the release of aerosols and droplets. Use of an electric micro-incinerator or disposable loops are the preferred alternative to minimize this issue.

Use of Absorbent Materials

Work surfaces should be covered with absorbent pads or "diaper" sheets to absorb splashes and drips and to minimize the spread of contamination. The absorbent paper should be changed at the end of the laboratory procedure as part of the final cleanup, or at least daily during use.

Utilization of Miscellaneous Aerosol-Producing Devices and Activities

Use of any of the devices listed below results in considerable aerosol production. Blending, cell-disrupting, and grinding equipment should be used in a BSC when working with biohazardous materials.

Blenders

Safety blenders, are designed to prevent leakage from the bottom of the blender jar. They provide a cooling jacket to avoid biological inactivation and can withstand sterilization by autoclaving. Some important guidelines:

- Always place the blender on a stable surface and make sure that the lid is secure before operating the blender. Ensure use of appropriate PPE and never place hands or utensils inside when it's operating.
- Safety blender rotors used for infectious materials should be leak-proof. They should initially be tested with sterile saline or dye solution prior to use with any biohazardous material.
- Always inspect the power cord for cracks or damage. DO NOT operate the blender if there is damage on the power cord.
- The use of glass blender jars is not recommended because of the potential for breakage. If they must be used, glass jars should be coated with a polypropylene to prevent spraying of glass and contents in the event the blender jar breaks. The blender must be operated within a secondary containment basin.

- DO NOT overfill the blender. Overfilled blenders can lead to potential spills. A towel moistened with disinfectant should be placed over the top of the blender during use.
- When opening blenders, be cognizant of potential contamination hazards in the form of droplets that might become airborne or fall on the surfaces, liquid residue on the cap, and possible expansion of the volume due to aeration.
- Before opening the blender jar, allow the unit to rest for at least one minute to allow the aerosol to settle.
- Placing the blender in a BSC will provide protection against airborne hazards and placement of a tray lined with absorbent pads would assist with contamination control.
- The device should be decontaminated promptly after use.

Lyophilizers
Depending on type and model, potential aerosol production may occur when material is loaded into or removed from the lyophilizer unit.

- If possible, sample material should be loaded in a BSC.
- The vacuum pump exhaust should be equipped with a filter to filter out any hazardous agents or, alternatively, the pump can be vented into a BSC.
- After lyophilization is complete, all surfaces of the unit that have been exposed to the agent should be disinfected.
- If the lyophilizer is equipped with a removable chamber, it should be closed-off and moved to a BSC for unloading and decontamination.
- Handling of cultures should be minimized and vapor traps should be used wherever possible.

Sonicators
Sonication is the use of sound-wave energy for dispersion, disruption, or inactivation of biological materials, such as viruses. Sonicators generate sound waves at very high frequencies (~20,000 + Hz range), which is outside normal hearing range. The following are hazards associated with sonicators:

- *Noise*: Although the 20,000-Hz frequency is outside normal hearing range, there are other sources of noise, such as vibration from any loose equipment or other items on the bench or the liquid itself. If the noise levels are high, hearing protection devices should be worn.
- *Aerosols*: Aerosols present a more serious potential hazard and must be taken into consideration. Precautions listed for blenders and lyophilizers should be observed.

Ampoules
Glass ampoules have been widely used in packaging cultured materials. Opening ampoules containing liquid or lyophilized cultured material should be performed in a BSC to control any aerosol produced. Sealed-glass ampoules used to store biohazardous material in liquid nitrogen have exploded, causing eye injuries from glass microparticles in the snap of the ampoule. The use of polypropylene tubes (cryovials) eliminates this hazard. These tubes are available dust-free or pre-sterilized and are fitted with polyethylene caps with silicone washers. Heat-sealable polypropylene tubes are also available.

- Individuals should be trained on the safe handle and opening of glass ampoules.
- Gloves and protective eyewear must be worn when opening ampoules or cryovials.
- To open a sealed-glass ampoule, nick the neck of the ampoule with a file, wrap it in disinfectant-soaked disposable towel, hold the ampoule upright, and snap it open at the nick.

- Reconstitute the contents of the ampoule by adding liquid slowly to avoid aerosolization of the dried material.
- Mix the contents without bubbling and withdraw it into a fresh container. Discard the disposable towel and the ampoule's top and bottom as medical waste.

Loop Sterilizers and Bunsen Burners
Sterilization of inoculating loops or needles in an open flame generates small-particle aerosols that may contain viable microorganisms.

- Alternatively, disposable plastic loops and needles may be used for culture work where electric incinerators or gas flames are not available.
- Continuous flame gas burners should not be used in a BSC. These burners can produce turbulence that disturbs the cabinet's protective airflow patterns. Additionally, the heat produced by the continuous flame may damage the HEPA filter. If a gas burner must be used, one with a pilot light should be selected. Electric sterilizers should also be considered.

Personal Protective Equipment (PPE)

Personal protective equipment (PPE) must be provided without cost to personnel. Although not a substitute for the use of BSCs and good laboratory practices, PPE is considered a primary barrier to infectious agents and proper use will reduce the likelihood of infection. PPE is the least-desirable exposure control method because its failure results in direct exposure to the agent.

PPE is most effective when used to supplement primary control methods such as biological safety cabinets, safety centrifuge cups, and other containment devices. Appropriate clothing may also protect the experiment from contamination

The following PPE requirements apply to all individuals visiting research and teaching laboratories:
- Full-length pants or equivalent must be worn at all times;
- Closed-toe footwear that covers the top of the foot must be worn at all times;
- Long sleeves or laboratory coat covering both arms.

All other individuals entering laboratory space must follow the PPE requirements described above for the visitor, as well as the following:
- Laboratory coats must be worn over personal clothing at all times;
- Long hair and facial hair must be secured or tied back;
- Flame resistant laboratory coats must be worn when working with pyrophoric chemicals, water reactive chemicals, and high volumes of flammable chemicals;
- Disposable gloves that are protective against the hazardous or potentially hazardous materials being used must be worn. Gloves must be replaced when soiled, contaminated or damaged;
- Eye protection must be available and used when danger from splashing of hazardous or potentially hazardous materials could occur.

Respirators

Respirators are selected based on the hazard involved and the protection factor required. Certain laboratory and clinical situations require respiratory protection to prevent inhalation of infectious

agents. Regulations, as well as good safety practice, require that personnel be medically evaluated, specifically trained, and fit tested **prior** to wearing respiratory protective equipment. **Note:** ***Use of respirators requires completion of the OSHA Respiratory Questionnaire for medical clearance from the ROHP, and fit testing by EHS.***

Contact EHS if respiratory protective equipment is required or if there are questions about the respiratory protection program.

Further guidance on the use of PPE can be found in the Personal Protection Equipment in Laboratories Policy and Chemical Hygiene Plan.

Storage and Labeling of Biological Agents

Biological agents must be stored using leak proof and sealed containers. Containers must be clearly labeled with the identity of the agent and should include the universal biohazard symbol (see below) as physical space on the container permits. At a minimum, secondary (or outside) containers must include the universal biohazard symbol (identity of contents is also desirable).

Freezers, refrigerators, and other storage areas must also be labeled with the biohazard symbol; exceptions to this policy will be considered on an individual basis by the IBC if pertaining to procedures in a proposed IBC protocol. Waste and contaminated equipment or other objects to be decontaminated must also be labeled with the biohazard symbol.

Universal Biohazard Symbol
The OSHA Bloodborne Pathogen Standard specifically requires that containers of human blood or other potentially infectious material (OPIM), contaminated waste, and refrigerators, freezers, and other storage containers used to store or transport blood or OPIM be labeled with the universal biohazard symbol (fluorescent orange or orange-red):

Biohazard Labels and Signs
Each laboratory must have a door sign at the entrance that provides safety information to visitors and service personnel. Room signs must contain designations for all laboratory hazards in use within the laboratory (carcinogens, acutely toxic agents, reproductive hazards, biohazards, radioactive materials, lasers, and magnetic fields). EHS will prepare the signs for each door in accordance with the requirements of NFPA 704.

Biohazard signs will be posted at the following:

- Entrances to laboratories that use agents classified as BSL2, BSL3 or BSL4.

- Entrances to animal rooms used for housing animals infected with ABSL2, ABSL3, or ABSL4 agents.

For examples of BU door signage, see Appendix P.

Certain other areas and pieces of equipment within a laboratory may also require signs. Refrigerators, freezers, cabinets, and other storage facilities require the biohazard symbol in the following situations: used to store infectious agents of Risk Group 2 or higher, human blood or blood products, unfixed tissues, cell or organ cultures, body fluids, or excreta. Large pieces of equipment for handling such materials (e.g., centrifuges, biological safety cabinets) must be similarly labeled.

Chapter 5
Laboratory Training

Training is a critical component of any integrated biological safety program. Training is intended to provide the understanding, technical knowledge, and tools that the trainee can use to improve his or her daily laboratory safety practices.

At a minimum, all personnel working with biological materials must have training in the following areas prior to the start of their experiments:

- Knowledge of this biosafety manual;
- Knowledge of hazards of the agent or sample being used:
- Experimental procedures to be performed;
- Safe use of laboratory equipment:
- Proper use of personal protective equipment:
- Procedures to follow in the event of an exposure incident;
- Decontamination and spill clean-up procedures;
- Safe handling methods for any infectious agent and/or recombinant DNA (rDNA);
- Proper methods for transporting infectious agents and other biohazardous materials;
- Proper waste handling, storage, and disposal;
- Procedures to follow and reporting after an injury;
- Bloodborne Pathogens Standard (if working with human blood or blood products, unfixed tissue, body fluids, organ, or primary tissue and/or samples contaminated with bloodborne pathogens);
- BPHC requirements (see Appendix Q for a summary); and
- Other specialized training as deemed appropriate by the IBC or the BSO.

PIs are responsible for ensuring that their employees receive proper training in the biohazards and controls specific to their laboratory and for the safe conduct of the experimental procedures to be used. EHS develops, maintains, and provides different types of training associated with the BU biological, chemical, and radiological safety programs. Each of these has its own driver and emphasis.

ROHP is also available as a resource to the IBC, PI's and the BSO to complement required training in the areas of biosafety and specific agents.

Biosafety Training

All research laboratory workers (e.g., faculty, staff, students, and visiting scientists) must complete the necessary biosafety training prior to engaging in any laboratory activities. EHS administers general online training on Laboratory Safety, Chemical Safety, BSL1 and 2, Bloodborne Pathogen, Shipping of Biological Samples for laboratory workers through SciSure (formerly known as SciShield). Each training module is completed based on the job function designated by the PI for each worker.

The PI is responsible for training workers on specific work procedures and how to perform the tasks safely, use of appropriate PPE, safety equipment, and engineering controls such as biosafety cabinets or chemical fume hoods. The PI, supervisor, or laboratory manager document and track training of laboratory personnel. This training is mandated and must be provided by the PI or laboratory manager

on a periodic basis to all laboratory personnel. Documentation of the training is required and must include, at minimum, the date and duration of training, name and position of the trainer, topics covered, and names of the trainees. Additional training may be required by law or BU policy, and in some cases, completion of such training must be documented with the BSO or EHS.

Training and knowledge of the NIH Guidelines and IBC policies
All PIs listed on an active IBC protocol and all individuals listed on an active IBC protocol involving recombinant or synthetic nucleic acids (even if they are not current working with these agents) are required to complete the online "IBC Policy / Recombinant DNA Training" and quiz before engaging in any protocol activities and at the time of three-year renewal before continuing to engage in the protocol activities.

Mandated Specific Training
Mandated specific training may also be required by law or BU policy. In some cases, it is administered and tracked by the BSO or EHS, who must maintain the training records. Examples of mandated specific training include, but are not limited to: non-human primate users training; agent specific trainings or other specific training required by the IBC or BPHC. Individuals working in laboratories classified as BSL3 and above, or who are potentially exposed to specific zoonotic diseases, must also undergo training.

HIV/HBV Laboratory Training
Personnel who work in research laboratories that culture, produce, or otherwise perform microbiological manipulation of human immunodeficiency virus (HIV) or hepatitis B virus (HBV) must receive additional training beyond the standard bloodborne pathogen training. The PI is responsible for ensuring that this training is provided and completed prior to working with HIV or HBV. Laboratory workers must demonstrate proficiency in standard microbiological techniques, and in the practices and techniques specific to the laboratory. Additionally, workers must have prior experience in handling human pathogens before working with HIV or HBV.

Personnel who do not have experience with human pathogens must be trained in the laboratory before working with HIV or HBV. Initial training must not include the use of infectious agents; rather, training and work activities should be progressive as proper techniques are demonstrated. Workers are permitted to handle infectious agents only after demonstrating proficiency to the laboratory supervisor's satisfaction.

Packaging and Shipping of Infectious Agents Training
Personnel who package and ship biological samples, infectious agents and diagnostic specimens such as microorganisms, blood samples, and clinical samples for pathological testing are required by federal and international regulations to receive training every two years. EHS offers this training monthly, online and upon request. The training is valid for two (2) years and must be retaken prior to the expiration date for persons responsible for shipping of these materials.

In-person training dates and locations can be found in SciSure (formerly known as SciShield). under "Classroom Shipping Biological training". Online training is also available in SciSure as "Shipping Biologicals".

Select Agents Training

Personnel authorized to use select agents are required to receive training that meets the specific requirements of 42 C.F.R. Part 73, 7 C.F.R. Part 331, and 9 C.F.R. Part 121. Training must be completed prior to any individual starting work with select agents and annually to continue working with them. EHS and the lab will maintain copies of the training records for reference. Contact the NEIDL BSO for more information.

Agent-Specific Training

Personnel authorized by the IBC to work with specific agents designated as Biological Agents with Potential to Cause a Laboratory Acquired Infection (LAI) are required to receive agent specific training assigned by their PI, supervisor or EHS ROHP is available to assist PIs on request with this training requirement and will provide a medical surveillance wallet card to be carried by those personnel, as well as access to Agent Information Sheets (AIS) with appropriate safety information.

The list of these agents is dynamic and is regularly reviewed by BU's LAI Committee for pathogens that researchers are proposing to use. As new agents approved by the IBC are introduced into the laboratory environment, they may be added to the list; agents that are no longer being used will be removed from the list.

Laboratory-Specific Training

Individual laboratories are required to develop specific training for the particular agents and procedures that personnel will perform in that laboratory. This training should be specific to the hazards in the laboratory and to each person's laboratory duties. Each person in the laboratory must understand the hazards associated with the agent and laboratory operations, how to prevent exposures to biological and chemical agents (see Chemical Hygiene Plan), and be trained on the laboratory standard operating procedures. Laboratory-specific training should not duplicate the general biosafety training, but instead should supplement it.

Laboratory safety training records are maintained by SciSure (formerly known as SciShield) for the courses offered on the system. If lab specific training needs to be documented use the Miscellaneous Training Roster on the Documents tab of the lab home page in SciSure and upload the training record with name, date and training objective. Ongoing training is required as new hazards and procedures are introduced into the laboratory.

Other Safety Training

Personnel who utilize other materials such as radioisotopes, controlled substances, or x-ray generating devices must complete additional laboratory safety trainings. These trainings can be found in the course directory in SciSure under Training.

Refresher Training

All laboratory workers and certain categories of building occupants will be subject to periodic mandatory refresher training. The scope and details of these refresher trainings is based on the risk of the agent and procedures performed and will range from annually to every three years based on regulatory requirements such as Bloodborne Pathogen Standard or Select Agents Rule. All laboratory personnel must also complete the annual Laboratory Safety Training.

Chapter 6
Decontamination and Sterilization

Definitions

Decontamination is a process or treatment that renders a device, instrument, or work surface safe to handle. A decontamination procedure can range from sterilization by autoclave or ethylene oxide to simple cleaning with soap and water. Sterilization, disinfection, and antisepsis are all forms of decontamination.

Sterilization is the use of a physical or chemical procedure to destroy all microbial life, including highly resistant bacterial endospores.

Disinfection eliminates virtually all pathogenic, non-spore-forming microorganisms but not necessarily all microbial forms on inanimate objects (work surfaces, equipment, etc.). Effectiveness is influenced by the types and extent of organisms, the amount of organic matter, and the surface type (i.e., porous or smooth) of the object to be disinfected and chemical exposure time, temperature, and concentration.

Antisepsis is the application of a liquid antimicrobial chemical to skin or living tissue to inhibit or destroy microorganisms. It includes using germicidal solutions for swabbing an injection site on a person or animal and for handwashing. Although some chemicals may be utilized as either a disinfectant or an antiseptic, adequacy for one application does not guarantee adequacy for another. Manufacturers' recommendations for appropriate use of germicides should always be followed.

General Procedures

Decontamination of cultures and objects contaminated by biological agents is routinely performed in microbiological laboratories. Decontamination is a vital component of microbiological safety practice and serves to protect laboratory personnel (as well as others) from contamination and potential infection and the release of infectious organisms to the outside environment (primarily through person-to-person transmission). Decontamination of media, work surfaces, and equipment is also necessary to prevent contamination of cultured organisms. The following measures should be taken:

- Infectious wastes such as contaminated liquid and solid will be handled, treated and disposed of according to biological waste policies and procedures. Liquid wastes such as bacterial or viral culture media from BSL2 labs will be treated with appropriate disinfectant prior to sink disposal. Solid wastes from the BSL2 laboratories will be segregated and placed in biohazard containers lined with biohazardous waste bags and disposed of as biological wastes. This waste is sealed by the laboratory and shipped off-site for sterilization (see Waste Chart posted in the laboratory for more information).
- A disinfectant should be chosen that is appropriate for the organism in use.
- All liquid biological cultures should be deactivated with appropriate disinfectant.
- All solid biological waste should be disposed of in the biohazard waste containers.

Methods of Decontamination

The three main categories of physical and chemical decontamination are heat, liquid disinfection, and vapors and gases.

Heat: Wet heat is the most dependable method of sterilization. Autoclaving (saturated steam under pressure of approximately 15 psi to achieve a chamber temperature of at least 250° F for a prescribed time) is the best method of rapidly achieving destruction of all forms of microbial life.

- In addition to proper temperature and time, prevention of entrapped air is critical to achieving sterility because of air's poor heat transfer properties.
- Material to be sterilized must come into contact with steam and heat. Indicators of proper autoclave operation (e.g., autoclave tape or autoclave-sensitive labels) must be used with each load to visually indicate successful processing.
- Use of autoclave tape alone is not an adequate monitor of the sterilization's success.
- The Massachusetts Department of Public Health Medical Waste Management Act has specific quality-control requirements for autoclaves used for sterilization of medical waste. Appendix D describes the procedures for such tests.

Liquid disinfection: A liquid disinfectant (e.g., 1:10 solution of household bleach yielding a final hypochlorite concentration of 0.5%) is used to wipe or soak potentially contaminated materials for a period of time to kill all pathogenic agents present. Each disinfectant requires varying amounts of contact time. When sink disposing of disinfected biological solutions, run water in the sink for at least 5 minutes in order to neutralize pH of the deactivated solution.

Gas and vapor: Potentially contaminated articles are exposed to a sterilizing gas (e.g., ethylene oxide, chlorine dioxide, or ETO) or vapors from a chemical (e.g., formaldehyde, vaporized hydrogen peroxide). Because of the hazardous nature of the gases and vapors used, this requires specially designed equipment and facilities.

Autoclaving

Autoclaving uses saturated steam under pressure (approximately 15 psi) to achieve a temperature in the autoclave of at least 121°C (250°F). Autoclaving can be used to destroy vegetative bacteria, bacterial spores, and viruses. When decontaminating biohazardous waste, it is recommended that the temperature **in the waste** reach a minimum of 115°C for a minimum of 20 minutes. The total processing time required to meet these conditions depends on several loading factors (see below), however, it is recommended that a minimum autoclave cycle of one hour be used when decontaminating waste. Please note that waste that has been designated by the EHS for autoclave treatment must be treated by autoclaving prior to disposal in biohazardous waste boxes and shipped off as regulated biohazardous wastes. The autoclaving process makes them safer for handling and transport, it does not change the disposal endpoint.

When using an autoclave, the following guidelines should be taken into consideration:

- Biohazardous materials should not be left inside the autoclaves overnight in anticipation of autoclaving the next day.
- Autoclaves should not be operated by untrained personnel.
- Special precautions should be taken to prevent accidental removal of material from an autoclave before it has been sterilized or the simultaneous opening of both doors on a double door autoclave.
- Always use the appropriate PPE when operating the autoclave including lab coat, disposable gloves, heat resistant gloves and face protection.

- Dry hypochlorite, or any other strong oxidizing material, must not be autoclaved with organic materials such as paper, cloth, or oil: **WARNING! OXIDIZER + ORGANIC MATERIAL + HEAT = POSSIBLE EXPLOSION**

Three factors in combination determine the effectiveness of autoclaving:

1. *Temperature:* an autoclave uses steam under a pressure of approximately 15 psi to achieve a chamber temperature of at least 121°C. Although the autoclave chamber may reach 121°C, this does not necessarily mean that the interior of the load will reach this temperature.
2. *Time:* a minimum autoclave cycle time of 20 minutes at a chamber temperature of 121°C (time does not begin as soon as the autoclave cycle is initiated) is commonly recommended for sterilization of clean items. However, the total processing time required to achieve decontamination depends on several loading factors, including the load container (heat transfer properties), the amount of water added to the load, and the weight of the load. For increased loads, an increased cycle time will be required to ensure effective decontamination.
3. *Contact:* steam saturation is essential for maximum heat transfer. Steam must contact all areas of the load. Autoclave bags and other containers should be left partially open (or otherwise permit entry of steam) to ensure adequate contact. Studies have shown that adding water to the interior of the bag improves the time-temperature profile of the autoclave cycle, thereby increasing the autoclave's sterilization efficiency.

Dry Heat

Dry heat method requires a higher temperature and longer contact time. It is less effective than moist heat (autoclaving). Nevertheless, dry heat is preferable to moist heat for decontamination of anhydrous materials and closed containers because the moisture component of the steam used in an autoclave will not effectively penetrate anhydrous materials and closed containers.

The highest dry heat equivalent temperature that these materials will reach in an autoclave is 121°C. The highest temperature that material will reach in a dry heat oven will be the actual temperature inside the oven. A temperature of 160°-180°C for three to four hours is recommended for decontamination of waste using a dry heat oven.

Chemical Disinfection

Disinfection is the decontamination of work surfaces, equipment, biological safety cabinets, and other inanimate objects using antimicrobial agents. Several chemical agents are used as disinfectants. Laboratory workers should remember that there are hazards associated with all of these chemical disinfectants.

- Inhalation and skin contact should be minimized, and contact with eyes avoided.
- Appropriate gloves and safety eyewear should always be worn when handling these chemicals.

Pertinent information for some of the common chemical disinfectants is summarized in table below:

Summary of Chemical Disinfectants

Disinfectant	Use Parameters	Effective Against[a]					Important Characteristics	Potential Application
		Vegetative cells	Lipophilic viruses	Tubercle bacilli	Hydrophillic viruses	Bacterial spores		
Alcohol (ethyl, isopropyl)	*conc.*: 70-85% *contact time*: 10-30 min.	+	+	+	±		Eye irritant, toxic, flammable, inactivated by organic matter.	Surfaces: work and equipment
Chlorine Compounds	*conc.*: 0.05-0.5% (commercial bleach 0.5%) *contact time*: 10-30 min.	+	+	+	+	±	May leave residue; corrosive; skin, eye and respiratory irritant; inactivated by organic matter; make up at least weekly.	Spills, equipment surfaces, instruments, glassware, water baths
Quaternary Ammonium Compounds	*conc.*: 0.1-2% *contact time*: 10-30 min.	+	+				Toxic, inactivated by organic matter.	Surfaces (work and equipment), BSCs, floor maintenance, glassware, instruments
Phenolic Compounds	*conc.*: 0.2-3% *contact time*: 10-30 min.	+	+	+	±		Leaves residue; corrosive; skin, eye and respiratory irritant; toxic; inactivated by organic matter.	Surfaces (work and equipment), BSCs, floors, spills, glassware, instruments, water baths
Iodophor Compounds	*conc.*: 0.47% *contact time*: 10-30 min.	+	+	+	±		Leaves residue; corrosive; skin and eye irritant; toxic; inactivated by organic matter.	Surfaces (work and equipment), BSCs, glassware, water baths

Disinfectant	Use Parameters	Effective Against[a]					Important Characteristics	Potential Application
		Vegetative cells	Lipophilic viruses	Tubercle bacilli	Hydrophillic viruses	Bacterial spores		
Formaldehyde[b] (Formalin)	*conc.*: 4-8% *contact time*: 10-30 min.	+	+	+	+	±	Leaves residue; skin, eye and respiratory irritant; toxic (carcinogen).	Less effective than other disinfectants but can be used for equipment surfaces, glassware, instruments
Glutaraldehyde	*conc.*: 2% *contact time*: 10-60 min.	+	+	+	+	+	Leaves residue; skin, eye and respiratory irritant; toxic.	Equipment surfaces, glassware, instruments

a:+ = very positive response, ± = less positive response. A blank denotes a negative response or not applicable.
b: due to its irritating characteristics and status as a carcinogen, formaldehyde should not be used without good local exhaust ventilation.

From *Laboratory Safety: Principles and Practices*, second edition, Diane O. Fleming, John H. Richardson, Jerry J. Tulis, and Donald Vesley, eds., American Society for Microbiology, Washington, D. C.

Chapter 7
Biohazardous Spill Response

Even with the most careful planning and implementation of a research project, the possibility of an incident or spill involving biological materials exists. The following procedures are intended to provide a planned response to such rare events.

In any spill scenario, the priority of actions is determined by the "**PEP**" rule - **P**eople, **E**nvironment and **P**roperty. The highest priority is to provide aid to injured personnel and prevent spill area access to others.

The following are the general requirements and guidelines for biohazardous spill response**:**

Preplanning for Biohazardous Spill Cleanup

All spills of biohazardous materials do not represent the same risk to personnel and the environment, making each spill somewhat unique. There are several factors that must be considered, including but not limited to: pathogenicity of the agent, route of exposure, volume of a spill, the quantity, and other aspects such as sharps.

The following should also be considered when conducting an assessment:

- Location (e.g., biohazard cabinet, countertop, floor, equipment);
- Nature (e.g., tip-over, aerosolizing (spray/splash), drop from a height);
- Toxicity/infectivity of the spilled material;
- Volatility and viscosity of the spilled material;
- Other properties of material (e.g., pH, normality, temperature);
- Nature of affected surfaces (e.g., absorbent, porous or smooth-pitted. Additional challenges (e.g. broken glass or sharps, clothing, mixing with other materials);
- Susceptibility of spilled material to neutralization/disinfection.

Pre-planning of spill response will lower the risk associated with cleaning up a spill and will increase the likelihood that the spill is handled appropriately. PIs or Laboratory Directors should prepare their laboratory for typical spill scenarios expected in the laboratory. Laboratory workers should be informed of the hazards of the biological agents used in the laboratory, the risk associated with these agents during spill scenarios, how to safely clean up the agents, and how to properly dispose of cleanup materials.

Spill Cleanup Materials

Each laboratory area should have spill cleanup materials available to respond to the largest spill anticipated for that area. At a minimum, the following spill cleanup materials should be available in the laboratory:

- Disposable gloves (thick, chemical-resistant gloves or double pair of thin, nitrile gloves are recommended);

- Safety goggles and masks or a full-face shield (strongly recommended to avoid splashes to the nose and mouth);
- Lab coat or smock to protect clothing and body;
- Absorbent pads;
- Paper towels;
- Disinfectant appropriate for the agents used in the laboratory;
- Forceps or other devices to pick up contaminated material (especially sharps);
- Sharps disposal container;
- Autoclavable biohazard bags; and
- Respirators as required.

The spill kits distributed by EHS to laboratories may not be adequate for the response to a biological spill. Additional items needed for the cleanup of biohazardous agents must be maintained in the laboratory.

Biohazardous Spill Cleanup Risk Assessment

Several factors must be considered when assessing the risk that a spill represents:

- Volume and concentration of the spilled material;
- The infectivity of the spilled material and routes of exposure;
- Location of the spill;
- Aerosolization potential of the agent resulting from the spill;
- Susceptibility of the spilled material to disinfection;
- Nature of the affected surface(s) and its ability to "hide" organisms from disinfection; and
- Immune status of immediate personnel.

As with any spill scenario (biological, chemical, or radiological), **the safety of personnel is the most important consideration**. Cleanup is to begin only after it is determined that the personnel who will clean up the spill have appropriate knowledge, training, and equipment.

Biological Material Spill Cleanup Procedures

The following are general biohazardous spill cleanup procedures that are appropriate for most spill scenarios; however, the appropriate response to any spill is based on an assessment of the risk associated with that particular situation.

If in doubt, immediately call the Medical Campus Control Center at (617) 358-4144 for the CRC EHS emergency telephone at (617) 353-2105. Both response lines are active 24/7/365.

Biohazardous Spills Inside Biological Safety Cabinets
If a biohazardous spill occurs inside a Biological Safety cabinet, the following procedures should be followed:

- Wear a laboratory coat (disposable is recommended when available), safety glasses, and gloves (appropriate for the biological agent and the chemical disinfectant) during cleanup.

- Allow the BSC to run continually during cleanup.
- Surround the affected spill area with absorbent material to prevent spread of the spill.
- Apply disinfectant appropriate for the biological agent and allow a minimum of 20 minutes of contact time (or as directed by manufacturer's instructions). Alcohol or other flammable liquids are not recommended.
- Wipe up the spill with a paper towels soaked with disinfectant.
- Wipe the BSC's walls and work surface, as well as any equipment in the cabinet, with a disinfectant-soaked paper towels.
- Place contaminated items in an appropriate container (biohazard waste bag, sharps container, or autoclavable pan with lid for reusable items) for autoclaving or disposal in biohazardous waste box.
- Allow items to have a minimum of 20 minutes of contact time with the disinfectant (or as directed by manufacturer's instructions) before removing them from the BSC.
- Remove disposable protective clothing, gloves and place in a biohazard waste bag.
- Thoroughly wash hands and forearms with soap and water.
- Allow BSC to run for a minimum of 10 minutes before resuming work in the cabinet or shutting off the cabinet.

Biohazardous Spills in the Laboratory, Outside the Biological Safety Cabinet

If a **BSL1 agent** or **less than 100 ml of a BSL2** agent is spilled, the following procedures should be followed:

- Remove any contaminated disposable protective clothing and place in a biohazard waste bag. Wash the hands and other areas affected by skin contact with soap and water.
- Wear a long-sleeved gown or lab coat (disposable is recommended), shoe covers, safety glasses (face shield also recommended), and gloves (appropriate for biological agent and disinfectant).
- Place absorbent pads over the spill (to absorb liquid), then place a second layer of disinfectant-soaked absorbent pads over the spill.
- Pour additional disinfectant around the spill, being careful to minimize any splatter or aerosolization, and work from the periphery toward the center, ensuring thorough contact between the spill and the disinfectant. Disinfect all items in the spill area.
- Allow a minimum of 20 minutes contact time (or as directed by manufacturer's directions) with the disinfectant.
- Wipe down all equipment, tools, etc., with disinfectant.
- Place contaminated items in an appropriate container (biohazard waste bag, sharps container, or autoclavable pan with lid for reusable items) for disposal as biohazardous waste or for autoclaving as necessary.
- Remove disposable protective clothing and place in a biohazard waste bag.
- Thoroughly wash hands, forearms, and face with soap and water. It is recommended that cleanup personnel shower as soon as possible.

In addition, the following procedures should be followed:

- Contact the BUMC Control Center at (617) 358-4144 or BUMC Public Safety (617) 358-4444 and notify EHS immediately for assistance with the cleanup.

- Remove any contaminated clothing and place in a biohazard waste bag for disposal or autoclaving as necessary and wash all areas affected by skin contact with soap and water.
- Wear a long-sleeved gown or lab coat (disposable recommended), shoe covers, safety glasses (face shield also recommended), and gloves (appropriate for biological agent and disinfectant).
- Place absorbent pads over the spill (to absorb liquid), then place a second layer of disinfectant-soaked absorbent pads over the spill.
- Pour additional disinfectant around the spill, being careful to minimize aerosolization, and work from the periphery toward the center, ensuring thorough contact between the spill and the disinfectant. Disinfect all items in the spill area.
- Allow a minimum of 20 minutes contact time (or as directed by manufacturer's directions) with the disinfectant.
- Wipe down all equipment, tools, etc., with disinfectant.
- Place contaminated items in an appropriate container (biohazard waste bag, sharps container, or autoclavable pan with lid for reusable items) for disposal as biohazardous waste or for autoclaving and necessary.
- Remove protective clothing and place in a biohazard waste bag for autoclaving.
- Thoroughly wash hands, forearms, and face with soap and water. It is recommended that cleanup personnel shower as soon as possible.

Biohazardous Spills Inside a Centrifuge

If a biohazardous spill occurs inside a centrifuge, the following procedures should be followed:

- Clear the area of all personnel and allow aerosol to settle (usually a minimum of 30 minutes) before re-entering the area.
- Wear a laboratory coat (disposable recommended), safety glasses, and gloves during cleanup.
- Transfer and open the centrifuge rotor and buckets in a BSC for cleanup.
- Using an appropriate disinfectant, thoroughly disinfect the inside of the centrifuge, the rotor, and buckets.
- Discard cleanup materials and protective clothing as biohazardous waste.
- Thoroughly wash hands, forearms, and other parts of the body with soap and water.

Biohazardous Spills Outside the Laboratory During Transport

When transporting biological materials between laboratories in the same campus, the samples must be contained in an unbreakable, leak-proof, primary container. The primary container must be placed inside a secondary container with absorbent material that is well-sealed and leak-proof container (see Chapter 11 for transportation guidelines). Both the primary and secondary containers must be labeled with the universal biohazard symbol with the identity of the agent. In the event a transport container drops and its contents are spilled, the following procedures should be followed:

- If necessary, clear the area of all personnel and secure the area.
- Cleanup should be initiated as soon as possible to prevent the release of potentially infectious aerosol. Attempt cleanup **only** if appropriate cleanup materials and protective clothing are available
- Notify the Medical Campus Control Center at 617-358-4144 or the CRC EHS emergency telephone at 617-353-2105. Both response lines are active 24/7/365.

Note: *Employees should become familiar with other non-spill emergencies, such as fire and medical emergencies. EHS has developed special emergency flip charts that are located in every lab and provide quick references to employees. Employees should review the charts so that in the event of an emergency, they are familiar with their location and content.*

Spill Response

When responding to a spill, the following rules should be followed:

- **Tend to the injured**: Ensure receipt of immediate medical care and do not attempt to move the injured individual(s) unless ambient conditions become life-threatening. Individuals splashed, sprayed with, or otherwise exposed to human blood or other body fluids or tissues during a spill will need to remove contaminated clothing and utilize basic first aid, washing any wounds immediately.
- **Await assistance**: Unless laboratory personnel are trained and properly supplied and equipped with appropriate personal protective equipment, **DO NOT** attempt to clean up the spill. Personnel should immediately call the Medical Campus Control Center at 617-358-4144 or the CRC EHS emergency telephone number, 617-353-2105. Both response lines are active 24/7/365.
- **Isolate the spill**: Evacuate the immediate spill area or the entire room in the case of an aerosolizing (splashing or spraying) spill or a spill of volatile material. Prevent others from entering the spill area with barricades or, if necessary, a sentry.
- **Contain the spill**: Place absorbent material around, on, or in the flow path of the spilled material *only if it can be done safely*.
- **Provide information**: Provide the information requested by the Control Center or EHS personnel and wait for the arrival of the emergency provider.
- **Clean up**: Clean up should take place **ONLY** if laboratory personnel are trained, properly supplied with personal protective equipment, and otherwise able to clean up and disinfect the spill safely.

Chapter 8
Biohazardous and Medical Waste Disposal

In the Commonwealth of Massachusetts, biohazardous waste is governed by the Department of Public Health regulation 105 CMR 480, "Storage and Disposal of Infectious or Physically Dangerous Medical or Biological Waste, State Sanitary Code Chapter VIII." Boston University has biological waste management guidance documents for each campus which are reviewed annually and posted on the EHS website. The information below is a brief summary of the instruction provided in the guidelines.

The regulation defines biohazardous waste as ***infectious or physically dangerous medical or biological waste*** that because of its characteristics may cause, or significantly contribute to, an increase in mortality or an increase in serious irreversible or incapacitating reversible illness; or pose a substantial present potential hazard to human health or the environment when improperly treated, stored, transported, disposed of, or otherwise managed.

The following types of waste are identified and defined as infectious or physically dangerous medical or biological waste, and shall be subject to the requirements of 105 CMR 480.000:

- **Blood and blood products**: Discarded bulk human blood and blood products in free draining, liquid state; body fluids contaminated with visible blood; and materials saturated with blood.
- **Pathological waste**: Human anatomical parts, organs, tissues, and body fluids removed and discarded during surgery or autopsy, or other medical procedures, and their containers.
- **Cultures and stocks of infectious agents and associated biologicals**: All discarded cultures and stocks of infectious agents and associated biologicals, biotechnological byproduct effluents, cultures of specimens from medical and pathological laboratories, cultures and stocks of infectious agents from research laboratories, wastes from the production of biologicals, and discarded live and attenuated vaccines intended for human use.
- **Contaminated animal carcasses, body parts and bedding**: The contaminated carcasses and body parts and bedding of all research animals known to be exposed to pathogens.
- **Sharps**: Discarded medical/research articles that may cause puncture or cuts, including but not limited to all, used and discarded hypodermic needles and syringes, Pasteur pipettes, broken medical glassware, scalpel blades, disposable razors, and suture needles.
- **Biotechnological byproduct effluents**: Any discarded preparations made from genetically altered living organisms and their products. Infectious or physically dangerous medical or biological waste shall be referred to as "Waste" in the subsequent provisions of 105 CMR 480.000.

Biohazardous Waste

Proper handling and disposal of biohazardous waste is necessary to prevent infection of personnel (laboratory workers, custodians, laboratory visitors, etc.) and release to the environment. OSHA and Commonwealth of Massachusetts regulations (105 CMR 480.000) require that biohazardous waste be properly labeled, stored, and disposed of.

Labeling Biohazardous Waste

At a minimum, all biohazardous waste must be labeled with the universal biohazard symbol and the word 'Biohazard'. Additional information, such as the type of waste (such as "sharps" or "liquid waste") and origin of the waste, is recommended.

Handling and Disposal of Biohazardous Waste

Sharps

Sharps include **all** syringes, lancets, scalpels, and other similar medical instruments (whether or not contaminated), as well as contaminated Pasteur pipettes and broken glass, and other instruments or materials that can cut or puncture personnel.

- Sharps must be collected in rigid containers that are leak-proof and resistant to puncture from the sharps. Sharps containers must be designed so that sharps can be safely introduced into the container but not easily retrieved.
- Containers must be red or orange in color and labeled with the universal biohazard symbol and the word 'Biohazard'. When the sharps container is approximately 3/4 full, personnel should seal the waste container and it will be picked up by the building facilities custodian or appropriate service personnel. CRC personnel should seal the waste container and fill out a pick-up request on the EHS website . Waste will be picked up by a waste management vendor that is contracted by EHS.

A licensed vendor retrieves the waste from each building at pre-determined intervals via SciSure (formerly known as SciShield) request on Charles River Campus, and Boston University Environmental Service (EVS) or BMC EVS removes biological waste on medical campus on pre-determined intervals or through call-in requests or work orders) to process the waste with an approved sterilization method.

Uncontaminated Laboratory Glassware and Broken Glass

Collect uncontaminated laboratory glassware and broken glass in rigid containers (separate from other waste) that will prevent cuts and punctures to personnel. Containers should be labeled "broken glass." Broken glass is to be disposed of as ordinary trash.

Solid Biohazardous Waste

Solid biohazardous waste includes cultures of microbiological agents, tissue culture, and contaminated material (such as petri dishes, pipettes, contaminated glass, etc.) These materials are collected in a cardboard box lined with two red bags with biohazard symbols. The cardboard box is labelled with the biohazard symbol.

- Personnel should close and seal the biohazard waste box with tape for pick up by the building facilities custodian or appropriate service personnel. Each box must be labelled with the building and room number (a Sharpie or similar permanent marker, if appropriate) on the top of the box to identify its origin.

- CRC personnel should seal the waste container and fill out a pick-up request on the CRC website. The building and room number should be written (a Sharpie or similar permanent marker is appropriate) on the top of the box to identify its origin.

A licensed vendor retrieves the waste from each building at pre-determined intervals to process the waste with an approved sterilization method. Medical Waste Tracking forms are maintained by EHS.

Liquid Biohazardous Waste

Liquid biohazardous waste includes all blood and liquid waste from humans or animals, and all other liquid biohazardous waste (such as microbial cultures). Collect liquid waste in closeable, rigid, plastic, leak-proof containers labeled with the universal biohazard symbol and the word "Biohazard".

- Human and animal blood and body fluids can be disposed of by flushing directly to the sanitary sewer (wear laboratory coat, safety glasses and face shield, and gloves, and be careful to minimize splashing).
- All other liquid waste must be autoclaved or treated with a disinfectant prior to sink disposal.
- Liquid waste treated with small quantities of bleach or other household disinfectants can be disposed of by flushing directly to the sanitary sewer after sufficient contact time. Liquid waste treated with other (wescodyne, for example) chemical disinfectants must not be disposed in the sink and must be collected and disposed of as hazardous chemical waste through EHS.

Animal Carcasses, Body Parts, Tissue and Bedding

All animal carcasses and parts, regardless of whether they have been experimentally infected or not, are disposed of as pathological waste.

- Animal carcasses and parts should be stored in a freezer or cold storage area prior to disposal. Ensure to secure any bones, **limbs and sharp parts from puncturing the bag and protruding to prevent injuries.**
- Animal tissues and animal bedding must be disposed of as pathological waste if the source animal was infected with a BSL2 agent or higher.

All animal wastes that are collected as biohazardous must be sent for incineration via a licensed vendor. who retrieves the waste from each building at pre-determined intervals. A yellow 'pathological' or 'incinerate only' sticker must be affixed to the closed biohazardous waste box if it contains animal carcasses, parts, tissues or bedding or a pre-printed "incinerate" box must be used. Medical Waste Tracking disposal forms are maintained by EHS.

Chapter 9
Federal Select Agent Program

The U.S. Department of Health and Human Services (DHHS) and the U.S. Department of Agriculture's (USDA) regulations require institutions that possess, use, or transfer certain biological agents and toxins (known as "select agents") be registered and approved by the Federal Select Agent Program (FSAP). The FSAP have identified specific biological agents and toxins they consider to be a severe threat to public health and safety because of their potential use as bioterrorism agents. These materials are referred to as "Select Agents". Their transfer, possession, use, and disposal are strictly regulated. On December 4, 2012, the Centers for Disease Control (CDC) and Animal and Plant Health Inspection Service (APHIS) (now the FSAP) implemented the amendment to the regulation and identified certain select agents and toxins as Tier I. These Tier I agents have additional requirements on security, biosafety and occupational health.

The current list of select agents is provided at the end of this chapter. Because the list of select agents may be revised, it is recommended that the Federal Select Agent Program website be checked before acquiring pathogenic agents and biological toxins.

The regulations associated with select agents are very complex and strict, and significant monetary fines and criminal penalties are associated with non-compliance. The information in this chapter is a summary of the select agent regulations; it is not a complete description of the regulatory requirements. **Investigators must review and understand the select agent regulations and their responsibilities prior to acquiring or working with any select agent** (for regulatory information, see the CDC website).

Responsible Official and Authorization

Responsible Official (RO)

Select agent regulations require that a RO be designated for each institution that possesses and uses select agents. **The RO has institutional responsibility for the biosafety, security, and regulatory compliance of select agents, and as such, must be contacted *prior* to obtaining any select agents.**

Authorization to Possess and Use Select Agents

Prior to personnel obtaining any select agent, the IBC must review and approve any proposed work. In addition, PIs who want to acquire, possess, or use any biological agent or toxin listed as a select agent **must be registered and approved with the FSAP *prior* to obtaining the agent(s) or toxin(s)**.

The FSAP must approve both the institution and the individual laboratory. Investigators who want to possess and use select agents must contact the RO for assistance with the registration application process. Approval by the FSAP can take several months, and PIs should plan research projects accordingly.

Exemptions and Exclusions

Diagnostic labs that do not maintain select agents are largely excluded from the Federal select agent regulations; however, notification and possession time limits and other requirements do apply. Additionally, the FSAP can grant exclusions for temporary public health emergency situations and other

special circumstances. Consequently, any laboratory that conducts diagnostic or verification testing for any select agent must identify itself by contacting the RO as soon as possible. **Identification of any select agent in a specimen or isolate must be reported to the RO immediately. The RO will assist the diagnostic laboratory with reporting to FSAP immediately after identification. Laboratories must fill out and submit "Report of the Identification of a Select Agents or Toxin" (Form 4) within 7 days of the identification; Form 4 can be found on the FSAP website.**

Several specific select agent microbial strains and toxin forms have been determined not to present a severe threat to public health and safety and are therefore excluded from the Federal select agent regulations. The list of excluded biological agents and toxins is dynamic and the most current list is available on the FSAP select agent website. Permissible amounts of the listed biological toxins do not fall under the regulation as long as they meet the threshold quantities. **Laboratories maintaining these exempt quantities of select agent toxins must register their toxins with the IBC via an IBC protocol, and keep an accurate inventory of toxin amounts to verify that total quantities are below the threshold.**

Security, Incident response and Biocontainment requirements

Each select agent laboratory must develop and implement three written plans: security; incident response; and biosafety; that address safety issues associated with that specific select agent.

Security Requirements

Each select agent laboratory must have a written security plan that addresses the following topics:

- Site-specific security risk assessment
- Information system and physical security control
- Storage control and inventory audits of select agents
- Select agent shipping and transfers
- Roles and responsibilities and training
- Reporting of unauthorized persons, suspicious activities and missing materials
- Access control and escort provisions for cleaning, maintenance, and repairs

Any theft or loss of select agents must be immediately reported to the RO, BU Police and EHS, which will notify the FSAP. In addition, the BPHC will be notified of such events. BU in collaboration with City Agencies (BPHC, Boston Fire Department, Boston Police Department and EMS) has developed an Emergency Notification and After-Action Guideline High Risk Materials Matrix outlining the list of agencies and events for which they will be notified. This plan must be available and up-to-date during a CDC inspection.

The Department of Justice must approve all persons who will have access to any select agent. Approval requires that each individual successfully pass a background security check (conducted by the FBI in accordance with the USA Patriot Act) and submit fingerprints to the FBI. Anyone who has not been approved for access to select agents must be denied access unless escorted by an approved person. Everyone who enters a laboratory where select agents are accessible must have security approval or be accompanied by an approved person. This includes visiting scientists (on or off-campus), maintenance workers, custodians, and vendors.

Please see the CDC/USDA Security Plan Guidance document for more information.

Incident Response

Each laboratory that possesses or uses select agents must develop a written emergency plan that is laboratory specific and coordinated with department, building, and Institutional emergency plans. The plan must address the following:

- Loss, theft, or release of a select agent or toxin
- Inventory discrepancies
- Security breaches (including information systems access controls to select agents and toxins)
- Severe weather and other natural disasters
- Workplace violence
- Bomb threats and suspicious packages
- Emergencies such as fire, gas leak, explosion, or power outage
- Planning and coordination with emergency responders
- Building evacuation, site security, and control
- Decontamination and emergency medical treatment, and other emergency response issues

Any exposure or potential exposure should be reported immediately, and the BU Incident Response Plan for Select Agent Laboratories and the Biological Spill Response Plan for Select Agent Laboratories implemented.

Affected personnel should immediately contact the ROHP for medical evaluation and treatment by calling (617) 358-7647 (ROHP).

Please see the CDC/USDA Incident Response Plan document for more information.

Laboratory personnel should become familiar with these plans.

Biocontainment

All persons approved for access to select agents must receive documented training covering the following:

- Hazardous characteristics of select agents and their safe handling, use, and disposal
- Safeguards for protecting against exposure to select agents, requirements and procedures
- Biological safety and personal protective equipment (PPE) requirements
- Disinfection, decontamination or destruction of select agents
- Handling of select agents in shared spaces

For more information on biocontainment, please see the CDC and USDA's Select Agents and Toxins Biosafety/Biocontainment Plan Guidance

Training of Personnel

Training in all aspects of delineated in the Security, Incident Response and Biosafety Plans is required before beginning work with select agents and annually thereafter.

Transfers of Select Agents

Select agents can only be transferred to, or between, entities that are currently approved by the FSAP to possess and use select agents. All transfers of select agents require *prior* approval of the FSAP.

Both the sender and recipient must complete a common transfer form (Form 2), and the recipient must submit the form 2 request to the FSAP for approval. Form 2 requires the signature of the RO at both the sender and recipient facilities. When the select agent is consumed or destroyed, the recipient must notify FSAP through an update of their registration.

Procedures for Select Agent Procurement and Receipt

For the transportation of Select Agents, Boston University follows the regulations of international, federal, state, and local authorities including the Department of Transportation, the FSAP/CDC, International Air Transportation Authority, and World Health Organization to ensure pathogens are safely shipped to and from the labs. The general requirements are issued by the US Department of Transportation, which sets down strict requirements for packaging, labeling, and documentation of the materials, and requires training for employees involved in shipping.

All transportation of Select Agents will involve Department of Transportation (DOT) compliant triple packaging and will be placed in a non-crushable, liquid-tight, solid container for an added layer of safety. These packages will be transported via exclusive-use vehicles and will be secured in the transport vehicle away from potential impact on outer walls. In addition, shippers will adhere to pre-determined travel routes and strictly defined schedules for pick-ups and deliveries, and both package and vehicle will be monitored by BU and local emergency responders using GPS.

The FSAP/CDC also regulates the shipment of Select Agents. Qualified carriers must meet all federal, state and local regulations to transport select agent materials. There will be notifications to the Boston Public Health Commission, Boston Police and Fire Departments, and Boston Emergency Medical Services in advance of shipments and GPS monitoring of the vehicle and package. The transportation of Select Agents is tightly controlled, and BU has worked closely with city, state, and federal authorities to ensure that the transportation of select agents complies with all regulations.

For more information about select agent transportation, see “Transportation of Select Agents” in Chapter 10.

Inventory and Disposal of Select Agents

An accurate record of all select agents, from receipt to destruction or disposal, must be maintained. The inventory must include specific information on individual containers and vials, as well as a record of each use, and ultimate disposal. The select agent inventory must be verified twice a year to account for all quantities and containers of select agents. Any discrepancies between the inventory record and the actual inventory must be reported immediately to the RO.

For more information, please see Guidance on the Inventory of Select Agents and Toxins.

Records Required for Select Agents

Select agent regulations require that several records be maintained, including the following:

- Biocontainment certifications
- Laboratory notebooks
- Institutional Biosafety and Animal Use Committees minutes
- Records associated with occupational health and suitability programs
- Training records
- Transfer documents (Form 2)
- Safety and security incident reports

Each laboratory/institution is responsible for maintaining these records. The recordkeeping requirements are complex, and therefore the FSAP regulations should be reviewed for a complete description of the recordkeeping requirements.

Recordkeeping for Select Agent Laboratories

The following is a list of required recordkeeping for all select agent inventory and access records, per FSAP regulations.

Inventory Records

The logbook Inventory must contain the following elements:

- Agent name, designation
- Date of creation, quantity acquired, source
- Storage location (e.g., building, room, and freezer)
- Name of the PI
- Quantity, volume used, purpose of use, destruction or disposal date
- Transfers: quantity, date, recipient PI
- Lost, unaccounted for, stolen
- Written explanation of any discrepancy

Access Records

Access Records must contain the following elements:

- Access to SA: Name, SA, date. Access to Area: Name, date, and time entered and left the area; uncleared individuals must be accompanied by approved individuals and recorded as such.

Records are reviewed routinely by the RO and EHS to ensure that they are being maintained and updated appropriately. Records must be maintained for three (3) years.

Further information regarding select agents and records will be given to laboratories working with select agents in specialized safety training by laboratory personnel authorized to work with such agents.

HHS and USDA Select Agents and Toxins

The following biological agents and toxins have been determined to have the potential to pose a severe threat to both human and animal health, to plant health, or to animal and plant products. An attenuated strain of a select agent or an inactive form of a select toxin may be excluded from the requirements of the Select Agent Regulations. For the most updated list, please refer to the Federal Select Agent Program website. Excluded agents and toxins are listed separately:

1. Abrin [6]
2. *Bacillus cereus* Biovar *anthracis* [1]
3. Botulinum neurotoxins [1][6]
4. Botulinum neurotoxin producing species of *Clostridium* [1]
5. Conotoxins (Short, paralytic alpha conotoxins containing the following amino acid sequence $X_1CCX_2PACGX_3X_4X_5X_6CX_7$) [6]
6. *Coxiella burnetii* [8]
7. Crimean-Congo haemorrhagic fever virus
8. Diacetoxyscirpenol [6]
9. Eastern Equine Encephalitis virus [4][5]
10. Ebolavirus [1]
11. *Francisella tularensis* [1]
12. Lassa fever virus
13. Lujo virus
14. Marburg virus [1]
15. Monkeypox virus [4]
16. Reconstructed replication competent forms of the 1918 pandemic influenza virus containing any portion of the coding regions of all eight gene segments (Reconstructed 1918 Influenza virus)
17. Ricin [6]
18. *Rickettsia prowazekii*
19. Severe acute respiratory syndrome coronavirus (SARS-CoV) [5]
20. SARS-CoV/SARS-CoV-2 chimeric viruses resulting from any deliberate manipulation of SARS-CoV-2 to incorporate nucleic acids coding for SARS-CoV virulence factors
21. Saxitoxin [6]

South American Haemorrhagic Fever viruses:

22. Chapare
23. Guanarito
24. Junin
25. Machupo
26. Sabia
27. Staphylococcal enterotoxins (subtypes A,B,C,D,E) [6]
28. T-2 toxin [6]
29. Tetrodotoxin [6]

Tick-borne encephalitis complex (flavi) viruses:

30. Far Eastern subtype [5]
31. Siberian subtype [5]
32. Kyasanur Forest disease virus [5]
33. Omsk hemorrhagic fever virus [5]
34. Variola major virus (Smallpox virus) [1]
35. Variola minor virus (Alastrim) [1]
36. *Yersinia pestis* [1]

Overlap Select Agents and Toxins

37. *Bacillus anthracis* [1]
38. *Bacillus anthracis* Pasteur strain
39. *Burkholderia mallei* [1]
40. *Burkholderia pseudomallei* [1]
41. Hendra virus
42. Nipah virus [1]
43. Rift Valley fever virus
44. Venezuelan equine encephalitis virus [4][5][8]

USDA Veterinary Services (VS) Select Agents and Toxins

45. African horse sickness virus
46. African swine fever virus
47. Avian influenza virus [4]
48. Classical swine fever virus [5]
49. Foot-and-mouth disease virus [1][5]
50. Goat pox virus
51. Lumpy skin disease virus
52. *Mycoplasma capricolum* [4]
53. *Mycoplasma mycoides* [4]
54. Newcastle disease virus [3][4]
55. Peste des petits ruminants virus
56. Rinderpest virus [1]
57. Sheep pox virus
58. Swine vesicular disease virus [5]

USDA Plant Protection and Quarantine (PPQ) Select Agents and Toxins

59. *Ralstonia solanacearum [7]*
60. *Rathayibacter toxicus*
61. *Sclerophthora rayssiae [7]*
62. *Synchytrium endobioticum*
63. *Xanthomonas oryzae*
64. *Xanthomonas oryzae*

[1] Denotes Tier 1 Agent
[2] C = Cysteine residues are all present as disulfides, with the 1st and 3rd Cysteine, and the 2nd and 4th Cysteine forming specific disulfide bridges; The consensus sequence includes known toxins a-MI and a-GI (shown above) as well as a-GIA, Ac1.1a, a-CnIA, a-CnIB; X1 = any amino acid(s) or Des-X; X2 = Asparagine or Histidine; P = Proline; A = Alanine; G = Glycine; X3 = Arginine or Lysine; X4 = Asparagine, Histidine, Lysine, Arginine, Tyrosine, Phenylalanine or Tryptophan; X5 = Tyrosine, Phenylalanine, or Tryptophan; X6 = Serine, Threonine, Glutamate, Aspartate, Glutamine, or Asparagine; X7 = Any amino acid(s) or Des X and; "Des X" = "an amino acid does not have to be present at this position." For example if a peptide sequence were XCCHPA then the related peptide CCHPA would be designated as Des-X.
[3] A virulent Newcastle disease virus (avian paramyxovirus serotype 1) has an intracerebral pathogenicity index in day-old chicks (Gallus gallus) of 0.7 or greater or has an amino acid sequence at the fusion (F) protein cleavage site that is consistent with virulent strains of Newcastle disease virus. A failure to detect a cleavage site that is consistent with virulent strains does not confirm the absence of a virulent virus.
[4] Select agents that meet any of the following criteria are excluded from the requirements of this part: Any low pathogenic strains of avian influenza virus, Madariaga virus, Clade II Monkeypox, any strain of Newcastle disease virus which does not meet the criteria for virulent Newcastle disease virus, all subspecies Mycoplasma capricolum except subspecies capripneumoniae (contagious caprine pleuropneumonia), all

subspecies Mycoplasma mycoides except subspecies mycoides small colony (Mmm SC) (contagious bovine pleuropneumonia), and any subtypes of Venezuelan equine encephalitis virus except for Subtypes IAB or IC, provided that the individual or entity can verify that the agent is within the exclusion category.
[5] For determining the regulatory status of nucleic acids that are capable of producing infectious forms of select agent viruses, please reference guidance here.
[6] For determining the regulatory status of Recombinant and/or Synthetic nucleic acids that encode for the toxic form(s) of any select toxins if the nucleic acids (i) can be expressed in vivo or in vitro, or (ii) are in a vector or recombinant host genome and can be expressed in vivo or in vitro; please reference guidance here.
[7] Select agents or toxins that meet any of the following criteria are excluded from the requirements of this part: Any subspecies of *Ralstonia solanacearum* except race 3, biovar 2 and all subspecies of *Sclerophthora rayssiae* except var. *zeae,* provided that the individual or entity can identify that the agent is within the exclusion category.
[8] *Coxiella burnetii* Phase II, Nine Mile Strain, plaque purified clone 4 with reversion to wildtype *cbu0533* is a select agent

Chapter 10
Transportation of Biological Materials

PLEASE NOTE: Training is required prior to shipping of materials off-campus. This chapter does not meet training requirements.

Background

The packaging and transportation of biological materials are subject to strict local, state, federal, and international regulations. This is particularly so if the material is transported through the "public domain," namely, those roadways, airways, and sea lanes accessible to the public. Therefore, unless the material is being moved within a specific campus, legal requirements governing packaging, labeling, and handling must be followed.

The intent of the packaging and transportation regulations is to prevent accidental exposure of personnel who may handle the material during its shipment. Therefore, certain general criteria apply to all possible transportation scenarios.

Transportation

Prior to transporting any biological materials, the following controls must be in place:

- Emergency procedures (e.g., contact names and information, spill cleanup, disinfection protocols, etc.) must be known to the person carrying the materials;
- Container must be appropriate for the material being transported;
- Material must be packed so that it will stay upright during transportation;
- The containers must be properly labeled;
- Proper PPE must be worn during the packaging of the material;
- Hands should be washed after handling materials;
- Open cuts or other wounds should be covered before handling the materials;
- Aerosol generation must be avoided when handling and packing the materials;
- The person packaging the material must ensure that the exterior surfaces of each package are free of any potential contamination by the packed material.

Transportation within a Campus

The following requirements must be observed during the transportation of biological materials within a campus (e.g., between two laboratories):

- At a minimum, all laboratory materials must be transported in a secondary container that is sealed, shatterproof, and leak-proof. Materials should never be carried in hands or pockets.
- The secondary container should be closeable and easy to decontaminate, an absorbent pad (or similar material) should be placed inside the secondary container to absorb any spills.
- A laboratory coat should be worn during transport.
- Label information must include the identity of the biological material or agent, the universal biohazard symbol (if the material or agent is in, or above, Risk Group 2.
- Each individual container must have enough label information to identify its contents. Other information should be on the outside of the package.

- The container should be carried directly to the intended laboratory and not taken to offices, cafeterias, or other public or inappropriate locations.
- Upon delivery, the receiving laboratory personnel should be informed and the material properly stored.
- The package should be carefully inspected for signs of leakage or other contamination and, if necessary, decontaminated before opening.

Transportation between Campuses (Ground)

Transportation of biological samples between campuses (e.g., BUMC and CRC) is subject to the general conditions described above. Biological materials should not be transported on the Boston University Shuttle (BUS). In addition, because transportation takes place through the public domain, the following other conditions apply:

- All biological samples must be packed according to Department of Transportation/International (DOT)/International Air Transport Association (IATA) regulations; this includes triple-packaging all samples, even if exempt materials.
- The specimen should be placed inside a primary container with a tight-fitting, leak-resistant top (e.g., full round-threaded screw cap with seal or stopper).
- The primary receptacle or secondary container should be labeled with the universal biohazard symbol if it contains bloodborne pathogen materials, as per required under the OSHA Bloodborne Pathogen standard 29 CFR 1910 1030.
- The primary receptacle is placed within a secondary (outermost) container that must have enough extra space to hold absorbent and cushioning materials around the primary receptacle.
- Label information must include the category of the infectious biological material or agent, (e.g. Category B or exempt human or animal specimen), and the sending and receiving laboratory identification. Category A CANNOT be transported by hand between laboratories.
- Each individual container must have enough label information to identify its contents. In addition, a sheet containing a description of contents should be placed inside the container between the outer and secondary packaging.
- Any dry ice or other coolant can now be added between the secondary and outer packaging layers. This coolant material should be placed in a shipping box that contains a Styrofoam liner or other appropriate material to ensure that the outer box is not damaged by moisture from cold packs or other coolants.
- All required DOT/IATA labeling and marking information should be on the outside of the package.
- The BU shuttle system, MBTA, taxi cabs, driving services, or other payment for transport methods must not be used for transportation of infectious agents or other biohazardous materials.
- If the package contains exempt human or animal specimens, or materials that fall under the "Category B Infectious Substances" category, the package may be moved over U.S. roadways by a member of the laboratory. This exclusion, called "Materials of Trade" (MOT) by the U.S. Department of Transportation, allows some materials that are exempt or Category B Infectious materials to be transported by a research or clinical laboratory personnel. Courier services fall under the "Exclusive Use" Exemption under DOT. This exclusion does not apply to Category A infectious substances or other categories of Dangerous Goods. This individual must have undergone shipping training in the last two years. This package must follow all requirements as

described above. Contact the Office of Research Safety, EHS for further information and questions about these DOT exemptions.

- The container should be shipped directly to the intended laboratory and not taken to offices, cafeterias, or other public or inappropriate locations.
- Upon delivery, the receiving laboratory personnel should be informed and the material properly stored. The package should be carefully inspected for signs of leakage or other contamination and, if necessary, decontaminated before opening.

Air Transport of Materials

Occasions do arise when a PI must either ship or receive biological materials from another institution. Such activities are governed by strict federal and international guidelines. See Appendix A for detailed requirements pertaining to international (import/export) shipments. Before exporting biological material, you should check 1) whether the particular material requires an export permit by reviewing the Commerce Control List, Category 1; 2) check the recipient against the "Restricted Parties Lists"; 3) verify whether license is needed for the country of import; and 4) verify that the material will not be used to design weapons of mass destructions or another military development. After determining U.S. export control requirements, you should verify the import requirements for the destination country and apply for import permits if required. Boston University's Export Control Office can provide assistance with this process.

All anticipated purchases/shipments of materials subject to the International Traffic in Arms Regulations (ITAR) must be reported to the Export Control Office in advance of the purchase/shipment to ensure that laboratories are briefed on the licensing requirements for the use of such materials.

In addition to U.S. export regulations, you are required to review the dangerous goods shipping requirements outlined below.

The International Civil Aviation Organization (ICAO) is the United Nations entity that governs all international civil aviation matters. The ICAO's *Technical Instructions for the Safe Transport of Dangerous Goods by Air* govern the shipping of dangerous goods. These technical instructions have been incorporated into U.S. law and are an acceptable method of transport in the United States (49 CFR 175).

Packaging and shipping biological materials involve certain risks with numerous potential liabilities. The International Air Transport Association's (IATA) *Dangerous Goods Regulations* (DGR), latest edition, is the worldwide gold standard for shipping. The IATA regulations apply to *all* air transport, both domestic and international flights. Following IATA's DGR ensures that a package will also meet U.S Department of Transportation requirements for ground transport.

All responsibilities for packaging and shipment of these agents have been assigned to the shipper. Only properly trained personnel may offer infectious materials for transport. The following is only a summary of the requirements for packaging and shipping infectious agents. All persons must complete the shipping training prior to the packaging and shipment of their materials.

Dangerous goods: articles or substances capable of posing significant risk to health, safety, property, or the environment when transported by surface or air. Most infectious or biological materials are considered dangerous goods and therefore are subject to shipping regulations.

Infectious substances: substances known or reasonably expected to contain pathogens. Pathogens are defined as micro-organisms (including bacteria, viruses, rickettsiae, parasites, fungi) and other agents, such as prions, which can cause disease in humans or animals. For the purposes of shipping classification, infectious substances are broken into two categories:

- **Category A**: an infectious substance transported in a form that, when exposure to it occurs, is capable of causing permanent disability or life-threatening or fatal disease in otherwise healthy humans or animals.
- **Category B**: an infectious substance that does not meet the criteria for inclusion in Category A.

Biological products: those products derived from living organisms manufactured and distributed in accordance with the requirements of national governmental authorities (e.g., the FDA). They may have special licensing requirements and are used either for prevention, treatment, or diagnosis of disease in humans or animals, or for developmental, experimental, or investigational purposes related thereto. Biological products manufactured and packaged in accordance with the requirements of appropriate national authorities that are transported for the purposes of final packaging or distribution and for personal health-care use by medical professionals are NOT subject to dangerous goods regulation. However, biological products not governed by national authorities and that are known or reasonably believed to contain infectious substances MUST be classified and shipped according to dangerous goods regulations.

Dry Ice: Commonly used a refrigerant.

Exempt Patient/Animal Specimens: patient or animal specimens for which there is a minimal likelihood that pathogens are present are exempt from most of the shipping regulations. However, they must be marked with the words “exempt human specimen” or “exempt animal specimen” and must be triple-packed as described below.

Genetically Modified Micro-Organism/Organisms (GMMO/GMO): materials in which genetic materials have been altered through genetic engineering which does not occur naturally.

Completely Exempt Substances: materials that are totally exempt for consideration under the shipping regulations:

- Substances containing micro-organisms that are non-pathogenic to humans or animals
- Substances in a form so that any present pathogens have been neutralized or inactivated such that they no longer pose a health risk (the chemical itself may be regulated – Contact EHS prior to shipping)
- Environmental samples (including food and water samples) that are not considered to pose a significant risk of infection, and
- Dried blood spots, collected by applying a drop of blood onto absorbent material, fecal occult blood screening tests, blood or blood components intended or prep to be used for transfusion, and tissues or organs intended for transplantation.

Classification and Identification

The substance to be shipped must be classified as completely exempt from regulation, an exempt human/animal specimen, or a Category A or B infectious substance. Once classified, proper shipping names and identification numbers can then be assigned to the material. Exempt human specimens require shipping names. However, Category A and B materials are assigned the following names and numbers:

- **Category A**: assign one of two (2) identifiers, depending on whether or the material infects humans:
  - UN 2814 Infectious substance affecting humans
  - UN 2900 Infectious substance affecting animals

  Note*: If a material infects both humans and animals, use the Infectious substance affecting human code, UN 2814.*
- **Category B:** UN 3373 biological substance category B
- **Dry Ice:** Un 1845 Dry Ice
- **GMMO/GMO:** UN 3245 Genetically Modified Micro-Organism/Organisms
- **Exempt Materials:** Exempt Human or Animal Specimen

Packaging

All regulated infectious substances, including Category A, Category B, and exempt patient specimens, must be triple packed:

- The innermost primary receptacle(s) is leak-proof.
- A leak-proof secondary receptacle with absorbent material and cushioning material placed between the primary and secondary receptacles to prevent the release of liquid during transport and to shield multiple primary receptacles from coming in contact with one another.
- Rigid, tertiary outer packaging that is at least 100 mm (4 in) in its smallest external dimension.

Additionally, shipments of Category A and Category B materials must be packaged according to IATA Packing Instructions 620 and 650, respectively. These guidelines require the following:

- Shipments must be prepared in such a way that they arrive at their destination in good condition and present no hazard to persons, animals and/or environment during shipment.
- Outer packaging must meet structural strength requirements and carry defined specification markings.
- Packages must be at least 100 mm (4 in) in their smallest external dimension.
- An itemized list of contents must be enclosed between the secondary container(s) and the outer packaging.
- All packages containing infectious substances must be marked durably and legibly on the outside of the package with the name and telephone number of the person responsible for the shipment.
- The shipper must make advance arrangements with the recipient and the operator to ensure the shipment can be transported and delivered without unnecessary delay.
- Substances shipped at ambient temperatures or higher must be in primary receptacles made only of glass, metal or plastic, with a positive means of ensuring a leak-proof seal. Screw caps must be reinforced with adhesive tape.
- Substances shipped refrigerated or frozen must carry the refrigerant between the secondary container and outer packaging. Wet ice is not recommended for shipping as it may cause the

package to leak during transport, thus delaying or causing rejection of the package by the transporter. If dry ice is used, the packaging must permit the release of CO_2 gas.

- Primary and secondary containers must meet temperature and pressure requirements set out in the regulations.
- Category A shipments require UN Spec packaging.

Labeling

Package labeling is in the form of standardized pictures that must be affixed to the outside. The color and design of each label is prescribed in the IATA regulations. All labels must be at least 4 inches on the smallest side.

For the purposes of infectious substances, five (5) different labels and marking must be considered:

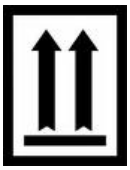
Category A:

Category B:

Dry Ice:

Cargo Aircraft Only: must be affixed if shipping volumes greater than 50 ml of a Category A substances can be halved for use on a smaller category A box.

Orientation Arrows: if shipping liquids, two such labels must be affixed to the package, on opposing sides.



Marking

Markings are the words and numbers required to be on the outside of a package. The following markings must be present on any package containing a Category A or Category B material:

- *UN Number and Proper Shipping Name*:
  - UN 2814 Infectious substance affecting humans
  - UN 2900 Infectious substance affecting animals
  - Biological substance category B
  1. Genetically Modified Micro-organisms/Organisms

  Note: The UN number is part of the label for Category B substances.

- *Contact Information*
  - Name and telephone number of the responsible person
  - 24-hour emergency telephone number in case of transportation emergency
  - "To" and "from" information

If shipping material under dry ice, the following additional marking is required:

- UN 1845 Dry Ice (the weight in kilograms of the dry ice present should also be noted)

If shipping an exempt patient or animal specimen, the only marking required is:

- Exempt Human Specimen; or
- Exempt Animal Specimen

Training Requirements

Those involved in the packaging and shipping of infectious substances must undergo training every two years or when regulations change. It is the department's responsibility to ensure training is completed; EHS can provide this training.

Shipping

The shipper is obligated to receive further qualification when shipping hazardous materials of a class or division (chemical, radiological, Li-Ion Battery, etc.) where current training is insufficient or contact EHS for more information. Below is an example of a record-keeping template:

	INSTRUCTIONS
1	Shipper's: Name Address Phone number
2	Receiver's: Name Address Phone number
3	Line out the item that does not apply. Passenger aircraft can only be used to ship quantities less than 50 ml. Cargo aircraft must be used to ship quantities between 50 ml and 4 L.
4	Line out the item that does not apply.
5	Proper Shipping Name (infectious substance, affecting humans or infectious substance, affecting animals) Identify the specimen by name in parenthesis *ex. Infectious substance, affecting humans (rabies virus)*
6	Class or Division * Always 6.2
7	UN Code * UN 2814 or UN 2900 (UN 3373 does not require shippers dec.)
8	Packaging Group * There is no packaging group for biological agents.
9	Identify by stating the number of containers by the quantity in each container. (e.g., 5 X 10ml) Identify type of outer container for the shipment
10	Packaging Instructions * 602 or 650 (also 904 if dry ice included)
11	24-hour emergency contact number for the shipper (PI, Lab Supervisor), The statements, "Prior arrangements as required by the IATA Dangerous Goods Regulations 1.3.3.1 have been made." And "Prepared according to ICAO/IATA."
12	Name and Signature of the shipper.

* As described in the latest edition of the IATA Dangerous Goods Regulations

Note*:*

- *When shipping biomaterials on dry ice, remember that dry ice is itself considered a dangerous good and must also be listed on the shipping documents as UN1845, Packing Group III, Packing Instruction 954.*
- *BU has adopted additional shipping requirements for "high hazard materials" with strict requirements for approved carriers, including a dedicated vehicle, point-to-point delivery, and specified shipping routes. For more information, contact EHS.*
- *The transportation must also meet the NIH guidelines.*

Transportation of Select BSL3 and BSL4 Agents

Regulatory requirements establish defined roles and responsibilities for the principal participants of the shipping and transportation process including the sender, the carrier and the receiver. In accordance with local, national and international guidance, BU has taken an active role in the shipper-carrier-

receiver collaboration and has established specific criteria for actively seeking pre-qualified carriers that agree to meet or exceed these requirements prior to the selection of a final carrier or carriers.

BU is committed to using ground transportation for shipment from within the United States or Canada unless doing so increases risks associated with the transportation. Shipment originating from locations where ground transportation is not an option will follow the IATA regulations for transportation of dangerous goods during the air transportation, and DOT during the ground transportation.

Additionally, BU has committed in the NEIDL Comprehensive Emergency Management Plan (CEMP) to ongoing communication, training, and drills/exercises with external responders in collaboration with the Boston Public Health Commission.

BU has adopted additional requirements for the transportation of Select Agents including the use of dedicated vehicles operated by dedicated drivers who meet background check requirements, GPS tracking of packages and the vehicle, predetermined routes that avoid residential areas, and coordination throughout transport with local emergency responders and regulators. BU will have personnel with expertise in the material being shipped available to provide direction if an incident occurs.

The below describes the NEIDL-specific processes that will be followed for shipments of BSL3 and BSL4 biological, infectious Select Agent (and other transportation events as determined by a risk assessment):

Approval and Transportation Process

- PI notifies the BU BSO that a select agent needs to be ordered.
- BSO confirms that the PI is an authorized select agent user.
- BSO confirms that IBC approvals, training, and inspection status are valid and current.
- BSO confirms that BPHC approvals and notification of the City of Boston are in place.
- BSO confirms that appropriate laboratory and storage facilities are available.
- RO or Alternate RO submits request for approval by the Federal Select Program (FSAP).
- For imported select agents, the RO or Alternate RO works with the PI to obtain CDC Import permit, USDA and/or U.S. Public Health Service (PHS) permit if required.
  The sending and receiving RO, upon approval from FSAP, coordinates transport of the select agent with the transportation / shipping provider.
- Upon completion of the shipment and verification of the agent received, the RO or designee confirms the receipt with the shipper and all applicable regulatory / response agencies.

Select Agent Shipment from NEIDL

If there is a need to ship packages containing BSL-3 and BSL-4 agents from the NEIDL to another entity, the requirements will be the same as those described above; however, the roles would change in that in those instances the NEIDL will be the “shipper”, and the other entity will be the “receiver”.

Notification Process for Select Agent Shipments

Regulatory agencies and first responders will be notified on shipping logistics / information / status in real time as well as confirmation of receipt and condition of packages and paperwork.

Transportation Regulatory Framework including Select Agents

The transportation of biological agents and toxins, as described above, are supplemented by BU

transportation standards for Select Agents as well as guidance from those below.

World Health organization

- IATA requirements follow the United Nations, World Health Organization, Guidance on Regulations for the Transport of Infectious Substances 2013–2014.
- *Biosafety in Microbiological and Biomedical Laboratories*
  The CDC / NIH Biosafety in Microbiological and Biomedical Laboratories (BMBL), 6th Edition, 2020, contains guidelines for microbiological practices, safety equipment, and facilities that constitute the four established biosafety levels.

Key References and Resources

- U.S. Department of Health and Human Services, *Biosafety in Microbiological and Biomedical Laboratories,* 6th Edition, June 2020
- U.S. Department of Transportation, 49 CFR Part 171 Final Rule
- Current Revised International Air Transport Authority, Dangerous Goods Regulations
- U.S. Public Health Service (HHS)/ CDC 42 CFR Part 73.0, "Possession, Use & Transfer of Select Agents and Toxins,"
- National Institutes of Health *Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules.*
- APHIS/CDC Guidance Document for Report and Transfer of Select Agents and Toxins (APHIS/CDC Form 2)
- APHIS/CDC Guidance Document for the Report of Theft, Loss or Release of Select Agents and Toxins (APHIS/CDC Form 3):
- Federal Select Agent website

Appendix A
Importation and Exportation of Infectious Biological Agents

Multidisciplinary and multi-institutional research is a common practice that involves collaboration among faculty from various institutions and countries. At times it is necessary to share biological samples or materials with collaborators for outbreak surveillance, research and development of diagnostics, vaccines, and therapeutics, to benefit from unique laboratory testing available in the U.S. and to support research to better understand the potential threats posed by these agents. Federal regulations strictly control the importation and exportation of infectious biological agents, equipment and technologies used for the study and processing of these agents. The regulations require that anyone wishing to import infectious biological agents, infectious substances, or vectors must first obtain a permit issued by CDC. The following outlines two major requirements that must be followed.

Note: All importation and exportation of infectious biological agents must be processed through the Biosafety Program. Contact the EHS office for assistance with this matter. Laboratories that will import infectious or potentially infectious materials must first be registered with the IBC before the sample can be obtained and stored in the laboratory.

CDC Etiologic Agent Import Permit Program

The CDC Import Permit Program, or IPP, regulates the importation of infectious biological materials that could cause disease in humans in order to prevent their introduction and spread into the U.S. The program ensures that the importation of these agents is monitored and that facilities receiving permits have appropriate biosafety measures in place to work with the imported agents.

Materials requiring import permits:

- Infectious biological agents capable of causing illness in humans
- Materials known or reasonably expected to contain an infectious biological agent
- Vectors of human disease (such as insects or bats)

Subsequent transfers of pathogens of high consequence

Infectious biological agents are those microorganisms and microbial toxins that cause disease in humans and include bacteria, bacterial toxins, viruses, fungi, rickettsiae, protozoans, and parasites. These disease-causing microorganisms may also be referred to as infectious agents. Arthropods and other organisms that transmit pathogens to animals (including humans) are called vectors.

Infectious biological agents, vectors, and materials containing infectious biological agents are recognized as hazardous materials. Materials containing infectious biological agents are regularly transported from one location to another by common land and air carriers. Materials containing infectious biological agents must be appropriately packaged to prevent breakage or leakage in order to avoid exposing the package contents to package handlers, transporters, and the general public. Materials containing infectious biological agents must be packaged, labeled, and transported in accordance with all applicable regulations. Material containing infectious biological agents being imported into the United States must be accompanied by a U.S. Public Health Service importation permit.

Importing Biological Agents and Materials into the United States

All imports into the United States must be processed by a licensed Customs Broker. Most freight forwarders such as World Courier, FedEx etc. have customs brokers on staff to help you. Before you decide to import biological materials or equipment associated with biological research into the United States, ensure that you review the U.S. import requirements to determine if an import permit or license is required. Securing an import permit may take weeks so it is critical that you start this process early. Boston University's preferred customs broker is Watchpoint Logistics. It is recommended that you contact Linda Amiro, Import Manager at Watchpoint Logistics) and the EHS Biosafety Officer in connection with export licenses or import permit application. See below:

Contact information:
Linda Amiro
International Import Manager
Watchpoint Logistics Inc.
100 Griffin Brook Drive
Methuen, MA 01844
Phone: (617) 567-6800
C 781 710 8881
www.watchpointlogistics.com
Email:Linda.Amiro@Watchpointlogistics.com

Please see here for detailed guidelines on importing into the United States by US Customs and Border Protection. You have approximately ten (10) days to file import paperwork with US Customs and Border Protection after your shipment has arrived into United States. If you don't have permits and documentation in place, your shipment will be placed into Customs storage and you will be charged daily fees and penalties. The ultimate issue with biological shipments, of course, is that your materials may get destroyed during this process before you are able to secure an import permit.

Import Permits
Import permits are issued only to the importer of record, which must be located in the United States. Import Permits may take several weeks and must be secured prior to the importation of any material. The permit, with the proper packaging and labeling, will help expedite ensure clearance of the package of infectious materials through the U.S. Public Health Service Division of Quarantine and release by U.S. Customs.

The importer is legally responsible for ensuring that the foreign personnel package, label, and ship the infectious materials according to federal and international regulations. Shipping labels with the universal biohazard symbol, the importer's address, the permit number, and the expiration date are also issued to the importer with the permit. The importer must send the labels and one or more copies of the permit to the shipper. The permit and labels inform the U.S. Customs and Border Protection and U.S. Division of Quarantine personnel of the package contents.

Boston University researchers are encouraged to contact the Office of Environmental Health and Safety and the Export Control Office prior to submitting a permit application. The Export Control Office will assist

in determining the Export Control Classification Number (ECCN) of the shipment coming to BU, and to determine if any "deemed" export licenses are needed prior. A deemed export is the sharing of controlled technology or technical data to a foreign national within the United States, treating it as if it were an export to their home country.

Federal Regulations
Regulatory authority for the CDC import program is given to the Secretary of Health and Human Services through the Public Health Service Act, which allows for the development and enforcement of regulations to prevent the introduction, transmission, or spread of communicable disease from foreign countries into and throughout the U.S. or its possessions. The importation of infectious biological agents is governed by the following federal regulation: USPHS 42 CFR - Part 71 Foreign Quarantine. Part 71.54 Import regulations for infectious biological agents, infectious substances and vectors.

A person may not import into the United States, nor distribute after importation, any etiologic agent or any arthropod or other animal host or vector of human disease, or any exotic living arthropod or other animal capable of being a host or vector of human disease unless accompanied by a permit issued by the CDC Director.

Any import coming within the provisions of this section will not be released from custody prior to receipt by the District Director of U.S. Customs Service of a permit issued by the Director (Centers for Disease Control and Prevention).

Items Requiring Permits

Infectious Biological Agents
It is impractical to list all etiologic agents in this document. In general, an import permit is needed for any infectious agent known or suspected to cause disease in humans. Use the CDC import permit online tool to find out if the material to be imported requires and import permit.

Biological materials
Unsterilized specimens of human and animal tissues (such as blood, body discharges, fluids, excretions, or similar material) containing or suspected to contain an infectious biological agent may require a permit in order to be imported.

If you are importing biological materials that are not infectious, you may be required to submit such declaration to U.S. Customs at the time of the import. This document should be on a sender's university/hospital/company letterhead and signed by an individual who can legally bind the entity. It should accompany all import shipments into the United States to expedite Customs clearance.

Hosts and vectors
- *Animals*: any animal known or suspected of being infected with an organism capable of causing disease in humans requires a permit issued by CDC. Importation of live turtles of less than 4 inches in shell length and live nonhuman primates is regulated by the CDC's Division of Global Migration and Quarantine.
- *Bats*: all live bats require an import permit from the CDC and the U.S. Department of Interior, Fish and Wildlife Services. The application for a CDC import permit for live exotic bats is at CDC Importation of Animals website.

- *Arthropods*: any living insect or other arthropod that is known or suspected of containing an infectious biological agent requires a CDC import permit.
- *nails*: snail species capable of transmitting a human pathogen require a CDC permit.

Packaging Requirements

Infectious materials imported into this country must be packaged to withstand breakage and leakage of contents and be labeled, as specified in the following federal regulations:

- DOT 49 CFR PART 173 - General Requirements for Shipments and Packaging
- For international shipments, the International Air Transport Association's (IATA) *Dangerous Goods Regulations* must be consulted.

Importation of Animal Pathogens and related biological materials

USDA and APHIS permits are required for infectious agents of livestock and biological materials containing animal material. Tissue culture materials and suspensions of cell culture-grown viruses or infectious biological agents containing growth stimulants of bovine or other livestock origins are controlled by the USDA because of the potential risk of introduction of exotic animal diseases into the United States. For more information, contact USDA/APHIS at their website:

Principal Investigators must submit USDA/APHIS permit applications via EHS. More information about the application process can be found in the IBC Policies page.

Importation of Wildlife and Animals

U.S. Fish and Wildlife Service permits are required for certain live animals, including bats. For more information, call (800) 344-WILD or visit their website here.

Importation of Select Agents

Individuals wishing to import select agents and toxins must be registered with the Federal Select Agent Program (FSAP) in accordance with 42 CFR Part 73 (Possession, Use, and Transfer of Select Agents and Toxins, Interim Final Rule) for the select agent(s) and toxin(s) listed to the imported permit application. Also, in accordance with 42 CFR Part 73.16(a), an FSAP Form 2 must be completed and submitted to the FSAP and granted approval prior to the shipment of the select agents or toxins under the import permit. Additional information can be found at Form 2 Transfer Guidance. Clinical specimens suspected of containing select agents require a CDC approved import permit. If the select agent has been already identified in the clinical specimens, a FSAP approved Form 2 is required for the transfer of the material into the USA (instead of an import permit).

Importation of Research Equipment

Research equipment: Some equipment used in biological and medical research must be approved by the Food and Drug Administration (FDA) and must have appropriate safety certificates. Before purchasing equipment abroad, ask the manufacturer if this equipment requires FDA approval and if the company has such approvals in the United States.

Exportation of Infectious Materials

There are two (2) sets of export controls that govern the export of biological materials: the Export Administration Regulations (EAR) and the International Traffic in Arms Regulations (ITAR). The EAR

govern the export of dual-use agents, genetic material, vaccines and equipment used in biological research. Prior to the export, you should review the Commerce Control List (CCL) (Category 1) to determine if your material is controlled under an Export Control Classification Number (ECCN). If the material is listed on the CCL, contact the Export Control Office to help you determine if a license is required. Licensing requirements are based on the material, customer and end-use. If the material is not listed on the CCL and is not controlled under the U.S. Munitions List, it is designated as EAR99. EAR99 material can be exported abroad without an export license unless your collaborator is listed on the "Restricted Parties Lists" or you ship to a country where U.S. maintains a comprehensive embargo administered by the Office of Foreign Assets Control; or the end use will be weapons of mass destructions or military end use.

If you are unable to determine the ECCN of your material, the Export Control Office will assist, and if necessary will submit a commodity classification request with the Department of Commerce, Bureau of Industry and Security on your behalf. Classification requests can take anywhere from few days to several months so it is important to plan ahead.

Biological agents, pathogens, toxins specifically modified, developed, configures or adapted for military use are controlled on the U.S. Munitions List (Category XIV) under the International Traffic in Arms Regulations (ITAR). Research with these agents is heavily regulated and licenses are required for you to share these materials with non-U.S. Persons whether abroad or in the United States. Contact the Export Control Officer before shipping ITAR controlled materials abroad as a license is required.

Click here for more information on export controls and biological materials under the Bureau of Industry and Security (BIS) Export Administrative Regulations (EAR). **Export controls regulate more materials/agents than CDC and USDA regardless of quantity or attenuation, therefore, it is critical that you review the Commerce Control List and the US Munitions List prior to any export shipment. DO NOT ASSUME THAT YOUR MATERIAL IS NOT REGULATED.**

In addition to the product/material based regulations, you may need an export license based on the destination and customer. Office of Foreign Assets Controls manages economic sanctions and embargoes and you may need an export license to ship to any of these countries. Before you make any export shipments, please verify by screening the Restricted Parties List that no license is required for the receiving researcher, entity or end-user. Some universities are on those lists.

Export licenses or requests for material classifications may take months so you are encouraged to start this process early.

In addition, to the licensing requirements, you should understand that you will need to complete export shipping paperwork. Standard documents include Commercial Invoice and Air Waybill; however, additional documentation may be required depending on the importing country requirements. You should also verify the requirements of the importing country for licensing biological materials prior to dispatching export shipments. Contact the receiving entity or your freight forwarder to provide assistance with foreign country import requirements.

Please review the BU Export Shipping Process below. To learn more about export controls at Boston

University please sign up for Export Control Training here.

Foreign Trade Regulations

All export shipments valued over $2,500 per commodity classification or subject to an export license must be reported via the Automated Export System to U.S. Census Bureau prior to the export. Your freight forwarder will be able to handle the reporting, Alternatively, regardless of carrier, Watchpoint Logistics will assist you. Please visit here under the heading “Shipment/Transport Questions to Answer” in advance of contacting Watchpoint in anticipation of the information they will need up front.

Recordkeeping

All shippers are required to keep records on file five (5) years from the date of the export or expiration of an export license whatever period is longer.

Appendix B
Laboratory Ventilation and Containment for Biosafety

Laboratory-ventilated containment equipment fall into three (3) major categories:

Laboratory Chemical (Fume) Hoods
Traditional laboratory chemical (or fume) hoods are designed to capture and control chemical vapors and pull them away from the worker. Although the inward flow of air protects the user, chemical hoods do not protect the product (the desired organism being manipulated).

Other Local Exhaust Ventilation (LEV) Systems

Horizontal Laminar Flow Clean Bench
With horizontal laminar flow clean benches, HEPA-filtered air flows horizontally across the workspace directly toward the user. These clean benches provide product protection and were originally designed to provide a particulate-free environment for the manufacture of semiconductor components.

Clean benches provide product protection against microbial contamination, but they *do not* provide personal or environmental protection. In fact, the horizontal flow of air will blow biological agents directly toward the user and into the laboratory. Clean benches are not a biological safety cabinet, and they should not be used with any materials (biological, chemical, or radiological) requiring containment for protection of personnel or the environment.

Clean benches are acceptable for use with materials that do not present risks to the laboratory workers (including immunocompromised individuals who may frequent the lab). Human cell lines and nonhuman primate cell lines are generally considered to be BSL2 agents and would not be suitable for use in a clean bench.

Biological Safety Cabinets

Biological safety cabinets (BSCs) are divided into Class I, II, and III (see schematic below). Class II BSCs are subdivided into type A and type B. All BSCs provide personnel and environmental protection, with Class II BSCs also providing product protection.

- Personnel protection is achieved by inward airflow through the front of the cabinet.
- Product protection is achieved by downward HEPA-filtered airflow from the top of the cabinet.
- Environmental protection is achieved by HEPA filtration of exhaust air.

New NSF Classification (Adopted 2002)	Previous NSF Classification	General Description
A1	Class II, Type A	- Maintain minimum average inflow velocity of 75 ft/min (0.38 m/s) through the work access opening; - have HEPA/ULPA filtered downflow air that is a portion of the mixed downflow and inflow air from a common plenum (i.e., a plenum from which a portion of the air is exhausted from the cabinet and the remainder supplied to the total work area; - may exhaust HEPA/ULPA filtered air back into the laboratory or to the environment through an external exhaust system connected to the cabinet with a canopy connection; and - have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums. If using chemicals with toxic vapors, the unit shall be connected to an external exhaust system. Type A1 cabinets may be used for work with volatile chemicals if deemed appropriate by a chemical risk assessment NOTE — Type A1 BSCs manufactured prior to 2010 are not suitable for work with volatile chemicals due to the contaminated positive pressured plenums that are not surrounded by negative pressure plenums.
A2	Class II, Type (formally Class II A/B3) When exhausted to the environment were formally designated Type B3	- Maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening; - have HEPA/ULPA filtered downflow air that is a portion of the mixed downflow and inflow air from a common exhaust plenum; - may exhaust HEPA/ULPA filtered air back into the laboratory or to the environment through an external exhaust system connected to the cabinet with a canopy connection; and - have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums. If using chemicals with toxic vapors, the unit shall be connected to an external exhaust system. Type A2 cabinets may be used for work with volatile chemicals if deemed appropriate by a chemical risk assessment
B1	Class II, Type B1	- Maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening; - have HEPA/ULPA filtered downflow air composed largely of uncontaminated recirculated inflow air; - exhaust contaminated downflow air from a region of the total work area via an internal dedicated exhaust plenum and through HEPA/ULPA

		filter(s) to a dedicated external exhaust system for BSCs with a direct connection and exhausted to the atmosphere; - recirculate the balance of the downflow and inflow air through a supply HEPA/ULPA filter(s); and - have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums. Type B1 cabinets may be used for work with volatile chemicals if permitted by a chemical risk assessment
B2	Class II, Type B2	- Maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening; - have HEPA/ULPA filtered downflow air drawn from the laboratory or the outside air (i.e., downflow air is not recirculated from the cabinet exhaust air); - exhaust all inflow and downflow air to the atmosphere through a dedicated external exhaust system for BSCs connected to the cabinet with a direct connection after filtration through a HEPA/ULPA filter without recirculation in the cabinet or return to the laboratory; and - have all contaminated ducts and plenums under negative pressure or surrounded by directly exhausted (non-recirculated through the total work area) negative pressure ducts and plenums Type B2 cabinets may be used for work with volatile chemicals if permitted by a chemical risk assessment
C1	Class II, Type C1	- Maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening; - have HEPA/ULPA filtered down-flow air composed largely of uncontaminated recirculated inflow air; - exhaust contaminated down-flow air from a region of the total work area via an internal dedicated exhaust plenum and blower, and then through HEPA/ULPA filter(s); - recirculate the balance of the down-flow and inflow air through a supply HEPA/ULPA filter(s); - have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums; and - may exhaust HEPA/ULPA filtered air either back into the laboratory or via a canopy connection to an external system that exhausts to the atmosphere. If working with volatile chemicals, the unit must be connected to an external exhaust system. Type C1 cabinets may be used for work with volatile chemicals if permitted by a chemical risk assessment

Certification of BSCs

Generally, BSCs are tested by the cabinet manufacturer in accordance with NSF International criteria. Cabinets that meet the NSF 49 criteria for performance characteristics, including biological containment, ventilation, cabinet leakage, and HEPA filter leakage, are NSF certified.

Field certification of BSCs is also required to ensure that the cabinet still performs as it did when it obtained NSF certification at the factory. NIH requires field certification under the following circumstances: (1) upon installation of a new BSC, (2) annually thereafter, (3) after repair or maintenance is performed, and (4) after the BSC is moved and relocated.

CDC recommends that BSCs be recertified annually to ensure for proper function. They will also be recertified after being moved to ensure that they have not been damaged. Laboratories are responsible for ensuring that the BSCs are recertified in a timely manner. Laboratories at CRC will contact EHS at (617) 353-4094. Laboratories at BUMC will contact the certification contractor directly. The contact information to reach the contractor is indicated on the certification sticker affixed on the front of the BSC.

NSF/ANSI 49 provides criteria for construction of BSCs, testing by manufacturers (including biological containment testing), and field certification. NSF has also established a certification program for field certifiers to ensure a minimum level of competency and professionalism. It is recommended that NSF field certifiers be used for field certification of BSCs. Field certification tests include:

Primary tests (BSC performance):

- Inflow test
- Down-flow test
- Smoke pattern test
- HEPA filter leakage
- Cabinet leakage (Applies to Class II BSC designed with biologically contaminated plenums under positive pressure to the lab old type A BSC.) Perform tests when BSC is newly installed, relocated, or maintenance has been completed.

Additional tests (worker comfort and safety), performed at discretion of certifier:

- Noise
- Vibration
- Lighting
- Electrical leakage, polarity, and ground circuit resistance

Appendix C
Autoclave Quality Assurance Program

Autoclaving is an accepted procedure for the decontamination of certain biohazardous waste. Biological cultures and stocks, contaminated solid waste, and liquid waste can be sterilized through autoclaving. After sterilization in a steam autoclave, these materials are considered non-infectious. All autoclaved waste is placed into the solid biohazard waste stream. Wastes from the BSL4 and BSL3 containment laboratories are first autoclaved before being removed from containment and disposed as biological waste. Materials that contain hazardous chemicals or radioisotopes are not to be autoclaved.

Sterilization by autoclaving is accomplished through exposure and penetration of the contaminated material by superheated steam for an adequate amount of time. Because steam will not penetrate a sealed plastic autoclave bag, the bags containing dry loads must not be tightly sealed (rubber band closures will allow bags to "breathe") to allow the steam to enter the bag or an adequate amount of water must be added to the load. Consult the manufacturer's instructions for sterilizing materials inside plastic autoclave bags. Liquid waste may also be autoclaved in lieu of adding an appropriate chemical disinfectant and disposed of in the sink.

To ensure that biohazardous waste is properly decontaminated during autoclaving, the following procedures should be followed by laboratory personnel.

1. Infectious waste must be treated in an autoclave for a ***minimum of 30 minutes at 121°C (250°F)****;* however, the total processing time required to decontaminate infectious waste depends on the specific loading factors (container type, water content, quantity, etc.) A total processing time of 60 minutes is recommended for gravity displacement autoclaves and 10 minutes for vacuum-type autoclaves (132°C).

2. All autoclaved waste must include a chemical sterilization indicator; (the use of biohazard bags with a "built-in" indicator is recommended).

3. Steam autoclaves used to treat infectious waste must operate at a minimum temperature of 121°C. The operating temperature of the autoclave must be verified for each run by maintaining a record of the temperature either as a chart or paper tape recording or a manual recording in a logbook.

4. On a monthly basis, confirm that adequate sterilization conditions are being met through the use of Biological Indicators (BI) containing heat-resistant spores (e.g. Geo*bacillus stearothermophilus*) placed in the center of an autoclave load. In conjunction with the BI testing, measure and record the maximum temperature achieved during the autoclave cycle through the use of a maximum registering (or "holding") thermometer or calibrated data logger for full cycle.

5. Maintain records of BI testing and maximum autoclave temperature recordings for a minimum of three one year (see Autoclave QC Log at end of appendix).

Monthly Spore Testing Procedure

1. Place BI spores and holding thermometer or data logger in the center of an autoclave load.

2. Process the load under normal operating procedures.

3. The highest temperature indicated on the holding thermometer is entered on the Autoclave QC Log. If this temperature is less than 121°C, the autoclave is not to be used to treat infectious waste until it has been repaired and passes retesting. In the interim, tag the autoclave as “Not Approved for Infectious Waste.”

4. Incubate the autoclaved BI and a non-autoclaved, control BI according to the manufacturer’s instructions (usually 55°-60°C overnight).

5. If a color change (cell growth) occurs in the autoclave BI, the sterilization process was unsuccessful. Discontinue use of the autoclave until it is checked, repaired as needed. and passes retesting. Tag the autoclave as “Not Approved for Infectious Waste” until the autoclave passes retesting.

6. Indicate test results on Autoclave QC Log available from EHS and retain for at least three years.

Appendix D
Biosafety Level 2 (BSL2) Requirements

Background

Biosafety Level 2 (BSL2) is suitable for experiments involving agents of moderate potential hazard to personnel and the environment.

For example:

- Microorganisms of low biohazard potential, such as those in Risk Group 2 or BSL2.
- Recombinant DNA activity requiring BSL2 physical containment including animal studies that involve the construction of transgenic animals.
- Non-recombinant cell and/or tissue culture systems that require this level of containment.
- Oncogenic viral systems classified as low risk.
- Production activities with Risk Group 1 organisms.

The control of potential biohazards at the BSL2 level is provided by use of standard microbiological practices with the addition of personnel protective equipment (lab coat and gloves). The following are procedures used with BSL2 containment requirements. They are based on the recommendation of the *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) 6th Edition, 2020 and BU policies and procedures.

Standard Microbiological Practices

- Access to the laboratory is limited or restricted at the discretion of the laboratory director when experiments are in progress.
- Persons wash their hands after they handle viable materials, after removing gloves, and before leaving the laboratory.
- Eating, drinking, smoking, handling contact lenses, and applying cosmetics are not permitted in the work areas. Food is stored outside the work area in cabinets or refrigerators designated for this purpose only.
- Personnel who use contact lenses will consult with EHS if required to use eye protection in the lab.
- Use of personal electronic devices such earbuds and cell phones should not be practiced when working in the lab.
- Mouth pipetting is prohibited; mechanical pipetting devices are used.
- Policies for the safe handling of sharps are instituted.
- All procedures are performed carefully to minimize the creation of splashes or aerosols.
- Work surfaces are decontaminated upon completion of work, or at the end of the day, and after any spill or splash of viable material with disinfectants that are effective against the agents of concern.
- An insect and rodent control program is in effect.

- Access to the laboratory is limited or restricted by the laboratory director when work with infectious agents is in progress. In general, persons who are at increased risk of acquiring infection, or for whom infection may have serious consequences, are not allowed in the laboratory or animal rooms. For example, persons who are immunocompromised or immunosuppressed may be at increased risk of acquiring infections. The laboratory director in consultation with ROHP has the final responsibility for assessing each circumstance and determining who may enter or work in the laboratory or animal room.
- The Principal Investigator (PI) or Laboratory Director establishes policies and procedures whereby only persons who have been advised of the potential hazards and meet specific entry requirements (e.g., immunization) may enter the laboratory.
- A biohazard sign must be posted on the entrance to the laboratory when etiologic agents are in use. Appropriate information to be posted includes the agent(s) in use, the biosafety level, the investigator's name and telephone number, any personal protective equipment that must be worn in the laboratory, and any procedures required for exiting the laboratory.
- Laboratory personnel receive appropriate immunizations or tests for the agents handled or potentially present in the laboratory (e.g., hepatitis B vaccine or TB skin testing).
- Biosafety procedures are incorporated into standard operating procedures or in a biosafety manual adopted or prepared specifically for the laboratory by the laboratory director. Personnel are advised of special hazards and are required to read and follow instructions on practices and procedures.
- The Principal Investigator or Laboratory Director ensures that laboratory and support personnel receive appropriate training about the potential hazards associated with the work involved, the necessary precautions to prevent exposures; and the exposure evaluation procedures. Personnel receive annual updates or additional training as necessary for procedural or policy changes.
- A high degree of precaution must always be taken with any contaminated sharp items, including needles and syringes, slides, pipettes, capillary tubes, and scalpels.
- Needles and syringes or other sharp instruments should be restricted in the laboratory for use only when there is no alternative, such as parenteral injection, phlebotomy, or aspiration of fluids from laboratory animals and diaphragm bottles. Plasticware should be substituted for glassware whenever possible.
- Only needle-locking syringes or disposable syringe-needle units (e.g., needle is integral to the syringe) are used for injection or aspiration of infectious materials. Used disposable needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal; rather, they must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal. Non-disposable sharps must be placed in a hard-walled container for transport to a processing area for decontamination, preferably by autoclaving. Syringes that re-sheathe the needle, needle-less systems, and other safety devices are used when appropriate. Non-disposable sharps must be also be stored in a secondary container with lid when not in use.
- Broken glassware must not be handled directly by hand, and must be removed by mechanical means such as a brush and dustpan, tongs, or forceps. Containers of contaminated needles, sharp equipment, and broken glass are decontaminated before disposal according to any local, state, or federal regulations.

- Cultures, tissues, specimens of body fluids, or potentially infectious wastes are placed in a container with a cover that prevents leakage during collection, handling, processing, storage, transport, or shipping.
- Laboratory equipment and work surfaces should be decontaminated with an effective disinfectant on a routine basis, after work with infectious materials is finished, and especially after overt spills, splashes, or other contamination by infectious materials. Prior to its removal from the facility, contaminated equipment must be decontaminated according to any local, state, or federal regulations before it is sent for repair or maintenance or packaged for transport in accordance with applicable local, state, or federal regulations.
- Spills and accidents that result in overt exposures to infectious materials are immediately reported to the Principal Investigator and Laboratory Director. Medical evaluation, surveillance, and treatment are provided as appropriate, and written records are maintained.
- Sinks in the BSL2 area should be made free of clutter and cleaned routinely with appropriate disinfectant such as a 10% bleach solution and flushed down with running water after a few minutes. Water baths and all water reservoirs should be washed periodically with a suitable chemical decontaminant.
- Once a month, work spaces should be cleaned and disinfected, as well as other lab areas where clutter accumulates (e.g., storage areas).
- The laboratory will set up a routine schedule to perform surface cleaning with appropriate chemical disinfectant of large equipment (such as incubators) as part of laboratory good practices.
- Supplies should be rotated and outdated material discarded. Unlabeled material should be eliminated.
- Eliminate and clean clutter.
- Custodial services: only personnel with appropriate authorization may enter a BSL2 facility while BSL2 research activity is in progress.
- Animals not involved in the work being performed are not permitted in the lab.

Safety Equipment (Primary Barriers)

Properly maintained biological safety cabinets, preferably Class II, or other appropriate personal protective equipment or physical containment devices are to be used when:

- Procedures that have the potential to create infectious aerosols or splashes are conducted. These may include centrifuging, grinding, blending, vigorous shaking or mixing, sonic disruption, opening containers of infectious materials whose internal pressures may be different from ambient pressures, inoculating animals intranasally, and harvesting infected tissues from animals or embryonate eggs;
- High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory with sealed rotor heads or sealed centrifuge safety cups are used, and if these rotors or safety cups are opened only in a biological safety cabinet;
- Face protection (i.e., goggles, mask, face shield, or other splatter guard) is used for anticipated splashes or sprays of infectious or other hazardous materials to the face when the microorganisms must be manipulated outside the BSC;
- Protective laboratory coats, gowns, smocks, or uniforms designated for lab use are worn while in the laboratory. This protective clothing is removed and left in the laboratory before leaving for non-laboratory areas (e.g., cafeteria, library, administrative offices). All protective clothing is

either disposed of in the laboratory or laundered by the institution; it should never be taken home by personnel;

- Gloves are worn when hands may contact potentially infectious materials, contaminated surfaces, or equipment. Wearing two pairs of gloves may be appropriate. Gloves are disposed of when overtly contaminated and removed when work with infectious materials is completed or when the integrity of the glove is compromised. Disposable gloves are not washed, reused, or used for touching "clean" surfaces (keyboards, telephones, etc.). They should not be worn outside the lab. Alternatives to powdered latex gloves should be available. Hands are washed following removal of gloves.

Procedures for Receiving and Inspecting Samples

The following procedures should be used for receiving and inspecting samples:

- The PI will designate a responsible person for the purchase of all infectious materials to be used in the BSL2 lab.
- Infectious materials will be shipped to the laboratory in accordance with the appropriate Department of Transportation (DOT) and the International Air Transportation Association (IATA) standards for shipping of infectious biological materials. (*See Appendix A for more information*)
- Upon receipt of the package, it will be placed on a tray covered with absorbent material and opened in the Biological Safety Cabinet to prevent any potential exposure to personnel in case the container leaked during transport.
- Personnel assigned to open packages will wear lab coat, gloves, and eye protection.
- If any containers are found to be damaged, leaking or otherwise contaminated, they will be immediately isolated into a plastic bag along with all packaging materials. The spill will be disinfected and cleaned up. The Principal Investigator, lab director or designee will be notified immediately. The incident will be reported to EHS and as necessary, to other appropriate agencies.
- If, after inspection, the samples are intact, they can be placed into labeled secondary containers (unbreakable plastic containers or metal tubes) and then transferred to a storage area.
- Only staff who are authorized to do so can remove samples from storage. Removal and use of all such materials must be entered into the logbook (logbook documentation is usually not required for most BSL-2 agents except may be high titers/concentrations of certain BSL-2 viruses/bacteria.)
- Unused cultures can be returned to storage after the outer container has been properly disinfected.

Laboratory Facilities (Secondary Barriers)

In a BSL2 lab, the following conditions are required:

- Doors that can be locked and secured should be installed for facilities that house restricted areas.
- Consideration should be given to locating new laboratories away from public areas.
- Each laboratory contains a sink for handwashing.
- The laboratory is designed so that it can be easily cleaned. Carpets and rugs in laboratories are inappropriate.

- Bench tops are impervious to water and resistant to moderate heat and the organic solvents, acids, alkalis, and chemicals used to decontaminate the work surfaces and equipment.
- Laboratory furniture is capable of supporting anticipated loading and uses. Spaces between benches, cabinets, and equipment are accessible for cleaning. Chairs and other furniture used in laboratory work should be covered with a non-fabric material that can be easily cleaned and decontaminated.
- Biological safety cabinets should be installed in such a manner that fluctuations of the room's air supply and exhaust air do not cause them to operate outside their parameters for containment. Locate BSCs away from doors, windows that can be opened, heavily traveled laboratory areas, and other potentially disruptive equipment to avoid disruption of the BSC's air flow parameters.
- An eyewash station is readily available for use.
- Illumination is adequate for all activities, avoiding reflections and glare that could impede vision.
- There are no specific ventilation requirements. However, planning of new facilities should consider mechanical ventilation systems that provide an inward flow of air without recirculation to spaces outside of the laboratory. If the laboratory has windows that open to the exterior, they are fitted with fly screens.

Appendix E
Guidelines for Work with Toxins of Biological Origin

Biological Toxins

Biological toxins are poisonous substances produced by living organisms including animals, plants, or microbial sources that have the ability to cause death or severe incapacitation at relatively low exposure levels. Biological toxins can be small molecules, small peptides, or multi-subunit proteins, but do not replicate and therefore, are not considered infectious. These substances typically interfere with important physiological processes at the cellular level, such as inhibition of critical enzymatic activity, receptor activity, or some macromolecular interactions. Because their mode of action is potent and specific, they are often used in biomedical research laboratories, as well as in clinical research. Because of their use as import tools in biological research, it is important that laboratory workers understand and are able to assess risks associated with the use of biological toxins; these toxins are difficult to transmit from person-to-person, they are non-volatile, and usually are not dermally-active (mycotoxins are an exception), and they tend to be more toxic per weight than many chemical agents. These guidelines are intended for biological toxins handled under BSL2 conditions.

General Information

Working with biologically-derived toxins may present health risks in the event of accidental injection, absorption through skin or mucus membrane, inhalation or ingestion. However, these toxins can be handled safely by properly-trained laboratory personnel when the work is carried out under appropriate containment, such as Class II or III biosafety cabinets or fume hoods as well as PPE (i.e., lab coats, eye protection, masks, and gloves). Under such conditions, work with biological toxins generally do not pose a serious risk to the local community. The laboratory facilities, equipment, and procedures for work with these toxins must reflect the intrinsic level of hazard and potential risks. If both toxins and associated infectious agents are used, both must be considered when containment equipment is selected and policies and procedures are written. If animals are used, animal safety practices must also be considered. The IBC and EHS will evaluate all such uses and determine if the proposed safeguards are adequate or not.

Select Agent Toxins

The US Government has designated a set of specific biological toxins that are considered a potential threat to public, animal or plant health, as Select Agent Toxins. Use of these select agent toxins are heavily regulated under the Federal Select Agent Program (FSAP), requiring strict security measures and registration for possession, use, and transfer (more detail below). The federal list of select agents and toxins and exemptions can be found here.

Registration and Inventory

The use of any biological toxin in a lab when their 50% lethal dose (LD_{50}) is less or equal to 100 µg/kg will need approval from the IBC. Standard practices listed under BSL2 and/or BSL3 should be reviewed and incorporated as appropriate into protocols for work with biological toxins. Select agent toxins are not regulated under the Federal Select Agent Program if the amount stored and possessed by the lab at any given time does not exceed the permissible toxin amounts. The permissible amount for each select

agent toxins is listed here. A list of commonly-used biological toxins and their LD_{50}, including possession limit is provided in the table below.

PLEASE NOTE: When a biological toxin is exclusively used in **vivarium for animal work** (such as bacterial **lipopolysaccharide endotoxin (LPS),** registration with the IACUC in an animal work protocol is sufficient. Such registration must include the details of safety practices in the creation of a stock solution, exposure mitigation plan, use of PPE, spill management, and disposal of animal excreta.

Table: LD50 of commonly used Biological Toxins

Toxin Name	LD_{50} (µg/Kg body weight)	Select Agent Toxin? (Possession Limit)
Abrin	0.7	Yes, 1000 mg
Aerolysin	7	
Botulinin toxin A	0.0012	Yes, 1 mg
Botulinin toxin B	0.0012	Yes, 1 mg
Botulinin toxin C1	0.0011	Yes, 1 mg
Botulinin toxin C2	0.0012	Yes, 1 mg
Botulinin toxin D	0.0004	Yes, 1 mg
Botulinin toxin E	0.0011	Yes, 1 mg
Botulinin toxin F	0.0025	Yes, 1 mg
b-bungarotoxin	14	
Caeruleotoxin	53	
Cereolysin	40-80	
Cholera toxin	250	
Clostridium difficile enterotoxin A	0.5	
C. difficile cytotoxin B	220	
C. perfringens lecithinase	3	
C. perfringens kappa toxin	1500	
C. perfringens perfringolysin O	13-16	
C. perfringens enterotoxin	81	
C. perfringens beta toxin	400	
C. perfringens delta toxin	5	
C. perfringens epsilon toxin	0.1	
Conotoxin	12-30	Yes, 200 mg
Crotoxin	82	
Diphtheria toxin	0.1	
Lipopolysaccharide (LPS)	0.001-0.004	
Listeriolysin	3-12	
Leucocidin	50	
Modeccin	1-10	
Nematocyst toxins	33-70	
Notexin	25	
Pertussis toxin	15	
Pneumolysin	1.5	
Pseudomonas aeruginosa toxin A	3	
Ricin	2.7	Yes, 1000 mg

Saxitoxin	8	Yes, 500 mg
Shiga toxin	0.25	
Shigella dysenteriae neurotoxin	1.3	
Streptolysin O	8	
Staphylococcus enterotoxin B	25	Yes, 100 mg
Staphylococcus enterotoxin F	2-10	Yes, 100 mg
Streptolysin S	25	
T-2 Mycotoxin	1	Yes, 10,000 mg
Taipoxin	2	

(adapted from University of Florida EHS)

Standard Practices

All laboratory personnel must have experience biosafety training specific for the toxins being used. Training is documented and repeated annually or as needed for new personnel before use. Toxin-specific training should include description of the toxin, its importance to public health and/or research priorities, medical consequences of any exposure, appropriate lab safety measures including decontamination policies, and instructions for how to deal with accidental exposure. If needed and when requested by the PI, toxin-specific training can be provided by the ROHP and EHS. Specific attention must be paid to the storage, decontamination, prevention of aerosolization and choice of PPE while working with biological toxins.

Biological toxins have varying susceptibility to inactivation and decontamination measures. For example, most are resistant to freeze-thaw, but susceptible to 10% (v/v) fresh bleach solutions. Refer to the CDC BMBL (6th Edition) Appendix I Guidelines for tables detailing toxin-specific inactivation and decontamination recommendations as appropriate.

Biological toxin stocks must be kept in secured storage rooms, cabinets, or freezers with restricted access. If toxins are stored in the laboratory, containers should be sealed, labeled, and secured to restrict access; refrigerators and other storage containers should be clearly labeled and provide contact information for trained, responsible laboratory staff.

Dry forms of toxins present a high aerosol risk. Primary containers of dry forms of toxins should be handled in a chemical fume hood, a glove box, or a biological safety cabinet or equivalent containment system. All work should be done within the operationally effective zone of the hood or biological safety cabinet, and each user should verify the inward airflow before initiating work, and whenever possible, reconstitute entire vial of powdered toxin by injecting diluent through septum. HEPA and/or charcoal filtration of the exhaust air may be required, depending on the toxin.

If infectious agents and biological toxins are used together in an experimental system, lab members should consider both when selecting protective clothing and equipment. When vacuum lines are used with systems containing toxins, they should be protected with a HEPA filter to prevent entry of toxins into the lines. Water aspirators should be avoided.

In the event powdered form of toxin must be handled, use of anti-static nitrile gloves are recommended. When handling toxins that are percutaneous hazards (irritants, necrotic to tissue, or extremely toxic

from dermal exposure), select gloves that are known to be impervious to the toxin. Additional PPE such as disposable, long-sleeved gown may be necessary.

For Select Agent Biological Toxins and selected low molecular weight toxins, an inventory control system should be in place to account for toxin use and disposition according to the CDC BMBL (6th edition) Guidelines for Work with Toxins of Biological Origin. Select Agent Toxins require inventory detail to ensure that total quantities remain below the regulated amount. Select Agent Toxin inventories should include date and quantity of each acquisition (purchase, transfer, etc.), use and disposal. For other biological toxins, an inventory control system is not required unless mandated by the IBC (generally for biological toxins with more than 100 µg/Kg LD50). Details of LD50, storage, biosafety and containment practice and disposal of the biological toxins (including select agent biological toxins) to be used in research must be clearly stated in the IBC application. A list of commonly used biological toxins and their LD50, including possession limit is provided in the table below (adapted from University of Florida EHS).

Standard Laboratory Practices

Appropriate laboratory practices will depend on a number of parameters, such as the toxin used or the type of operations performed. General guidelines below are for biological toxins recommended to be handled under BSL2 conditions. In addition to standard BSL2 PPE, other protective equipment may be required depending on the characteristics of the biological toxin and the containment system. For example, additional respiratory protection may be necessary if aerosols may be generated and it is not possible to use containment equipment or other engineering controls.

Each laboratory should develop Standard Operating Procedures (SOPs) for toxin handling and policies (or a chemical hygiene plan specific to the toxin(s) used in that laboratory. The SOPs should include:

- Description of hazards that will be encountered during expected use of the toxin and those that could be encountered in case of a spill or other accident. This is typically a summary of the biosafety training for that toxin.
- Policies and practices to be used to minimize risks such as:
    - Containment and personal protective equipment;
    - Toxin-appropriate methods for decontamination of PPE, equipment, and bench areas;
    - Transport of toxin outside the laboratory. Toxins should be transported only in leak-/spill-proof secondary containers;
    - Management of spills;
    - Medical surveillance;
    - Procedure for accidental exposure.
- Safety receipt, inspection, storage, and disposal of the toxin used.

All high-risk operations (e.g., use of toxin at levels considered to be unusually high) should be conducted with two knowledgeable individuals present. Each must be familiar with the applicable procedures, maintain visual contact with the other, and be ready to assist in the event of an accident.

When biological toxins are in use, the room should be posted to indicate “Biological Toxins in Use - Authorized Personnel Only.” Any special entry requirements should be posted at the entrance(s) of the room. Only personnel whose presence is required should be in the room while toxins are in use.

Appendix F
Guidance on Physical and Biological Containment for Recombinant or Synthetic Nucleic Acid Molecule Research Involving Plants

Background

Plants are used as tools by research laboratories to understand natural life processes, values in medicine, and questions about agriculture and environmental health. Their research does not generally pose a hazard but will need to comply with established regulations, standards, and policies. Section III-D-5, Experiments Involving Whole Plants in the NIH Guidelines for Research Involving Recombinant DNA or Synthetic Nucleic Acid Molecules covers the specific requirements for research work involving transgenic or genetically modified plants. Laboratory plant containment and guidelines are set in place to prevent un-intended consequences when they are accidentally released into the environment.

Plant Containment Levels

There are four (4) laboratory containment levels associated with plant research at BU:

- BL1-P is required when working with transgenic or genetically modified plants that are not noxious weeds, do not easily disseminate, and are not detrimental to the environment.
- BL2-P is required when working with plants that are noxious weeds or can interbreed with noxious weeds; transgenic plans that contain the genome of a non-exotic infectious agent, and transgenic plants or plant pathogens that may have a detrimental impact to the environment.
- There are currently no BL3-P or BL4-P experiments at the BU.

Plant Handling and Containment Housing

All laboratories conducting plant research as described above must develop Standard Operating Procedures (SOPs) to describe the process to contain plants, if there is organisms and agents associated with the work, and methods for waste storage and disposal. Physical containment and handling of plants may include the following:

- Laboratory greenhouse or growth room;
- Establish procedures and practices for removal or inactivation of plant reproductive structures (seeds and pollen) as necessary;
- Establish procedures and practices for housekeeping, decontamination, and waste; disposal; all waste generated are disposed as biohazardous wastes;
- Establish personal protective equipment use;
- Establish procedure for transport (as necessary);
- Establish security of the laboratory facility and plant containment;
- Maintain and update inventory of plants.

Routine Practices

BL1-P

- Post appropriate BL1-P placard and information on the door;
- Access to the plant room/ greenhouse shall be limited or restricted;
- All research personnel with access shall be appropriately trained on the SOPs and procedures;
- All research personnel and visitors shall wear and use appropriate personal protective equipment (PPE) before access and follow the appropriate removal and disposal;
- Keep appropriate records and inventory of plants stored and used in the plant room/greenhouse;
- Appropriate inactivation and disposal of plants and plant materials as required;
- Conduct appropriate housekeeping and decontamination routinely or as necessary;
- Follow procedures for accidents or incidents and report to EHS;
- Clean up spills immediately;
- Report any accidental release of plant and plant material into the environment to the IBC and EHS;
- Other procedures established with EHS.

BL2-P

- Follow and establish procedures described above in BL1-P;
- Post appropriate BL-2 placard and information on the door;
- Appropriate engineering control shall be used when working on conducting procedure that could disseminate plant materials such as seeds, pollen, other similar materials;
- Appropriate treatment and disposal of waste materials and plants;
- Assess the plant species, inserted gene(s), reproductive traits, potential for gene flow, and ecological/environmental risks;
- Consider whether the modification could enhance survival, spread, or introduce new traits of concern.

Oversight and Additional Responsibilities

- Obtain IBC approval before starting any work with the above; IBC should ensure reviewers/consultants with plant biotechnology and containment expertise are involved;
- PIs are responsible for compliance, training, and incident reporting;
- APHIS/USDA acquire a permit before conducting any field work; secure all necessary permits and approvals as applicable to the research work;
- EHS will conduct inspections of laboratory and plant containment facility.

For further information, please contact EHS.

Appendix G
Guidelines on Prion and Prion-like Proteins Research

Background

A central biochemical feature of prion and Prion-like diseases is the conversion of normal cellular proteins to an abnormal, misfolded and aggregated form which are toxic to normal cells and potentially can kill the host cell. The pathogenic isoform of Prion protein PrP^{Sc} (named for "scrapie") is the first functionally characterized member of such group of proteins. The PrP^{Sc} is transmissible and self-propagating that causes a group of neurodegenerative diseases called *transmissible spongiform encephalopathies* (TSE) (or prion diseases) which affect humans (e.g., Kuru, Fatal Familial Insomnia and Gerstmann-Sträussler-Scheinker syndrome of humans) and a variety of domestic and wild animal species (e.g., Scrapie of sheep, Bovine Spongiform Encephalopathy (BSE) or "Mad Cow Disease" of cattle and dairy cows, Chronic Wasting Disease (CWD) of deer and moose). These misfold aggregated proteins are highly resistant to inactivation by heat and chemicals and thus require special biosafety precautions.

Prion disease is generally transmitted through accidental inoculation, consumption of raw infected tissues, transplantation or transfusion of contaminated extract during medical procedures. However, prion diseases also has significant genetic link, such as random mutation, hereditary link, or germline mutation. More detail available on the CDC website.

There are reports and experimental evidence that protein aggregates found in number of neurological diseases also cause toxicity in the host cell and have a causal relationship to the neuronal disease. These proteins include: alpha-synuclein protein (Parkinson's disease), tau and beta-amyloid (Alzheimer's disease), TDP-43 (ALS/Lou Gehrig's disease), polyglutamine-containing protein or PolyQ protein (Huntington's disease) and more. Although there is limited information supporting the infectious nature of these proteins, similar to that of the Prion protein, data from experimental models suggests the possibility of human-to-human transmission of these proteins. Therefore, work with these proteins should follow the same safety precautions as work with prion.

Laboratory Safety and Containment Recommendations

Work with prion and prion-like proteins require IBC approval particularly when such work involve a) production and/or use of such proteins; b) any mutated version of those proteins are created; and c) handling of unfixed tissues infected with or containing such proteins. The CDC classifies prions as Risk Group 2 agents requiring BSL2 containment (BMBL 6th edition Section VIII-H: Prion). In the laboratory, prions from human tissue and human prions propagated in animals should be manipulated at BSL2. BSE (Bovine Spongiform encephalopathy) prions can likewise be manipulated at BSL2. However, due to the recent history of transmission of BSE prion to humans, work involving direct contact with BSE-infected specimens may require the use of BSL3 facilities and practices. All other animal prions may be manipulated at BSL2.

Safety Procedures

The following safety procedures are recommended:

- Prion agent must be treated as biohazardous material. Solid wastes must be autoclaved. All equipment that has come in contact with the agent, packaging, containers and all unused portions and derivatives from the agent must be disinfected before disposal.
- Access to the laboratory must be restricted to trained personnel when work is being conducted on tissue.
- All fixed, non-fixed, or frozen tissues that contain the agent must be placed within watertight containers and labeled with the universal biohazard symbol and the notation "Infectious Materials." The use of conventional autoclaving protocol as the sole treatment has not resulted in complete inactivation of prions. Formalin-fixed and paraffin-embedded tissues, especially of the brain, may remain infectious after normal autoclaving. The recommended autoclaving procedure is in table below.
- Personnel working with the agent must not have contact with any animal colonies in the laboratory complex or with susceptible animal species without IACUC approval.
- Personnel must wear gloves and gowns while handling tissues that are potentially contaminated. All protective clothing must be removed before leaving the laboratory. Eye protection is recommended depending on the procedure. Sonication or homogenization of tissues must be performed in a properly certified Class II Biological Safety Cabinet (BSC). Personnel working with these materials should receive specific instruction on the procedures for handling such agents from experienced PI or the EHS. These instructions include a) nature of the protein agent, b) route of transmission, and c) specific hazards associated with the exposure and other laboratory specific issues as appropriate.
- The main precaution to be taken by laboratory members working with prion-infected or contaminated material is to avoid accidental puncture of the skin. Persons handling contaminated specimens should wear cut-resistant gloves if possible. If accidental contamination of unbroken skin occurs, the area should be washed with detergent and abundant quantities of warm water (**avoid scrubbing**); brief exposure (1 minute) to 1N NaOH or a 1:10 dilution of bleach may be considered for maximum safety.

Incident Reporting

The PI must contact the BSO and ROHP regarding spills and accidents that result in overt exposure to tissues. The report must include the following:

- Specification of amount released, time involved, and explanation of procedures used to determine the amount involved.
- Description of the area involved and the extent of employee exposure.
- Report of medical treatment provided.
- Corrective action taken to prevent the reoccurrence of the incident.

Records

ROHP must maintain health records for a period of 30 years. Records must be provided upon request by representatives of the Chief and/or Director of NIOSH.

Inactivation of Prions

Table 1. Prion Inactivation Methods for Reusable Instruments and Surfaces (BMBL 6th edition Section VIII-H: Prion Disease Table 4)

1. Immerse in 1 N NaOH or sodium hypochlorite (20,000 ppm available chlorine) for 1 hour. Transfer into water and autoclave (gravity displacement) at 121ºC for 1 hour. Clean and sterilize by conventional means. [Note: Sodium hypochlorite may be corrosive to some instruments, including autoclaves.]
2. Immerse in a pan containing 1 N NaOH, heat in a gravity displacement autoclave at 121ºC for 30 minutes. Clean-rinse in water and sterilize by conventional means.
3. Immerse in 1 N NaOH or sodium hypochlorite (20,000 ppm) for 1 hour. Remove and rinse instruments with water, transfer to open pan and autoclave at 121ºC (gravity displacement) or 134ºC (porous load) for 1 hour. Clean and sterilize by conventional means.
4. Surfaces or heat-sensitive instruments can be treated with 2 N NaOH or sodium hypochlorite (20,000 ppm) for 1 hour. Ensure surfaces remain wet for entire period, then rinse well with water. Before chemical treatment, it is strongly recommended that gross contamination of surfaces be reduced because the presence of excess organic material will reduce the strength of either NaOH or sodium hypochlorite solutions.
5. 2% Environ LpH® (EPA Reg. No. 1043-118; no longer commercially available) may be used on washable, hard, non-porous surfaces (such as floors, tables, equipment, and counters), items, such as non-disposable instruments, sharps, and sharp containers, and/or laboratory waste solutions (such as formalin or other liquids). This product is currently being used under FIFRA Section 18 exemptions in a number of states. Users should consult with the state environmental protection office prior to use. Items may be immersed for 0.5–16 h, rinsed with water, and sterilized using conventional methods.

Working Solutions 1 N NaOH equals 40 grams of NaOH per liter of water. Solution should be prepared daily. A stock solution of 10 N NaOH can be prepared and fresh 1:10 dilutions (1 part 10 N NaOH plus 9 parts water) should be prepared frequently enough to maintain a fully effective alkalinity.

20,000 ppm sodium hypochlorite equals a 2% solution. Many commercial household bleach sources in the United States contain 6.15% sodium hypochlorite; for such sources, a 1:3 v/v dilution (1 part bleach plus 2 parts water) would produce a solution with 20,500 ppm available chlorine. This relatively easy method provides a slightly more concentrated solution (extra 500 ppm) that should not pose a problem with decontamination procedures or significantly increase chemical risks in the laboratory. Bleach solutions can off-gas and working solutions should be prepared frequently enough to maintain adequate available chlorine levels.

CAUTION: *The above solutions are corrosive and require suitable personal protective equipment and proper secondary containment. These strong corrosive solutions require careful disposal in accordance with local regulations. Sodium, hypochlorite and sodium hydroxide solutions may corrode autoclaves.*

Precautions in using NaOH or sodium hypochlorite solutions in autoclaves: NaOH spills or gas may damage the autoclave if proper containers are not used. The use of containers with a rim and lid designed for condensation to collect and drip back into the pan is recommended. Aluminum should not be used. Persons who use this procedure should be cautious in handling hot NaOH solution (post-autoclave) and in avoiding potential exposure to gaseous NaOH; exercise caution during all sterilization steps; and allow the autoclave, instruments, and solutions to cool down before removal. Immersion in sodium hypochlorite bleach can cause severe damage to some instruments. Neutralization of hypochlorite with thiosulfate prior to autoclaving is recommended to prevent the release of chlorine gas.

Appendix H
Table Summary of Safety Requirements for Biosafety Levels

Safety Guideline	BSL1	BSL2	BSL3	BSL4
Laboratory personnel must wash their hands after handling cultures, removing gloves, and before leaving the laboratory.	Y	Y	Y	Y
Eating, drinking, and application of cosmetics is prohibited.	Y	Y	Y	Y
Personnel must be familiar with basic biosafety procedures, including this manual.	Y	Y	Y	Y
Personnel should wear face protection such as goggles, face shields or other device providing protection against any possible splashes and aerosols.	Y	Y	Y	Y
Pipetting by mouth is prohibited.	Y	Y	Y	Y
All laboratory procedures should be performed to minimize aerosol generation.	Y	Y	Y	Y
Work surfaces must be decontaminated at least daily, after each use for infrequent users, and after any spill of viable materials.	Y	Y	Y	Y
Sharps must be placed in specially designed puncture- and leak-proof sharps containers and disposed of appropriately as medical waste.	Y	Y	Y	Y
Laboratories must be kept neat; good housekeeping procedures must be in place and in regular use.	Y	Y	Y	Y
All medical waste is decontaminated before disposal by an approved decontamination method and/or disposed of as medical waste.	Y	Y	Y	Y
Insect and rodent control programs are instituted.	Y	Y	Y	Y
Laboratory contains a sink for handwashing.	Y	Y	Y	Y
Laboratories are designed for ease of decontamination (e.g., no carpets, sealed surfaces, no unreachable areas, etc.).	Y	Y	Y	Y
Bench tops are impervious to water, moderate heat, and chemicals.	Y	Y	Y	Y
Laboratory furniture must be secured, and spaces between benches, cabinets, and equipment must be accessible for decontamination.	Y	Y	Y	Y
All laboratory windows must be fitted with fly screens.	Y	Y	Y	Y
Laboratory coats or gowns and gloves must be worn.	Y	Y	Y	Y
Autoclaves are required for waste treatment prior to disposal -biohazardous waste.	N	N	Y	Y
Autoclave quality control program is required for use specified above.	Y	Y	Y	Y
Instructions for safety precautions are posted by the Principal Investigator.	Y	Y	Y	Y

Safety Guidelines	BSL1	BSL2	BSL3	BSL4
Animals not involved in the experiment are not permitted in laboratory.	N	Y	Y	Y
Biological safety cabinets are required and must be certified annually.	N	Y	Y	Y
Laboratory personnel require specific training in the handling of pathogenic materials.	N	Y	Y	Y
Safety centrifuge cups are required.	N	Y	Y	Y
Access to facility is limited or restricted during experiments.	N	Y	Y	Y
The universal biohazard symbol must be posted on the access door to the laboratory.	N	Y	Y	Y
Immunization and/or serological testing for agents to be handled may be required.	N	Y	Y	Y
All laboratory procedures must be performed in a properly certified biological safety cabinet.	N	N	Y	Y
Laboratory requires controlled entry, unidirectional air flow, and other special design features.	N	N	Y	Y
Windows must be closed and sealed.	N	N	Y	Y
No material or equipment can leave the laboratory unless it is autoclaved or decontaminated.	N	N	Y	Y
Autoclaves must be located inside the laboratory.	N	N	Y	Y
Access is through an airlock system.	N	N	N	Y

Appendix I
Table Summary of Requirements for Animal Biosafety Levels

Safety Guideline	ABSL1	ABSL2	ABSL3	ABSL4
Access is limited or restricted at the discretion of the laboratory for satellite laboratories or the Attending Veterinarian for all laboratories.	Y	Y	Y	Y
Personnel must wash their hands after handling cultures and animals, removing gloves, and before leaving the facility.	Y	Y	Y	Y
Eating, drinking, and application of cosmetics is prohibited.	Y	Y	Y	Y
Personnel must be familiar with basic biosafety procedures, including this manual.	Y	Y	Y	Y
Personnel should wear goggles or face shields if the possibility of splashes and aerosols exists.	Y	Y	Y	Y
Pipetting by mouth is prohibited.	Y	Y	Y	Y
All procedures should be performed to minimize aerosol generation.	Y	Y	Y	Y
Work surfaces must be decontaminated at least daily with an approved disinfectant, after each use for infrequent users, and after any spill of viable materials.	Y	Y	Y	Y
Sharps must be placed in specially designed puncture- and leak-proof sharps containers and disposed of appropriately as medical waste.	Y	Y	Y	Y
Facilities must be kept neat; good housekeeping procedures must be in place and in regular use.	Y	Y	Y	Y
All medical waste is decontaminated before disposal by an approved decontamination method and/or disposed of as medical waste.	Y	Y	Y	Y
Insect and rodent control programs are instituted.	Y	Y	Y	Y
Doors to animal rooms are kept closed when experimental animals are present.	Y	Y	Y	Y
Facilities are designed for ease of decontamination (e.g., no carpets, sealed surfaces, no unreachable areas, etc.).	Y	Y	Y	Y
Bedding materials from animal cages are removed in a manner that minimizes aerosol production and are disposed of as medical waste.	Y	Y	Y	Y
Instructions for safety precautions are posted by the Principal Investigator.	Y	Y	Y	Y
Facility windows and vents that open must be fitted with fly screens.	Y	Y	Y	Y
Laboratory coats or gowns and gloves must be worn.	Y	Y	Y	Y
Autoclaves are required for waste treatment prior to disposal as biohazardous waste.	N	N	Y	Y

Safety Guideline	ABSL1	ABSL2	ABSL3	ABSL4
Autoclave quality control program is required for use specified above.	Y	Y	Y	Y
Cages are washed prior to release or reuse.	Y	Y	Y	Y
Air is exhausted to the outside without recirculation.	N	Y	Y	Y
Personnel baseline serum samples may be required.	N	Y	Y	Y
Facility personnel require specific training in the handling of pathogenic materials.	N	Y	Y	Y
Safety centrifuge cups are required.	N	Y	Y	Y
Access to facility is limited or restricted during experiments.	N	Y	Y	Y
The universal biohazard symbol must be posted on the access door to the facility.	N	Y	Y	Y
Immunization and/or serological testing for agents to be handled may be required.	N	Y	Y	Y
All procedures must be performed in a properly certified biological safety cabinet.	N	N	Y	Y
Facility requires controlled entry, unidirectional air flow, and other special design features.	N	YN	Y	Y
Windows must be closed and sealed.	N	N	Y	Y
No material or equipment can leave the facility unless it is autoclaved or decontaminated.	N	YN	Y	Y
Autoclaves must be located inside the facility.	N	N	Y	Y
Access is through an airlock system.	N	N	N	Y

Appendix J
Bloodborne Pathogen Standard

Avoiding occupational exposure to human blood, body fluids, and tissues is the primary way to prevent transmission of bloodborne pathogens. The goal of the initial and annual standard precautions training is to present information on how to prevent such exposures by administrative controls, workplace engineering controls, proper work practices, personal protective equipment, and a hepatitis B vaccine immunization program.

Personnel can be exposed to bloodborne pathogens by being stuck with contaminated needles, lacerations from contaminated sharp instruments, or being splashed with blood or body fluids on the mucous membrane of the eye, nose or mouth, or on abraded, non-intact skin (e,g., chapped skin or skin affected by dermatitis). Any direct contact (contact without barrier protection) to concentrated hepatitis B, hepatitis C, HIV, or any other infectious virus in a research laboratory or production facility is considered an exposure that requires clinical evaluation. All employees working with human cell cultures should be offered hepatitis B vaccination and be evaluated if an exposure occurs. Hepatitis B viral infection is one of the most frequent laboratory-associated infections, and laboratory personnel are recognized as a high-risk group for acquiring this infection (Centers for Disease Control and Prevention).

The OSHA Bloodborne Pathogens Standard applies to all employees who might come into contact with blood or other bodily fluids, including:

- Human blood
- Human blood components
- Products made from human blood, or other potentially infectious materials (OPIM) such as the following human body fluids:
    - Semen
    - Vaginal secretions
    - Cerebrospinal fluid
    - Synovial fluid
    - Peritoneal fluid
    - Amniotic fluid
    - Saliva in dental procedures
    - Body fluid that is visibly contaminated with blood and all body fluids in situations where it is difficult or impossible to differentiate between body fluids.
- Any unfixed tissue or organ (other than intact skin) from human (living or dead)
- HIV-containing cell or tissue cultures, organ cultures, and HIV-, HBV- or HCV-containing culture medium or other solutions; blood, organs, or other tissues from experimental animals with HIV, HBV or HCV.

Program Elements

The Bloodborne Pathogens Standard requires that an Exposure Control Plan be written and implemented. The Exposure Control Plan must include the following elements:

- Policies and procedures for elimination, or minimization, of exposure;
- Evaluation of employee exposure potentials;
- Medical surveillance program;

- Routine testing.

The following is a general outline of the BU Exposure Control Plan.

Roles and Responsibilities

PI/ Laboratory Director/Employee Supervisor

The PI/laboratory director/employee supervisor must identify employees under his or her supervision who may be at risk. Upon identifying these employees, the supervisor must:

- Reduce potential risk by providing personal protective clothing and equipment;
- Provide HBV vaccinations at no cost to the employee;
- Complete the OSHA-required Bloodborne Pathogen Training;
- Train the employees;
- Ensure that the BU Exposure Control Plan manual is adopted, reviewed, and updated annually as necessary by the lab and develop an effective hazard communication program;
- Ensure appropriate PPE, engineering controls, such as a biological safety cabinet or sharps container are available and used;
- Develop safe work practices and procedures, as well as internal notification procedures to report accidents;
- Review and evaluate safe alternatives for use of sharps with lab personnel;
- Ensure that all observation cited by EHS during lab inspection are addressed and closed-out;
- In conjunction with the academic department, review the list of at-risk employees on an annual basis to ensure that the list is current.

Laboratory Worker

- Complete required Bloodborne Pathogen Training initially on hire and annually thereafter;
- Complete training provided by the PI;
- Obtain medical clearance from ROHP prior to starting work.
- Use required PPE when working in the lab;
- Report and follow procedures in the event of accidental exposure;
- Accept or decline the HBV vaccination upon offer; complete the declination form when declining. Inform ROHP should the individual decide to accept the vaccination previously offered at any time during employment;
- Review the lab Exposure Control Plan manual.

Environmental Health and Safety

- Inspect laboratories to verify the lab compliance with the OSHA Bloodborne Pathogen requirements;
- Review lab SOPs upon request by the lab;
- Conduct follow up investigation of exposure and incidents, Determine the root cause and provide recommendations to prevent or minimize the incident from recurring;
- Act as a liaison during inspections and visits by regulatory agencies;
- Prepare, maintain and implement the OSHA-required Bloodborne Pathogen Training for completion by the PI and lab personnel.

Key Definitions

Regulated waste includes the following: liquid or semi-liquid blood or other potentially-infectious materials and contaminated items that would release blood or other potentially infectious material in a liquid or semi-liquid state if compressed, items that are caked with dried blood or other potentially infectious materials and are capable of releasing these materials during handling, contaminated sharps, and pathological and microbiological wastes containing blood or other potentially infectious materials.

Sharps waste means any device having acute rigid corners, edges, or protuberances capable of cutting or piercing, including, but not limited to, all of the following:

- Hypodermic needles, syringes, blades, and needles with attached tubing;
- Broken glass items, such as Pasteur pipettes and blood vials contaminated with other medical waste.

At-Risk Employee Identification

The Exposure Control Plan requires each employer to identify, in writing, all tasks, procedures, and job classifications where occupational exposure to blood may occur and to document the methods of compliance that will minimize the potential of occupational exposure.

Incident Reporting

Please see Reporting Laboratory Injuries or Exposures. All incidents reported to ROHP are forwarded to Environmental Health and Safety along with key BU stakeholders and can be found on the ROHP website. Please see Potential Exposure Incident Reports.

Written Policies and Procedures

This manual is intended to act as a primary source of policies and procedures designed to eliminate, or minimize, potential employee exposure to all biological materials, regardless of their hazard level. Employees are required to read and implement all sections of this manual that are relevant to their work environment and be fully familiar with standard precautions. Each PI must further develop site-specific SOPs to address local programmatic needs.

Medical Surveillance

Please see ROHP Requirements for Medical Clearance. The OSHA Bloodborne Pathogens Standard requires that all personnel with potential exposure to bloodborne pathogens be offered immunization against the hepatitis B virus and have a baseline Hepatitis B surface antibody titer on file. Please keep in mind the following requirements:

- Hepatitis B vaccinations must be offered to all workers with occupational exposure. This vaccination must be offered along with the required bloodborne pathogens training within 10 days of initial assignment to a job with occupational exposure.
- Personnel must indicate their consent or declination for the Hepatitis B Vaccine using the ROHP Consent or Declination forms. Both forms are available from the ROHP. The ROHP must retain this form on file for the duration of the employee's employment plus 30 years.

- An employee who declines hepatitis B vaccination may, at any time thereafter, change his or her mind and receive the vaccine. The acceptance statement must be signed at that time.
- The PI/laboratory supervisor must not make participation in a prescreening program a prerequisite for receiving the vaccination.
- The HBV vaccination is available at no cost to the employee.
- ROHP conducts risk assessment of potential exposure through evaluation of bloodborne pathogens such as Hepatitis B, HIV and HCV post exposure.
- BPHC must also be notified of all presumptive exposures.

The PI must ensure that all employees with the potential for occupational exposure participate in a training program provided by EHS at no cost to the employee during working hours. The PI, Supervisor and/ or Biosafety assigns Bloodborne Pathogens training. Please keep in mind the following requirements:

- Training must be given in accordance with the Bloodborne Pathogens Standard upon initial assignment, on an annual basis thereafter, or whenever modification of an existing job description may affect the employee's potential for occupational exposure.
- HIV/HBV research laboratories must ensure that their employees demonstrate proficiency in standard microbiological procedures prior to being allowed to work in the laboratory.
- Training must include a comprehensive discussion of this standard, including epidemiology, symptoms and transmission of bloodborne diseases, the Exposure Control Plan, the uses, limitations of, and procedures for using personal protective equipment, a discussion of the HBV vaccination (including the benefits of vaccination and efficiency of the vaccine to prevent disease), emergency procedures involving blood exposure or contamination and post-exposure follow-up procedures, hazard communication, and a question-and-answer discussion opportunity.

PI Responsibilities for Occupational Health Issues

In keeping with the OSHA Bloodborne Pathogen Standard, this policy requires annual standard precautions training, a hepatitis B immunization program, and a post-exposure medical management program.

It is the PI's responsibility to ensure that researchers, technicians, students, or volunteers who work in the laboratory and who have contact with animals, infectious agents, or bloodborne pathogens are medically evaluated prior to starting work and that anyone working with bloodborne pathogens is referred to ROHP for medical surveillance services such as Hepatitis B titer screening and hepatitis B vaccination as indicated in compliance with the Bloodborne Pathogen Exposure Policy. It is the PI's responsibility to ensure that any person present in a BU laboratory who has an incident involving potential exposure to an infectious agent is offered ***immediate*** access to a medical evaluation by or through the ROHP (listed below). An immediate evaluation is important, as efficacy of post-exposure medication for HIV and other infectious agents may be less effective if the initiation of treatment is delayed.

Personnel working with non-human primates or their tissues may also require evaluation for post-exposure prophylaxis against Herpes B virus.

If at any time, an employee has a potential exposure to bloodborne pathogens, they MUST immediately contact the ROHP.

ROHP Information

ROHP is located on the Boston University Medical Campus at 72 East Concord Street, 8th Floor, Room 825. Our normal hours of operation are Monday through Friday from 8:00am to 4:30pm. Our phone number is (617) 358-7647 (ROHP) and is available and supported by medical staff 24 hours per day/7 days per week to triage and evaluate laboratory exposures and related illnesses. Based on injury severity, location and time of day, ROHP will refer people to the appropriate health care location.

For lab exposures (e.g., needle stick, bite, cut, scratch, splash) involving animals or infectious agents on the Medical Campus or Charles River Campus, **call the ROHP 24/7 hour number (1-617-**358**-ROHP (7647) or 4-ROHP (7647) if calling from a Medical Campus location)** to be connected with the BU ROHP medical officer.

For unexplained symptoms or illness **call the ROHP 24/7 hour number (617) 358-ROHP (7647), or 4-ROHP (7647) if calling from a Medical Campus location)** to be connected with the BU ROHP medical provider.

If referred to a health care location under any of these scenarios, always inform the physician of your work in the laboratory and the agent(s) that you work with. If you have been given a wallet-size medical surveillance card, please provide it to your treating healthcare provider.

Appendix K
Working Safely with Animals

Working with animals poses potential additional health and safety hazards that require extra precautions. The specific requirements will depend on the types of activities (e.g., surgery, feeding animals, use of anesthetic agents, etc.) and the specific species used. Personnel should follow the following guidelines:

- Follow the specific requirements established in the IACUC-approved protocol and the facility requirements.
- Follow procedures established by the Animal Science Center and IACUC for ABSL2 as appropriate.
- Wash hands after handling an animal, anything that an animal has touched, or before exiting the animal facility. The most common way of contracting an animal-transmitted infection is placing the infectious material directly into the mouth.
- Never smoke, drink, or eat in an animal area.
- Wear protective clothing as recommended/required by the facility for the species and operations: Protective laboratory coats, gowns, or uniforms are recommended to prevent contamination of personal clothing; Protective clothing helps prevent potentially contaminated material from leaving an animal area; and do not wear the protective clothing outside of the animal area and do not take protective clothing home;
- Use the personal protective equipment (PPE) recommended/required for the species and operations.
- Workers shall wear the appropriate PPE (e.g., gloves, face shields, masks, and respirators) when required and follow their supervisor's instructions scrupulously.
- Gloves are worn to prevent skin contact with contaminated, infectious and hazardous materials, and when handling animals. Gloves may also be worn to protect the items being handled from contamination.
- Gloves and personal protective equipment should be removed in a manner that minimizes transfer of infectious materials outside of the areas where infectious materials and/or animals are housed or are manipulated.
- Persons must wash their hands after removing gloves, and before leaving the areas where infectious materials and/or animals are housed or are manipulated.
- Eye and face and respiratory protection should be used in rooms containing infected animals, as dictated by the risk assessment.
- Participate in the ROHP medical surveillance program (See Appendix P) that provides medical evaluations, testing, immunizations, and periodic screenings for allergies, infections, and other medical problems related to animal exposure.
- Seek medical attention promptly when injured. Follow the specific recommendations for the facility.
- Workers engaged in work involving vertebrate animals should inform their physician of their work when seeking treatment for illness, even if uncertain whether the illness is work related. All animal care workers are provided wallet-sized agent identification cards. The card indicates the card carrier works in a laboratory setting at Boston University and may be exposed to hazardous materials. The card also contains ROHP contact information in the event the physician should choose to seek further information on potential occupational exposure.

Physicians need such information to make an accurate diagnosis because many animal-transmitted diseases have flu-like symptoms.

- If there is any possibility of work-related illness or disease, the ROHP must be notified immediately at (617) 358-ROHP (7647).
- Get the appropriate training and contact a supervisor with any questions.

Basic Safety for the Necropsy of Infected Animals

- Ensure that the necropsy of infected animals is carried out in biological safety cabinets, downdraft table or designated necropsy space by trained personnel;.
- Wear gloves and a disposable or facility lab coat or disposable Tyvek™ jumpsuit over laboratory clothing;
- Use other PPE recommended by the facility for the infectious agents present. This may include double gloves, dedicated shoes or shoe covers, eye protection, surgical mask or respiratory protection, hair bonnet, and/or specialized PPE for biocontainment, such as PAPR, back fastening gowns, or positive pressure suits;
- Pin down or otherwise fasten small animals to metal in a tray;
- Before and after necropsy, disinfect the necropsy table, inside of the BSC, and other potentially contaminated surfaces with an approved disinfectant;
- Upon completion of necropsy, place all potentially biohazardous materials in suitable containers and then sterilize the materials;
- Segregate contaminated mixed waste and store for appropriate disposal;
- Place contaminated instruments in a bath that contains a suitable disinfectant;
- Follow the facility requirements for sterilization;
- Clean contaminated gloves in disinfectant before removal from the hands;
- Wearing gloves is not a substitute for handwashing; wash hands after necropsy and carcass disposal;
- Dispose of protective clothing and shoe covers in appropriate containers;
- Follow the facility’s guidelines for the disposal of animal carcasses.

Appendix L
Procedures for Working in Animal Biosafety Level 2 (ABSL2)
Rodent Biosafety Level 2 Procedures at BU

PLEASE NOTE: Before starting any Animal Biosafety Level 2 (ABSL2) work, a PI must 1) obtain IBC and IACUC approval and 2) make appropriate animal housing arrangements with the ASC director.

The purpose of this Standard Operating Procedures (SOP) is to provide guidance to those individuals working in rooms in which animals involved in chemical and biological hazards determined to be ABSL2 are housed and rodents are exposed to biological hazards determined to be Animal Biosafety Level 2 (ABSL2).

Overview

Access to the room where the work with animals is to be conducted is restricted. Laboratory personnel must have training in aseptic micro-isolator techniques, when applicable, and use of biological safety cabinets, in addition to specific safety training in handling the pathogenic and/or chemical agent(s) with which they are working.

Research and Animal Science Center Facility (BUMC or CRC) personnel should receive appropriate immunizations or tests for any agents handled or potentially present in the room prior to initiating the ABSL2 portion of their project. Procedures must be conducted in a Class II BSC.

Definitions

ABSL2: Animal Biosafety Level 2 includes pathogenic agents of moderate hazard potential (CDC Biohazard Class 2) and chemical hazard agents of moderate hazard potential.

PPE: Personal protective equipment

EHS: Environmental Health and Safety

Parenteral: Taken into the body or administered in a manner other than through the digestive tract, as by intravenous or intramuscular injection.

CCL: Chemical Containment Level

Assumptions

It is the responsibility of BU Environmental Health & Safety and ASC to ensure that all necessary safety training is provided to research and BUASC staff prior to those staff working with biohazards. Required training modules and their completion is documented in SciSure.

Equipment and Supplies Required:

- Biohazard Stickers for Cage Cards;

- Biohazard Signage for Rack(s) housing ABSL-2 cages;
- Biohazard Signage for posting on outside of door containing ABSL-2 cages;
- Approved disinfectant – 0.5% hydrogen peroxide solution (Peroxigard™);
- Biosafety Cabinet - Class 2 with current (unexpired) service record;
- Individually Ventilated Caging with either single-pass ducted exhaust or HEPA-filtration prior to recycling exhausted air.

Safety Requirements

- Access to the room where the work is conducted is restricted;
- Laboratory personnel must have training in proper microisolator techniques and use of biosafety cabinets in addition to specific safety training in handling the microbial or chemical agent(s) with which they are working;
- Research and BUASC personnel will receive appropriate immunizations or tests, as prescribed by the BU Research Occupational Health Program, for applicable agents handled or potentially present in the room prior to initiating the ABSL-2 portion of that project;
- Cage cards for ABSL-2 animals must have a biohazard sticker;
- ABSL-2 cages are restricted to a dedicated rack with a biohazard signage.
- Procedures must be conducted in a Class 2 biological safety cabinet.

Personnel Protective Equipment (PPE)

- Don designated PPE for handling animals or cage contents as depicted on ABSL-2/CCL signage. All required PPE is located on the supply cart (or cabinet) in the corridor outside of ABSL-2 rooms.
- ABSL-2/CCL-2/3 PPE are required in these rooms even if ABLS-1 or CCL-1 cages and animals are to be handled.

Personal Protective Equipment

Minimum PPE:

- Solid front gown:
- Hair cover
- Shoe covers
- Mask
- Double gloves

In addition:

- N95 may be required
- Face shield or other specific eye or face protection may be required

Equipment and Supplies

- Biohazard stickers for cage cards

- MB-10 (chlorine dioxide) disinfectant or Virkon-S

Responsibilities

It is the responsibility of EHS and ASC to ensure that all necessary project-specific safety training is provided to research and LASC/LACF staff prior to any project being initiated. It is also the responsibility of EHS to provide documentation to the LASC/LACF of such training.

The PI and individuals working in the facility are responsible for ensuring they have received proper training and that they are adhering to this SOP, as well as to posted precautions and guidelines in the facilities.

Procedures and Instructions

Entry – refer to required PPE instructions on the entry door. Change gloves between handling animals or cages of different ABSL designation.

- Remove the lab coat worn in the ASC facility and hang it on the garment rack provided outside of designated ABSL2 animal space.
- PPE *must* be worn while working in ABSL2 animal housing and procedure space. PPE is provided just outside specific ABSL2 rooms. Don designated PPE prior to entry into the ABSL2 areas.
- Proceed into the designated ABSL2 room using an access card or key.

Conducting a Procedure

General Information:

- For CCL procedures, refer to SAF 10.
- Investigators using the room will be assigned a cubicle and/or rack and shelf/shelves where their animals will be housed.
  - A cubicle or rack may hold cages belonging to more than one investigator.
  - Biohazard projects are not housed in cubicles that house ongoing chemical hazard projects.
- All animal work should be conducted within a class 2 biological safety cabinet. Open ABSL-2 cages in the class 2 biological safety cabinet.
- Only hypodermic needles and syringes are used for parenteral injection, aspiration of fluids from laboratory animals, and bottles with plastic/rubber diaphragms. Only luer-lock syringes or disposable needle syringe units (i.e., the needle is integral to the syringe) are used for the injection or aspiration of infectious fluids. Needles are not to be bent, sheared, replaced in the sheath or guard, or removed from the syringe following use. The needle and syringe must be promptly placed in a puncture-resistant “Sharps” container.
- Always spray and wipe down all interior surfaces of the class 2 biological safety cabinet with approved disinfectant before and after working. Allow appropriate contact time of the disinfecting agent prior to wiping the surfaces with disposable towels. Time may differ depending on disinfectant in use. Do not spray the top/front grille of the class 2 biological safety cabinets (see photo below). Discard used disposable towels in the ABSL-2 designated waste container (biohazard waste).

ASC personnel will perform daily health checks of animals on studies involving infectious agents if the animals are housed on racks not held in cubicles.

- ASC personnel will perform daily health checks of animals on studies involving chemical agents by viewing the animals through the window of the isolation cubicle.

- The ASC will notify PIs of any animal health issues.
- All animal work will be conducted within the confines of the Class II BSC.
  - Always open animal cages in the Class II BSC using aseptic micro-isolator technique.
- Hypodermic needles and syringes are used only for parenteral injection or aspiration of fluids from laboratory animals and bottles with plastic/rubber diaphragms.
  - Only needle-locking syringes (e.g. luer locking) or disposable needle syringe units (e.g., the needle is integral to the syringe) are used for the injection or aspiration of infectious fluids.
  - Needles should not be bent, sheared, replaced in the sheath or guard, or removed from the syringe following use.
  - The needle and syringe should be promptly placed in a puncture-resistant sharps container.

- Always spray or wipe down all interior surfaces of the Class II BSC with MB-10 or Virkon-S **before** and **after** working in the hood. Allow 10 minutes of contact time prior to wiping the surfaces with disposable towels. Do not spray the top grille of the Class II BSC (this is where the filter is located). Discard used towels in the waste container.
- Immediately following infection of animals with pathogenic agents, place a biohazard sticker on their cage card(s) (ASC provides these specific stickers). Fill out the following information on the biohazard sticker for cage cards:
  - PI name
  - 24-hour contact phone number
  - Protocol number
  - Pathogenic agent
  - Dose per animal
  - Date(s) infected
  - Husbandry by PI or ASC (circle one); Always spray and wipe down all interior surfaces of the class 2 biological safety cabinet with approved disinfectant before and after working. Allow appropriate contact time of the disinfecting agent prior to wiping the surfaces with disposable towels. Time may differ depending on disinfectant in use. Do not spray the top/front grille of the class 2 biological safety cabinets (see photo below). Discard used disposable towels in the ABSL-2 designated waste container (biohazard waste).
- Report all spills and accidents that result in overt exposure to infectious materials to a ASC Supervisor and the ASC office, (617) 638-4086 or LACF at (617) 353-5415. Refer to Spill assistance website for emergency phone numbers for reporting spills and possible exposures.

Removal of Dirty Cages and Bottles from Room

Biological agents

- Place a cage inside the BSC, remove the water bottle from the cage, and replace the lid.
  - Put soiled cage, wire lid, and bedding in a semi-clear biohazard autoclave bag and place on a cart.
  - Put water bottles in a separate, semi-clear biohazard autoclave bag on a cart.

- Closure ties are stored adjacent to the biohazard autoclave bags on the supply rack in the corridor outside rooms and in rooms.
- **Do not use** autoclave tape to close the bag.

- Thoroughly spray the exterior of all bags with MB-10 or Virkon-S after closing the bag with a nylon tie, beaded tie, or twist tie. Spray all surfaces and wheels of the cart(s).
- Cart(s) should be moved to the soiled side of the cage wash room. Put soiled cage, wire lid, plastic top, and bedding into ABSL-2 cage cart for autoclaving. Autoclave bags and closure ties are inside the room in which ABSL-2 cages are housed.
- Do not use autoclave tape to close the bag.
- Research staff are to leave their bags of dirty ABSL-2 caging in the designated 'dirty cage' space inside the animal housing room. This area is designated by a sign. BU ASC staff will remove dirty caging daily for autoclaving and processing.
- Thoroughly spray the exterior of all bags with approved disinfectant after closing the bag with a nylon tie, beaded tie, or twist tie. Spray all surfaces and wheels of the cart(s).
- Cart(s) with soiled ABSL-2 caging/water bottles are transported to the designated ASC autoclave for sterilization prior to cage wash.

Chemical agents

- Place a cage inside the BSC, remove the water bottle from the cage, and replace the lid.
    - Put soiled cage, wire lid, and bedding in a red biohazard bag and place on a cart.
    - Put water bottles in a separate red biohazard bag on a cart.
- Closure ties are stored adjacent to the biohazard autoclave bags on the supply rack in the corridor outside W-838/839 and in both rooms.
- **Do not use** autoclave tape to close the bag.
- Thoroughly spray the exterior of all bags with MB-10 after closing the bag with a nylon tie, beaded tie, or twist tie. Spray all surfaces and wheels of the cart(s).
- Cart(s) should be moved to the soiled side of the cage wash room on the 8th floor of W Building (W-8).

Disposal of carcasses

- Any dead animals must be removed from their cage (while in the BSC) and placed in a small, leak-proof red biohazard bag.
- Place a sticker with animal identification information on the outer bag, then thoroughly spray the bag with MB-10 or Virkon-S and place in the refrigerator. A cage card with a sticker showing the same animal identification information will be placed on the cage. Any deceased animals must be removed from their cage (while in the biosafety cabinet) and placed in a small red biohazard bag. Place a sticker with a dead animal number on the outer bag and place the bag in the refrigerator in the ABSL-2 /room. A beige cage card with a sticker showing the same dead animal number will be placed on the cage.
- The ASC will remove carcasses for incineration disposal three days after they are found in the refrigerator. BU ASC will remove carcasses for incineration within three days after they are found in the refrigerator.

<u>Exiting Procedures</u>

- When work is complete and the BSC and all other work spaces have been decontaminated with MB-10 or Virkon-S, outer gloves should be removed and placed in the biohazard waste container. BU ASC will remove carcasses for incineration within three days after they are found in the refrigerator.

- Discard disposable PPE (lab coat, gloves) into the red biohazard trash receptacle in the hallway outside the room.
- After leaving the ABSL-2 room and having discarded your PPE, sanitize your hands using the alcohol dispenser located on the wall immediately outside the ABSL-2 room. Wash your hands at the closest hand-washing sink with soap and running water.
- If handling other animals or animal-contaminated material, don new PPE to enter other vivarium housing spaces before proceeding from a dirty to a clean animal housing space.
- To exit the room, open the door and remove one shoe cover, stepping over the room threshold into the hallway with that foot.
- Remove the shoe cover from the other foot as it is brought into the hallway but before stepping into the hallway with the second foot
- Remove gown from the shoulders, turning it inside out.
- Discard disposable face protection, hair cover, mask, gown and gloves in the red biohazard trash receptacle in the hallway outside ABSL2 room.
    - After leaving ABSL2 room and discarding PPE, hands should be washed using the alcohol hand sprayer located on the wall immediately to the left of the doors to each ABSL2 room

Reference: Biosafety in Microbiological and Biomedical Laboratories (6th edition, 2020)

Appendix M
ROHP Medical Surveillance Program

BU provides medical surveillance to all researchers and research support staff (employees and students) who face workplace risks. The program is designed to evaluate potential health hazards associated with research and development activity with recombinant DNA, bloodborne pathogens, zoonotic diseases associated with laboratory animals, and hazardous biological agents and chemicals. The program also has requirements for Personnel at the NEIDL enrolled in the Entity's Tier 1 Suitability Assessment Program. The details of the medical surveillance program are provided below:

Objectives and Scope

The Medical Surveillance Program developed and implemented by the ROHP has the following objectives:

- Determine the initial and periodic medical surveillance requirements for those personnel that perform research and those groups that support research such as animal care workers, EHS, Public Safety, and Facilities as determined by Work Environment Requiring Medical Surveillance intervention. Refer to the ROHP website to identify who requires a medical clearance.
- Define medical surveillance requirements based on the work environment, occupational exposure risk, and access requirements for each position.
- Determine whether the employee or applicant can safely perform the essential functions of the job for which employment has been offered.
- Determine accommodation, if necessary, for an employee or applicant to perform the functions of the job in a safe and effective manner.
- Establish a baseline for comparison with future periodic evaluations.
- Establish a procedure for performing additional medical surveillance in support of the IBC and IACUC when new protocols are reviewed and changes in the job function or role, exposure to hazardous materials and access requirements for researchers arise.

Clinical Services

The clinical services provided as part of the Initial and Periodic medical surveillance profiles include questionnaires, physical examinations, laboratory testing, and screenings dictated by the job position, exposure type, work environment and access/location requirements for each position. See Table 1 below for a complete listing of components based on researcher's job activities assigned by their PI or supervisor.

Returning to Boston University Research:

- From leaves of absence of more than one year or previous employment at BU more than one year ago, a complete medical surveillance is required.
- From leaves of absence of less than one year or previous employment at BU less than one year ago, completion of an abbreviated health questionnaire is required. No other examinations are needed unless health risks are indicated in the abbreviated questionnaire.

Returning to BU Research that require access to the NEIDL:

- From leaves of absence of more than one year or previous employment at BU more than one year ago, a complete medical surveillance is required.

- From leaves of absence of less than one year or previous employment at BU less than one year are required to complete an abbreviated health questionnaire and undergo mental health and drug screens. No other examinations are required unless health risks are indicated in the abbreviated questionnaire.

PIs Supervisors and Managers

PIs and managers are responsible for assigning job activities and ensuring the student, employee or researcher has completed their entry and periodic medical health requirements prior to beginning their work. Managers, sponsors and PI's that are hosting a visitor should refer to the ROHP website for requirements. In addition, refer to the Summer Student Visitor Guidelines for their requirements. A Visitor Job Risk Assessment must be completed by the Sponsor, EHS or Biosafety officer and submitted to ROHP@bu.edu. Please contact ROHP for the Visitor Job Risk Assessment.

ROHP Services

Reviews the candidate's ROHP Health Questionnaire and Job Activities/Job Risk Assessment to:

- Define the medical surveillance required based on occupational exposure and potential risk of the work environment for the candidate's assigned work. Establish a baseline medical history for the candidate for ongoing medical surveillance, and
- Assess the candidate's ability to safely perform the functions of the position.
- Determine if additional medical documentation is needed, i.e. immunization records, tuberculosis screens, etc.
- Offer animal allergy screening and reproductive counseling services as needed;
- Employees are offered the necessary vaccines at no cost based on their work risk assessment. Appropriate vaccinations are offered to all candidates who access laboratories that contain an agent with an available vaccine. Please see current CDC Vaccine Information Sheets for information regarding risks and benefits of vaccines.
- Review results of all testing, screenings, examinations and communicates results of all testing, screening and exams to employee/student. A consultation is conducted with the ROHP Medical Director if medically indicated.
- Notify appropriate personnel of examination outcome.
- A written opinion identifying what a researcher or research support individual is medically cleared for will be generated by ROHP once the medical requirements for the specific job activity have been met. Some researchers will require annual or other medical requirements depending on their position or job activities assigned. They will be notified of these requirements and medical clearances in SciSure, the research compliance software system, and are expected to complete these requirements as indicated to meet compliance with their IBC or IACUC protocol and/or BU's policies. These medical clearances will be viewable to the researcher, supervisor, and key stakeholders (i.e. EHS, IBC and IACUC).
- Issue a medical surveillance wallet card to personnel who may be exposed to hazardous materials while working in a research or animal care facility. The card contains ROHP contact information and is used to facilitate prompt medical attention and appropriate medical care in the event the card holder should experience symptoms or illness while away from Boston University that may be related to activities or exposures in a laboratory research environment;
- Provide safety and exposure response information and BU resources for employees and students.

- Refer employees and students to the Agent Information Sheets (AIS) list for any agent that could be a potential for a laboratory-acquired infection (LAI. These provide education regarding the agent including modes of transmission, prevention of disease and signs, and symptoms of disease.

Recordkeeping

Refer to the BU Medical Records and Health Information Guidance. All medical information that is confidential and will be maintained in the secure BU Occupational Enterprise Health electronic medical record system. For more information, please refer to the BU Medical Records and Health Information Guidance. Electronic medical records will also be maintained for all personnel assessed by ROHP. Employees and students can view their immunization record and lab work as well as upload important required documents in the Boston University Employee Health Connect Portal. See table below:

TABLE 1. MEDICAL SURVEILLANCE – VACCINES AVAILABLE BASED ON JOB ACTIVITIES

Category	Job Activity	Available Vaccines *
Animal	Bats	Rabies
Animal	Ferrets	Influenza
Animal	Non-human primates	Influenza, Measles
Animal	Pigs	Influenza
Animal	Wild birds	Influenza
BSL2	Haemophilus influenzae (type B) ONLY	Hib
BSL2	Human source material - potentially infectious with Hepatitis B	Hepatitis B
BSL2	Human subject/ patient care spaces	MMRV, Tdap, Hep B, Influenza, COVID
BSL2	Cowpox virus	JYNNEOS
BSL2	Influenza virus	Influenza
BSL2	Neisseria meningitidis	Menigococcal ACWY and B
BSL2	Polio virus	Polio
BSL2	Respiratory syncytial virus	RSV
BSL2	SARS CoV2	COVID
BSL2	Streptococcus pneumoniae	Pneumococcal
BSL2	Vaccinia Virus: Replication-competent (i.e. Western Reserve, Vaccinia, NYCBOH strain)	JYNNEOS
BSL3	Chikungunya virus	Chikungunya
BSL3	Japanese Encephalitis	JE
BSL3	Mpox Clade 1 (select) &/ or Clade 2 (non-select)	JYNNEOS
BSL3	Yellow Fever Virus	YF
BSL4	Ebola Zaire	ERVEBO
Toxin	Diphtheria toxin	Tdap or Td
Toxin	Pertussis toxin	Tdap

**Updated 11/14/2025 based on current CDC VISs/Vaccines and Immunizations*

Appendix N
Laboratory and Equipment Decontamination Procedures

Decontamination of Lab Space and Equipment Punch List

1. Designate a "Move Coordinator". The coordinator will work with EHS and other stakeholders to facilitate and coordinate the move.
2. Contact an outside vendor for the decontamination of biological safety cabinets (tissue culture hoods).
3. Have appropriate personal protective equipment available (lab coat, gloves, eye protection).
4. Dispose of old chemicals and all other chemical waste as hazardous waste. Notify the Environmental Manager at (617) 358-7840 at BUMC or (617) 353-4094 at CRC upon termination of a hazardous waste accumulation area or with any questions on this issue.
5. Decontaminate all equipment that is either to be moved or left behind.
6. Contact EHS regarding the discarding of equipment.
7. On the decontamination certificate, list all decontaminated equipment by room (one sheet per room).
8. Small pieces of equipment can be decontaminated and boxed by lab personnel.
9. Any working equipment to be left behind without a new owner must be reported to Facilities Management and EHS.
10. Contact the Radiation Safety for the guidance on decontamination and moving of radiological materials and work spaces.
11. Decontaminate all labs, including fume hoods, the outside of tissue culture hoods, cold/warm rooms, darkrooms, etc.
12. If perchloric acid was used in a fume hood contact EHS at (617) 358-7840 at BUMC or (617) 353-4094 at CRC. This will require a special procedure.
13. Fill out one decontamination sheet for each room, tape one copy to the outside of the lab door (if it is a section of a lab, tape to bench), send one copy to the Biosafety Office, and keep one copy for the records.
14. Disinfectants: the most common are 10% freshly diluted bleach (leave on for 20-30 minutes, then wash off), 70% ethanol, or isopropanol. Phenolic agents are not recommended.
15. If refrigerators, freezers, incubators, etc., are to be moved with content inside, make sure the content is well protected from sliding, breaking, etc.
16. The Move Coordinator must ensure the following emergency procedures are covered: chemical spills, biological spills, fire, and personal injuries, such as slips, falls, cuts, etc.
17. Protective clothing and spill absorbent materials must be on hand.
18. Follow and complete the Laboratory or Equipment Decontamination certification form. EHS will inspect the laboratory space or equipment to ensure they have been appropriate cleaned and decontaminated.
19. EHS will affix a decontamination sticker on equipment that had been properly cleaned and disinfected. The equipment will be moved out within 15 days that the sticker is issued. The equipment will not be moved if the 15 days issuance lapses. EHS will inspect the equipment again and issue a new sticker.
20. Biohazard sticker will be removed once the equipment has been properly decontaminated.

The Environmental Health and Safety Laboratory or Equipment Decontamination Certification form can be found here.

Appendix O
Laboratory Decommissioning and Relocation Procedures

Purpose and Applicability

It is the policy of Boston University that laboratory decommissioning take place prior to the relocation of any laboratory space or upon vacating laboratory space or leaving either institution. In addition, safe-moving practices must be adhered to at all times. This policy is intended to minimize research and clinical lab downtime due to moving of a laboratory, and to protect contractors, laboratory personnel, and any other personnel involved in the process from laboratory hazards. This policy applies to all Boston University employees and tenants occupying laboratory space within Boston University buildings.

Definitions

Abandoned Laboratory: A clinical or research laboratory that is left vacant by a Principal Investigator (PI) or Laboratory Director and his or her laboratory staff, and has laboratory materials (biological, surplus chemical, radioactive), equipment or waste that has not been disposed of.

Biological Materials: All human, plant, and animal pathogens; all human blood, blood components and products, tissues and body fluids; all human and animal cultured cells; all infected animals and animal tissues; all cultures/stocks of biological agents, including recombinant DNA materials; and all biological toxins. Also includes biomedical waste and physically dangerous (sharp) waste.

Decommissioning: The process whereby a Principal Investigator or Laboratory Director and his or her laboratory staff decontaminate existing laboratory space and make a clinical or research laboratory safe prior to vacating the space.

Decontamination: The process whereby the PI or Laboratory Director and his or her laboratory staff clean and disinfect laboratory surfaces and equipment so they are safe to handle.

Roles and Responsibilities

The PI or Laboratory Director is responsible for the complete decommissioning of the laboratory space prior to vacating the laboratory. In cases where an abandoned lab is identified, the department that the PI or Laboratory Supervisor reported to will be responsible for the decommissioning and all costs associated with the process.

EHS will distribute this policy and attachments and advise PIs, Laboratory Directors, and laboratory personnel on how to implement the various aspects of the policy. They will also verify that a lab has been appropriately decommissioned before a Principal Investigator or Laboratory Director may leave or move his or her laboratory.

The Move Coordinator for the laboratory is appointed by the PI Laboratory Director and is responsible for coordinating the laboratory decommissioning and move. The Move Coordinator is the primary contact with EHS.

Other personnel (Facilities, moving personnel, and contractors) should be aware of this policy and should not handle laboratory materials, equipment, or waste unless instructed to do so by their supervisor and/or EHS.

Procedures: Preparation

Prior planning is key to a successful laboratory decommissioning and move. Preparation and communication with EHS will be a major factor in minimizing delays, protecting property against damage and loss, and most importantly, reducing the potential for personal injury. Contact EHS at (617) 358-4094 (CRC) or 617 958-7840 (BUMC) with any questions or for assistance.

Procedures: Waste Disposal

All biological waste and hazardous waste must be disposed of according to current EHS policies and procedures as outlined in the EHS *Policy Manual*. All radioactive waste must be disposed of according to Radiation Safety policies and procedures. Boxes and trash must not be left in corridors. Prior arrangements for regular trash must be made with Custodial or Environmental Services.

Chemical waste must be labeled with hazardous waste stickers regardless of whether or not they are labeled from the manufacturer.

Unwanted, unopened, or uncontaminated chemicals should be offered to other labs that may be able to use them before the chemicals are considered for disposal.

Any unknown chemical must be identified and labeled as hazardous waste. For chemical unknowns that cannot be identified by the Principal Investigator, Laboratory Director or laboratory personnel, the laboratory may be assessed a service fee for hazardous waste analysis prior to disposal.

Darkroom tanks must be drained and the contents disposed of as hazardous waste. Empty compressed gas tanks must be returned to the distributor prior to the move. Mercury thermometers must be disposed of as hazardous waste, and vacuum pumps must be drained of oil and the oil disposed of as hazardous waste.

Procedures: Decontamination

All laboratory bench-top surfaces must be decontaminated prior to vacating the laboratory, and all laboratory equipment that is either remaining in the laboratory or being moved to a new laboratory must be decontaminated if potentially contaminated with biological, chemical, or radioactive materials.

Lab equipment requiring decontamination includes, but is not limited to, animal cages, centrifuges, fermenters, fish tanks, incubators, water baths, refrigerators, and freezers (if not moving intact).

Fume hoods must be decontaminated. Contact the Industrial Hygienist at (617) 358-7840 for decontamination and certification advice. Notify the Industrial Hygienist if there is any current or past practices that may reveal potential problems. Certain chemicals such as perchloric acid and mercury may remain on surfaces or equipment or in building systems.

It is a routine practice that **Biological Safety Cabinets** (BSCs) and glove boxes be cleaned and surface decontaminated with an appropriate chemical disinfectant before and after each use by the laboratory. BSCs that have been used with potentially infectious or infectious materials must first be gas decontaminated using gaseous sterilant before being moved or disposed. This is performed by a qualified vendor that is trained and licensed to perform the work. BSCs that have only been used for non-infectious materials since their initial installation, may be exempt from this gaseous decontamination based on the certification provided by the primary user. An attestation form must be completed and submitted to the Biological Safety Officer (BSO) for review and approval, utilizing the Attestation for Use of Biological Safety Cabinet (BSC) Form. Additional information on use of the BSC may be found on EHS's Biological Safety Cabinet Use page.

Contact the BSO at (617) 358-7840 to discuss decontamination or moving of the BSC. BSCs must be recertified after they are moved to their permanent locations.

Procedures: Designation of New Laboratory Space

The PI or Laboratory Director must inform EHS of any new laboratory space, so that the appropriate safety signage may be provided.

The PI is responsible for notifying all applicable Boston University research committees and outside agencies, as necessary, of the move to new laboratory space. Research projects approved by the IBC must have updated laboratory location information. USDA Veterinary Service or Plant Service permits are laboratory site specific, as are CDC Select Agent registration permits. Contact the Biological Safety Officer at (617) 358-7840 for assistance.

Procedures: Packing and Moving Laboratory Materials

Laboratory personnel are responsible for collecting all packaging items needed before the move date. Carts, plastic bags, toweling, or other cushioning, absorbent materials, sealable plastic or plastic-lined boxes, labels (e.g. Fragile, Universal Biohazard, ID, Location, Caution, Radioactive Material), sturdy tape, and spill kits should be readily accessible. Each container or piece of equipment must be labeled. Labels must identify the agent, hazard, and necessary precautions.

The PI or Laboratory Director is responsible for establishing safety and emergency procedures for all phases of the move. Potential emergencies include material spills, fires, slips and falls, and cuts. Protective clothing and spill absorbent materials must be available during packing, moving, and unpacking.

Procedures: Packing and Moving Laboratory Chemicals

In order to minimize the amount of chemicals that need to be packed and moved, new chemicals should be ordered only as necessary and in small quantities. Laboratory personnel should plan in advance to

minimize the inventory of liquid volume and weight of materials being moved. In addition, reduction of active materials should be planned the week prior to the move. Laboratory chemicals must be packed and moved by an outside contractor approved by EHS. Prior to the packing and moving, laboratory personnel are responsible for labeling each chemical container with the chemical identity.

Compressed gas tanks that are to be moved must have regulators removed and caps secured prior to moving. If possible, have old tanks collected prior to a move and arrange for future tanks to be delivered to the new location.

Thermometers must be removed from refrigerators, water baths, and incubators prior to equipment moving.

Vacuum pump oil must be drained from pumps prior to equipment moving.

Procedures: Packing and Moving Biological Materials

Biological materials must be appropriately packed and moved by the laboratory personnel. Regulated materials and biological materials include all human, plant, and animal pathogens; all human blood, blood components and products, tissues and body fluids; all human and animal cultured cells; all animal carcasses and unfixed animal tissues; all cultures/stocks of biological agents including recombinant DNA materials; and all biological toxins.

Proper packaging consists of a primary sealed container placed within a secondary sealed, unbreakable container, with enough absorbent material in between to contain and absorb any spill. ***Some examples of proper packaging include*** petri dishes in a plastic sleeve within a plastic-lined box using paper towel spacers; stabs in a sealed Tupperware container with paper towels to cushion vials; sealed tubes in a rack placed into plastic sealable container with enough paper towels to absorb any spilled contents; tissue culture dishes placed into a plastic-lined dishpan or a sealable cardboard box with an absorbent. Freezers can be moved intact, provided all contents are in sealed, unbreakable containers and the freezer remains closed. Because shifting of contents may occur, enclose loose items in boxes, or fix in some other way to avoid breakage and spills when the freezer is reopened. Other equipment, such as fermenters, refrigerators, incubators, and biological safety cabinets must be empty and decontaminated prior to the move.

Labeling: Once packaged, all biological materials must be properly labeled. ***Labels must include*** the name, PI, new location, ID of agent, biosafety level, telephone number for assistance in the event of any breakage, and a FRAGILE notice if applicable. Also the **universal biohazard label** should be used whenever packaging a BSL2 or higher agent. Questions concerning the biosafety level of biological materials or requests for biohazard labels should be directed to the Biosafety Safety Officer at (617) 358-7840.

Procedures: Laboratory Furniture and Equipment

Furniture: The Move Coordinator must be informed if there is any furniture of particular concern (fragile, valuable, requires dismantling) not already mentioned. Different moving companies may have different requirements that should be ascertained in advance of the move.

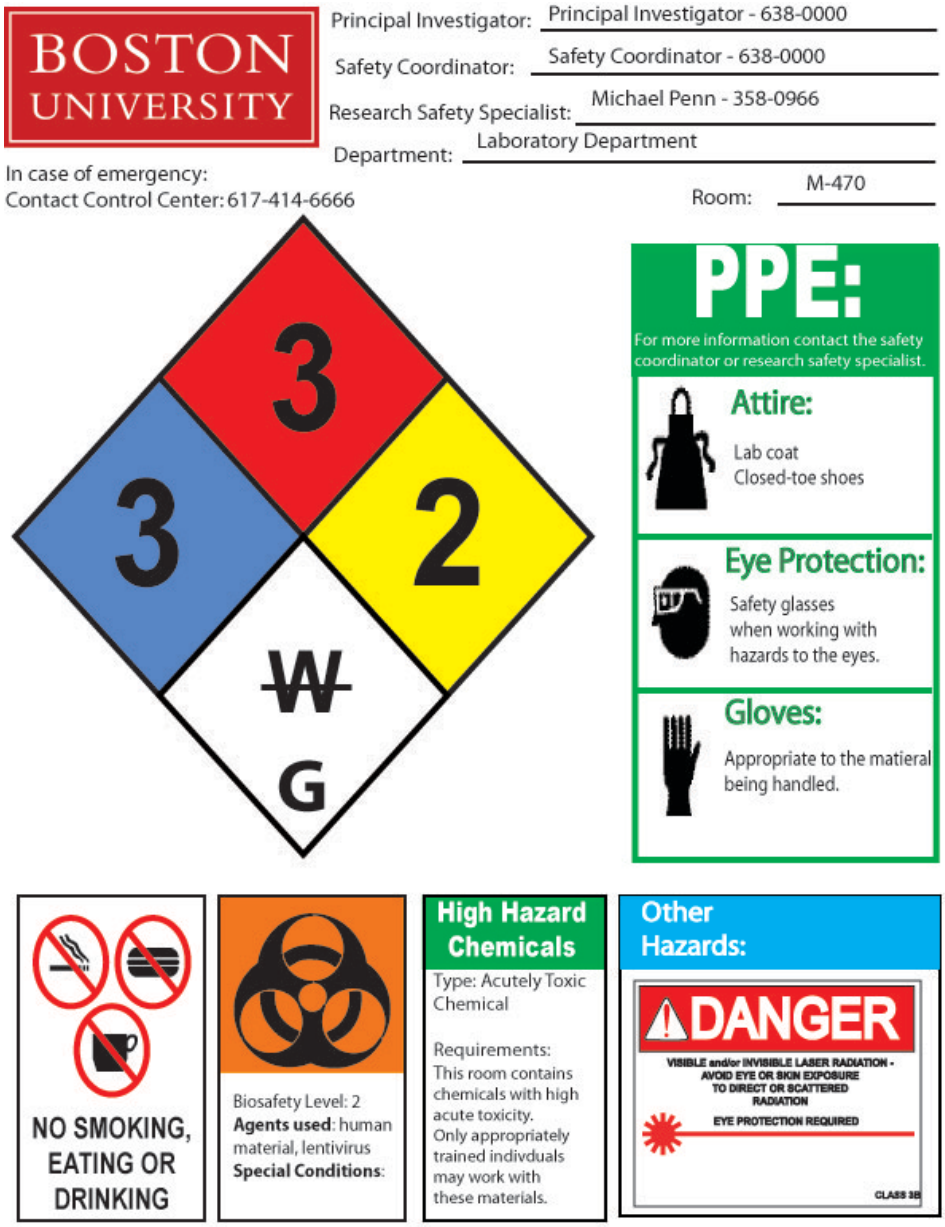
Special Requirements: The Move Coordinator must be informed in advance of any equipment under service contract, as well as equipment not under contract but requiring servicing and/or special handling.

Alarms: Laboratory personnel must disconnect alarms on freezers (if moving intact) and any other sensor alarms on or before the day of the move.

Keys and Combinations: Laboratory personnel must keep keys and combinations to locks readily accessible.

Appendix P
Laboratory Door Signage

Boston University Door Sign Sample: This is completed in accordance with the NFPA 704 Hazard Identification ratings system (the NFPA "hazard diamond") for health, flammability, and instability



Appendix Q
Policy on Disease Surveillance and Reporting for High-Risk Agents

Purpose and Applicability

This policy implements BPHC's *Guidelines for Implementation and Enforcement of Boston Public Health Commission's Disease Surveillance and Reporting Regulation.* The BPHC's guidelines require laboratory registration and a medical surveillance program for research laboratories working with high-risk agents. The guidelines are designed to ensure that BPHC receives timely access to information regarding incidence of disease syndromes, any outbreak or cluster of a disease, and potential exposures to reportable diseases deemed harmful to public health.

This policy sets forth the roles and responsibilities of researchers and compliance staff, as mandated by the BPHC guidelines.

This policy supplements, but does not replace or supersede, any other existing BU policies or procedures. For example, additional procedures relating to laboratory safety are set forth by EHS in the *EHS Manual*, and other subsidiary EHS documents.

Definitions

The **Associate Vice President, Research Compliance** (AVPRC) is the individual responsible for overall research compliance oversight at BU.

The **Biological Safety Officer** is the individual responsible for overall leadership of the biosafety program.

Expose or Exposure is any situation arising from, or related to, the work operation where an employee or community resident may ingest, inhale, absorb through the skin or eyes, or otherwise come into contact with any high-risk agent.

EHS Director is the Director of Research Safety in EHS of Boston University.

Occupational Health Officer (OHO) is the physician(s) who is the Occupational Health Officer of ROHP at Boston University. The Occupational Health Officer may also name a designee to perform occupational health assessments or evaluations, provided that the designee is also a licensed physician experienced in occupational medicine or a registered nurse experienced in occupational health nursing.

High–Risk Agent is a select agent, defined as:

- An agent that poses the greatest danger to public health and security or carries significant harm if used incorrectly.
- Agents in Risk Group (RG) 4 as specified in the National Institute of Health's *Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* and *Biosafety in Microbiological and Biomedical Laboratories 6 th edition,* published by the Centers for Disease Control and Prevention and the National Institutes of Health and the amendments and rulings made relative thereto from time to time.

- Other examples include Highly Pathogenic Avian Influenza, SARS Co–V or any other agent identified by the director of BPHC on a list to be posted on the BPHC's website or appearing on reporting forms. See details for applicable regulations.

Select Agent means microbial and toxic agents listed at 42 CFR 73.4, 42 CFR 73.5, and 9 CFR 121.2 and the rulings made by the CDC and U.S. Department of Agriculture relative thereto as amended from time to time.

Research Laboratory is a workplace or a work area of a workplace that is used primarily for research, development, non–routine testing, or experimentation activity in which any high-risk agent is used by or under the direct supervision of a technically qualified individual.

Work Area is a defined space, room or rooms, or other area where infectious agents or substances are produced, stored, or used, and where employees are present in the course of their employment. A work area may include an entire workplace.

Workplace is an establishment or business of an employer at one geographic location at which work is performed and contains one or more work areas.

Registration of Research Laboratories

The office of the AVPRC will be responsible for registering with BPHC all research laboratories possessing, producing, storing, or otherwise working with any high–risk agent.

Such registration shall be on a form, or electronic format, provided by the BPHC's Office of Environmental Health, and shall include the following:

- Name of the high-risk agent
- The location of each high-risk agent (but only if such disclosure is consistent with federal, state and institutional security restrictions and policies concerning select agents or high-risk agents).
- Principal Investigator responsible for the high–risk agent(s).
- Title and a brief description of the nature of the project.
- Grant identification number or other unique institutional identifier number for the project.
- Contact information for the IBC.
- Name and contact information for the Occupational Health Officer.

The information in the registration form shall be updated, on a form provided by the BPHC's Office of Environmental Health, twice a year, every July 31st and January 31st (or on the next business day if it falls on a holiday or weekend) following registration.

The AVPRC shall inform the OHO of all high-risk agents as they are identified and shall provide the OHO with a copy of each registration and update, simultaneously filing.

Responsibilities of Principal Investigators, Supervisors, Laboratory Directors

Ensuring Registration Prior to Project Commencement

The Principal Investigator (PI), Supervisor, or Laboratory Director of any research project that proposes to possess, produce, store, or otherwise work with any high–risk agent must first contact the AVPRC, or designee,

and ensure that a registration for the research laboratory is properly filed with the BPHC. The PI, Supervisors, and/ or Environmental Health and Safety/ Biosafety are responsible for assigning job activities and training based on their work environment in SciSure (formerly known as SciShield) so that ROHP is notified and can ensure proper medical surveillance and clearance. PI, Supervisors, and EHS/Biosafety are also responsible for ensuring staff are medically cleared and are trained prior to entering the lab space.

Develop Approved Plan Prior to Project Commencement

The Principal Investigator, Supervisor, or Laboratory Director of any research project who proposes to possess, produce, store, or otherwise work with any high–risk agent must:

- Develop a plan jointly with the OHO that will enable the Principal Investigator, Supervisor, or Laboratory Director to determine whether a significant exposure of personnel has occurred in the Research Laboratory and that will set forth a protocol for monitoring significantly exposed employees (for more information, see "Medical Surveillance of Employees Working With High-Risk Agents" under "Responsibilities of the Occupational Health Officer").
- Indicate in writing that the Principal Investigator, Supervisor or Laboratory Director, and OHO have each approved the plan
- The plan must also be approved by the AVPRC and the IBC before researchers on the project will be allowed access to a high-risk agent.

Mandated Reporting to OHO and EHS Director

The Principal Investigator, Supervisor, and/or Laboratory Director, and researcher shall promptly report to the OHO:

- Any diagnosis of any disease caused by a high-risk agent
- Any laboratory employee or other individual having access to a research laboratory that possesses, produces, stores, or otherwise works with any high-risk agent **who is absent from the workplace due to unexplained illness for a period of two or more consecutive workdays**.
- In addition, laboratory staff should call ROHP for any "unexplained symptoms or illness" such as: Fever 100.4°F (38°C) or higher that is not resolving or improving and / or accompanied by: other symptoms, such as body aches, chills, rash, nausea and/ or vomiting, or headache that are also not resolving or improving. Please see: Accident & Incident Reporting

The IBC, Principal Investigator, Supervisor, and/or Laboratory Director shall report to the AVPRC and the OHO any violation or breach of any laboratory procedures or any other incident that the IBC, Principal Investigator, Supervisor, or Laboratory Director **should reasonably believe resulted in exposure of laboratory personnel to a high-risk agent in the workplace or released any high-risk agent beyond the work area**.

Follow-up on Reporting Requirements of Laboratory Workers

Principal Investigators, Supervisors, and Laboratory Directors who learn of laboratory workers with reporting responsibilities as outlined in "Responsibilities of Laboratory Employees, Trainees, Students, and Others Who Have Access to High-Risk Agents" must confirm that such employees have reported to the ROHP **before returning to work** and that the employees have a written release to return to work provided by the ROHP.

Principal Investigators, Supervisors, and Laboratory Directors should refer any ill employee who has had access to a high-risk Agent to the ROHP for evaluation.

Responsibilities of Laboratory Employees, Trainees, Students, and Others Who Have Access to High-Risk Agents

Mandatory Reporting to OHO, AVPRC, and EHS

Laboratory workers or other individual having access to a research laboratory that possesses, produces, stores, or otherwise works with any high–risk agent and who are exposed to a high-risk agent from a spill or a breach in laboratory practices must immediately contact the OHO, AVPRC, and EHS Director of Research Safety to receive instructions as to appropriate immediate steps to be taken.

Mandatory Medical Evaluations

Any laboratory employee or other individual having access to a research laboratory that possesses, produces, stores, or otherwise works with any high–risk agent ***and*** who has been diagnosed with, is exhibiting symptoms of, or may have been exposed to, any high–risk agent ***or*** who has been absent from the work place due to illness for a period of two or more consecutive work days must report to the OHO prior to returning to work, for medical evaluation **before, and as a condition for**, returning to work.

Laboratory workers are **encouraged** to report **any** illness to the OHO if working with high–risk agents, even if the illness does not result in a two-day workplace absence.

Responsibilities of the Occupational Health Officer

Medical Surveillance of Employees Working With High-Risk Agents

The OHO is responsible for having in place a general plan to determine whether employees working with various high-risk agents have had significant exposure to a high-risk agent and a plan to monitor significantly exposed employees.

The OHO will work with the researcher, Principal Investigator, Supervisor, or Laboratory Director and EHS/ Biosafety on any research project that proposes to possess, produce, store, or otherwise work with any high–risk agent to:

- Perform a risk assessment and evaluate if there is an exposure concern.
- Develop the project-specific plan described in “Develop Approved Plan Prior to Project Commencement” as outlined in “Responsibilities of Principal Investigators, Supervisors, Laboratory Directors” which will enable the Principal Investigator, Supervisor, or Laboratory Director to determine whether a significant exposure of personnel has occurred in the research laboratory and which will set forth a protocol for monitoring significantly exposed employees; and
- Ensure that the project-specific plan is approved by EHS and the IBC before researchers have access to a high-risk agent.

The Occupational Health Officer will immediately notify the BPHC of all presumptive Lab Acquired Infections (LAI) followed by a confirmatory report once additional information is available. In the event of a medical incident, the Occupational Health Officer will act as a single point of contact and coordinate all Occupational Health activities and notify other agencies as appropriate.

The Entity is responsible for reporting and following the Laboratory Incidents Reporting Requirements to BPHC as identified in the BPHC Biological Laboratory Regulations Implementation & Enforcement Guidelines:

a) Any case or suspected case of illness or disease caused by a high-risk agent. Refer to the list, "Research Laboratories: Reportable Infectious Disease Agents and Toxins," which is available on the BPHC website.
b) Any accidental release, spill or accident which results in an exposure or potential exposure to a recombinant DNA material, or high-risk agent.
c) Any illness among persons caused or potentially caused by a recombinant DNA material, biological hazard, a high-risk agent or attenuated strain of a high-risk agent present in a laboratory.
d) Any employee who is absent from the workplace for two (2) or more consecutive workdays due to illness suspected of being related to occupational exposure to any high-risk agent used in the laboratory.
e) Any incident, problem, accident, or other event that caused or is suspected to have caused a serious threat to the public health; death; serious illness or bodily injury to any person; or extensive property damage.
f) Failure of any major mechanical systems occurring in the BSL-3/ABSL-3 or BSL4/ABSL-4 laboratory, even if redundant or backup systems are available and perform as designed during the incident. g) Any incident, problem or accident that must be reported to the Entity's IBC, NIH.

Report of Diagnosis, Symptoms, or Exposure

The OHO shall perform an occupational health assessment for any laboratory employee or other individual having access to the laboratory who:

- Has been diagnosed with;
- Is exhibiting symptoms of ***or***
- May have been exposed to any high-risk agent.

The findings of the assessment shall be immediately reported to the BPHC, via their Reporting Form, but in any event not later than one business day after completion of the assessment.

The OHO will conduct a follow-up assessment and provide information requested by BPHC regarding isolation and/or quarantine issues. If the determination is made that the illness is caused by a high-risk agent, BPHC will be consulted before an ill worker is allowed to return to work.

The OHO will send BPHC documentation that an exposed person has been cleared to return to work within three business days of clearance.

Report of Workplace Absence Due to Illness

The OHO shall perform an evaluation of any laboratory employee or other individual having access to a research laboratory that possesses, produces, stores, or otherwise works with any high–risk agent **and** who has been absent from the work place due to illness for a period of two or more consecutive work days.

The evaluation shall be completed prior to the employee's return to work.

If the OHO has a reasonable suspicion that the employee's illness may be related to exposure to any high-risk agent, the OHO shall immediately notify the BPHC.

If Occupational Health determines that the illness was caused by a high-risk agent and may be work related, the BPHC must be consulted at least three business days prior to the employee's expected return to work.

Report of Diagnosis of Disease

The OHO shall report to the BPHC any diagnosis of any disease caused by a high-risk agent. This report shall be made within one business day of the diagnosis.

Report of Violation of Laboratory Procedures Resulting in Release of High-Risk Agent

The OHO shall report to the BPHC any violation or breach of any laboratory procedures or any other incident which the OHO should reasonably believe released a high-risk agent beyond the work area. This incident shall be reported to BPHC within one business day of the breach or incident.

EHS Notification

The OHO shall provide the AVPRC and EHS Director of Research Safety with written notification of all reports made to the BPHC.

Manner of Reporting to ROHP

To report an injury or incident, please visit the Reporting Laboratory Injuries or Exposures page.

Appendix R
Boston University Institutional Biosafety Committee Noncompliance Policy

1. Purpose
The Boston University (BU) Institutional Biosafety Committee (IBC) has developed this policy for evaluating issues of noncompliance with IBC protocols, policies, and regulatory guidelines. Although uniform standards can serve as a guide, each individual situation is unique and will be judged on its own merits.

2. Reporting
All personnel involved in research overseen by the BU IBC have an obligation to report concerns of noncompliance to the IBC, Research Compliance, or via the processes outlined for including the

3. Applicability
The policy applies to all laboratories conducting IBC-approved activities and the personnel listed on BU IBC-approved protocols.

4. Authority
The BU IBC is charged with ensuring adherence with the National Institutes of Health Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines), OSHA Bloodborne Pathogen Standard (29 C.F.R. part 1910.1030), the Federal Select Agent Program (7 C.F.R. Part 331, 9 C.F.R. Part 21 and 42 C.F.R. part 73), The United States Government Policy for Oversight of Dual Use Research of Concern (DURC) and consistency with the guidance found in the Centers for Disease Control's (CDC) Biosafety in Microbiological and Biomedical Laboratories 6th Edition (BMBL). The IBC, therefore, monitors the conduct of research programs that fall under purview of BU for compliance with all the appropriate regulations, as well as institutional policies and procedures. Noncompliance may also be reported to the IBC as a result of annual/routine lab inspections performed by staff of the Environmental Health and Safety (EHS) program.

5. Definitions

Allegation of noncompliance: An unproven assertion of noncompliance.

Finding of noncompliance: A determination by the IBC that an assertion or allegation of noncompliance has been proven or substantiated. Findings of noncompliance by the IBC may include: violations of University policy, noncompliance with the other applicable federal, state and local laws or regulations governing the use of biohazardous materials and/or recombinant or synthetic nucleic acid molecules.

Serious noncompliance means noncompliance that adversely affects the health or welfare of research subjects (human and animal) and/or staff and:

- Harms or poses an increased risk of substantive harm to personnel; or
- Poses a risk of substantive harm to the general public or environment; or
- Compromises the integrity or validity of the research.

Examples of serious noncompliance may include, but are not limited to, the following:

- Failure of the Principal Investigator (PI) to adhere to the responsibilities outlined in Section IV-B-7 of the NIH Guidelines;
- Conducting procedures involving biohazardous materials and/or non-exempt recombinant/synthetic nucleic acid molecules without IBC approval;
- Continuing to conduct procedures involving biohazardous materials and/or non-exempt recombinant/synthetic nucleic acid molecules after an IBC protocol has expired;
- Working with an infectious agent, viral vector, or host system that is not documented in an approved IBC protocol;
- Deviating from approved IBC protocol in a way that could increase the exposure risk of personnel or the environment to biohazardous materials and/or non-exempt recombinant/synthetic nucleic acid molecules;
- Conducting of procedures by personnel not adequately trained but with a signed/approved personnel training form in the lab or on file with the IBC;
- Conducting procedures involving biohazardous materials and/or recombinant/synthetic nucleic acid molecules in a facility not approved for such use.

Continuing noncompliance means noncompliant activity that recurs after a report of the activity has been evaluated by the IBC (which may be either minor or serious) and after corrective action has been communicated in writing (e.g., email) to the PI. Note: determinations of continuing non-compliance that recur after IBC corrective action has been implemented may be reportable to external oversight authorities, as appropriate.

Minor noncompliance means any behavior, action, or omission in the conduct or oversight of research activities that deviates from the IBC-approved research plan, federal regulations, local, or institutional policies, but does not rise to the level of serious noncompliance.

Examples of minor noncompliance may include, but are not limited to, the following**:**

- Failure to respond to requests for revisions to protocols by the IBC in a timely manner;
- Addition of study personnel without notifying the IBC;
- Implementing minor wording or procedural changes in a study without first obtaining IBC approval.

6. Determinations and Corrective Actions

Initial Evaluation and Actions

A concern may be reported through various channels. Depending on the channel, and the nature of the concern, other offices and individuals will be informed, and in consultation with Research Compliance staff, the IBC Chair, EHS, and the Biosafety Officer, when applicable, immediate actions may be taken to address the concern. The PI will be notified in writing of these concerns and any required actions. During an evaluation it may be necessary to review research and other documentation, inspect facilities, and/or hold discussions with pertinent individuals including the PI, lab personnel and/or administrative personnel, as appropriate. In some cases, involvement by the Institutional Official (IO), BU Office of

General Counsel (OGC), and other University administration (e.g., Department Chair) may be required at the outset of an evaluation.

IBC Determination

Following an evaluation, the IBC Chair, in consultation with the above parties, may:

- bring the matter before the full IBC;
- appoint a subcommittee to review the reported concern;
- in the instance of minor non-compliance, may handle the matter or delegate handling of the matter to Research Compliance staff or the Biosafety Officer.

Determinations of Serious or Continuing Non-Compliance

If the IBC chair believes that the allegations may be serious or continuing non-compliance, the results of the evaluation, including all supporting documentation and the PI's corrective action plan, if developed, will be provided to the IBC for review at a convened meeting. Based on the information, the IBC will determine:

1. the nature of the concern as it relates to the *NIH Guidelines*, BMBL, University policies, and other applicable regulations;
2. the need for additional actions, such as further investigation or notification of other University officials as appropriate; and
3. further corrective measures to address the concern and prevent recurrence along with appropriate deadlines for response from the PI.

The IBC has the authority to address noncompliance based on *NIH Guidelines*, the BMBL, University policies, and other regulatory requirements. Findings of noncompliance may result in one or more of the following actions:

- Suspending the use of recombinant/synthetic nucleic acid molecules and/or biohazardous materials pending completion and acceptance by the IBC of a written plan by the PI for the correction and prevention of recurrence;
- Termination of approval for use of recombinant/synthetic nucleic acid molecules and/or biohazardous materials;
- Confiscation and destruction of the recombinant/synthetic nucleic acid molecules and/or biohazardous materials;
- Any other action necessary to protect personnel, the environment, the public and/or University, including restricting access to the laboratory in order to suspend activities.

The PI will be notified of the IBC's decision in writing. If the allegation involves other BU personnel for whom corrective actions may result, those individuals will be included in any appropriate communications.

Reporting to External Agencies

Findings of serious or continuing noncompliance will be reported to the appropriate agency, including, but not limited to, the study sponsor, NIH Office of Biotechnology Activities (NIH/OBA), the BPHC and the CDC. The IBC, in concert with Research Compliance, is responsible for reporting any significant problems (e.g., serious non-compliance) with, or violations of, the *NIH Guidelines* and any significant research-

related accidents or illnesses to the NIH/OBA within 30 days of the incident. These reports are not intended to be punitive toward the individuals involved, but rather are intended to assist the institution in developing new and better policies and practices to prevent future non-compliances from occurring.

References:

- Biosafety in Microbiological and Biomedical Laboratories (BMBL) 6th Edition
- NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules
- Occupational Safety and Health Standards for Bloodborne Pathogens
- Federal Select Agent Program
- United States Government Policy for Oversight of Dual Use Research of Concern (DURC)

Appendix S
BSL-3 and BSL-4 Biosafety Plan

For BSL3 and BSL4 Biosafety Plan, please contact NEIDL BSL3 or NEIDL BSL4 Biosafety Officer.