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UNIVERSITY OF
South Carolina

Biological Safety Manual

EH&S RESEARCH & LABORATORY SAFETY

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FOREWORD

This manual is a living document that is issued as a means of providing laboratory users of biological hazards with information on the biosafety policies and procedures of the University of South Carolina. These policies and procedures apply to laboratory activities involving recombinant or synthetic nucleic acids, human pathogens, human-derived samples, animal or plant pathogens, biological toxins, and other potentially infectious materials. The Biological Safety Manual is intended to provide practical guidance to assist labs with understanding biological safety principles and practices for the handling, use, storage, and disposal of biological materials used in laboratories. Another purpose for this manual is to promote compliance with biological safety regulations, standards, guidelines, or other requirements.

All university stakeholders are encouraged to take actions that will improve our laboratory safety culture. Each of us plays a critical role in creating a safe campus learning environment for all involved in education and research. Laboratory personnel using biological hazards should be familiar with the applicable biosafety requirements outlined in this manual. Principal Investigators should ensure their lab personnel understand the proper work practices, safety equipment, and other biosafety requirements for their research lab activities. The Biological Safety Program serves as a partner with the research community. The program provides lab inspections, trainings, consultations, and other biosafety services for labs to identify biosafety risks, mitigate the risk of exposures, and fulfill their compliance obligations.

The University has adopted an Enterprise Risk Management (ERM) oversight structure. The ERM process is led by senior leadership and extends the concepts of risk management to include identifying risks across the institution, assessing the impact of risks to the operations and mission, developing response or mitigation plans, and monitoring the identified risks. The ERM process is also intended to consistently monitor for emerging risks. The ERM committees have identified *laboratory safety* as a high-risk at the University, and this includes risks associated with research involving biological hazards. The Institutional Biosafety Committee (IBC) provides local review and oversight for research involving recombinant or synthetic nucleic acids, and other potentially infectious or hazardous biological materials. The IBC works in close collaboration with the Biological Safety Program in the EH&S Research Safety Bureau. The IBC reports to the ERM Research Safety and Compliance Oversight Committee.

EMERGENCY INFORMATION

If you are currently experiencing a health or safety emergency, immediately remove yourself from the hazard (*e.g.,* leave the lab if an inhalation hazard is present, use the eyewash or safety shower if exposed to a hazardous material). For emergency assistance, dial **911**. For USC police dispatch, dial **(803) 777-4215**.

Exposure Incident:

I. Stop work and immediately:
   a. *Needle stick or animal bite:* Wash or flush the exposed area with soap and water for 10 minutes.
   b. *Mucous membrane exposure:* Flush the exposed area with water. If exposure is to the eyes, flush eyes (holding open) using the eyewash station for 10 minutes.

II. Follow steps outlined in the **USC Workers' Compensation Guidance for Work Related Accidents or Injuries** if an employee of the University. For volunteers working in a laboratory, report injury immediately to supervisor and, if necessary, proceed to the nearest treatment facility.

III. Complete and submit the **USC Laboratory Incident Report Form** to the Biosafety Officer (BSO) **Sherika Smith** and **Mark Robbins** within 3 days of the incident.

Note: The exposed employee or volunteer and/or their supervisor should provide the healthcare professional evaluating the exposure incident with a description of the job duties relevant to the exposure incident, route(s) of exposure, circumstances of exposure, biological agent or hazard involved in the incident (*e.g.,* HIV+ blood, *Vibrio parahaemolyticus*, lentiviral vector), and relevant medical records.

Employees must receive medical treatment for work related non-emergency injuries at the medical facility designated by CompEndium Services. The supervisor and injured employee together should immediately call CompEndium Services (available 24/7) at 877-709-2667 to report the injury. If a supervisor is not available, another supervisor or HR Contact may assist the injured employee with this process.

Special requirements for reporting incidents to the NIH related to research subject to the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules*:

The *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* (*NIH Guidelines*) states that "...any significant problems, violations of the *NIH Guidelines*, or any significant research-related accidents and illnesses" must be reported to NIH Office of Science Policy (OSP) within 30 days. Certain types of accidents must be reported on a more expedited basis. Spills or accidents in BL2 laboratories resulting in an overt exposure must be immediately reported to the NIH OSP.

Any spill or accident involving recombinant or synthetic nucleic acid research of the nature described above or that otherwise leads to personal injury or illness or to a breach of containment must be reported to OSP. These kinds of events might include skin punctures with needles containing recombinant or synthetic nucleic acids, the escape or improper disposition of a transgenic animal, or spills of high-risk recombinant materials occurring outside of a biosafety cabinet. Failure to adhere to the containment and biosafety practices articulated in the *NIH Guidelines* must also be reported to OSP. Minor spills of low-

risk agents not involving a breach of containment that were properly cleaned and decontaminated generally do not need to be reported.

All incidents related to research subject to the *NIH Guidelines* must be reported by emailing a completed copy of the USC Incident Reporting Template to USC's Biological Safety Officer (BSO) at smiths69@mailbox.sc.edu, and Institutional Biosafety Committee (IBC) at mrobbins@mailbox.sc.edu. For incidents in BL2 labs resulting in an overt exposure that must be immediately reported to NIH OSP, the Principal Investigator must submit the Incident Report Form as soon as possible to the BSO and IBC with sufficient information to understand the nature and consequences of the incident, as well as its cause. Following initial reporting, a more detailed report can be provided to the NIH OSP later that includes the measures taken in response to mitigate the problem and to preclude its reoccurrence.

SECTION 1: OVERVIEW OF BIOLOGICAL SAFETY PROGRAM

The primary goal of the Biological Safety Program is to facilitate the implementation of administrative policies, work practices, safety equipment, facility design, and training programs to mitigate the risk of work involving biological hazards. Biological hazards are defined as organisms or substances derived from organisms that pose a risk to humans, animals, or the environment. Biological safety encompasses measures taken to identify biosafety risks, reduce the risk of exposure to biohazards (lab-associated infections) as well as promote compliance with biological safety regulations, standards, and guidelines.

Lab-associated Infections

Lab-associated infections (LAIs) are defined as infections acquired through lab related activities regardless of their clinical or subclinical manifestations. LAIs occur mainly due to human error or equipment failure. Although LAIs are often underreported, there have been well documented cases of LAIs, and it is estimated that a majority (~58%) happen in research laboratories[1]. The Biological Safety Program employs a risk assessment approach that takes into consideration an agent's characteristics (infectious dose, host range, viability in environment, etc.) as well as other factors to minimize the risk of lab-associated infections (refer to Risk Assessment for more information).

Purpose

The purpose of the Biological Safety Program is to provide biosafety services intended to protect research lab personnel by minimizing the risk of laboratory-associated exposures from potentially infectious materials, and to promote full compliance with applicable biosafety regulations, standards, and guidelines. The program also provides limited biosafety support services to teaching labs upon request.

Scope

All research and teaching laboratories utilizing biological hazards at the University of South Carolina. Biological hazards include:

- Infectious microorganisms (bacteria, viruses, fungi, etc.)
- Human-derived materials (*e.g.,* blood, unfixed tissues)
- Recombinant or synthetic nucleic acids
- Transgenic plants and animals
- Biological toxins

ROLES AND RESPONSIBILITIES

The Institutional Biosafety Committee (IBC) and the Biological Safety Program serve as partners in oversight of biological research at the University. The IBC provides local review and oversight for research involving biological hazards. The Biological Safety Program conducts lab inspections, training programs, consultations, and other biosafety and compliance services. Other institutional stakeholders also have responsibilities to provide an infrastructure that promotes research safety and compliance such as programs for research occupational health or services to manage lab coats.

[1] Laboratory-associated Infections: Summary and Analysis of 3921 Cases. Health Lab Sci 1976 Apr;13(2):105-14

Where unsafe practices involving the use of biohazards or actions in violation of established policies and guidelines are observed, the IBC is empowered with the authority to enforce the *NIH Guidelines* and to ensure that IBC approval conditions are fulfilled. In the event of a significant research-related incident, the IBC may suspend, limit, or terminate a Principal Investigator's authorization to conduct research pending a formal investigation. The IBC may take further actions deemed appropriate if a Principal Investigator has repeated compliance violations that are not corrected, any serious safety violations, or multiple less serious safety concerns are identified that create a significant risk to laboratory workers, other persons, or the environment.

A complete list of roles and responsibilities can be found in the IBC Charter.

INSTITUTIONAL BIOSAFETY COMMITTEE

The Institutional Biosafety Committee (IBC) is comprised of a minimum of six members. Membership selection is based on previous work experience and education to ensure they collectively have experience and expertise to effectively assess the safety and risks of research proposals. The committee meets at least quarterly. The meeting schedule and protocol deadlines are posted on the IBC website.

The Institutional Biosafety Committee is responsible for the administrative oversight of activities involving biological agents at USC. These responsibilities include, but are not limited to:

- Review research projects involving biological hazards. Review recombinant or synthetic nucleic acid molecules research conducted at or sponsored by the university to determine compliance with the *NIH Guidelines* and approve research projects that are found to conform with the *NIH Guidelines*. Review research involving human/animal/plant pathogens, human materials that may contain bloodborne pathogens, and HHS/USDA select agents and toxins.
- Periodically review and approve new or significantly amended biosafety policies.
- Adopt emergency plans covering accidental spills and personnel contamination resulting from recombinant or synthetic nucleic acid molecules research. Emergency plans will emphasize the prevention of occupational infections or environmental contamination.
- Open IBC meetings when possible and make meetings available upon request.

BIOLOGICAL SAFETY OFFICER

The Biological Safety Officer (BSO) is responsible for daily management of the biosafety program.

Specific responsibilities include:

- Conduct periodic assessments of laboratories to ensure compliance with regulatory requirements and university policies.
- Maintain biological safety policies, procedures, and guidance documents.
- Investigate laboratory accidents and report problems, violations, and injuries or illnesses associated with biological agents, to the Institutional Biosafety Committee.
- Provide biosafety training to laboratory personnel
- Manage the contract and records for certification of biosafety cabinets. Provide guidance to labs on the selection, installation, and use of biosafety cabinets.

PRINCIPAL INVESTIGATOR

The Principal Investigator is the individual who submits the application to employ biohazardous agents in his or her work. This individual is *fully responsible* for adherence to all guidelines and regulations. The Principal Investigator is also fully responsible for the safe use of such agents by himself/herself and those under his or her direction.

Responsibilities of the Principal Investigator:

- Never initiate or modify research involving biological materials which require IBC approval until that research, or the proposed modification has been approved by the IBC and met other requirements of the *NIH Guidelines*.
- Enforce institutional policies that control safety in and access to the laboratory.
- Ensure that laboratory staff is appropriately trained in the practices and techniques required to ensure safety and the procedures for dealing with accidents. Ensure all laboratory personal complete required EH&S biosafety training courses.
- Arrange and document the training of students and employees regarding biosafety procedures in the laboratory including routine and emergency procedures.
- Make available to all laboratory staff the protocols that describe the potential biohazards and the precautions to be taken
- Provide adequate personal protective equipment and instruction on its proper use.
- Ensure that biohazardous wastes are properly prepared for disposal.
- Adhere to Institutional Biosafety Committee approved emergency plans in the event of accidental spills and/or personal contamination.
- Report immediately to EH&S any suspected personnel exposures, theft of material, or other incidents regarding biosafety or biosecurity.
- Correct work errors and conditions that may impede safety or containment for biological hazards, correct all reported safety or compliance deficiencies identified during lab inspections, and submit corrective action plan by the requested due date)
- Comply with shipping requirements for biohazardous materials.

INDIVIDUAL

The individual worker is the person who deals with the biohazardous agent on a regular basis and must be familiar with the potential hazards and requisite safety procedures associated with the agent.

Responsibilities of the Individual:

- Have a working knowledge of relevant emergency and decontamination procedures.
- Complete all required biosafety training.
- Properly dispose of all biohazardous wastes.
- Report immediately to the Principal Investigator all suspected personnel exposures, theft of material, and any other biohazard related accidents. If the Principal Investigator is not available, the report should be made to EH&S/Biological Safety Officer

SECTION 2: GUIDELINES AND REGULATIONS

Occupational Health and Safety Administration (OSHA)

OSHA's Bloodborne Pathogens Standard (29 CFR 1910.1030) applies to all employees working with human-derived materials at the University.

National Institutes of Health (NIH)

NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (*NIH Guidelines*) must be followed by all working with recombinant or synthetic nucleic acids at USC.

Center for Disease Control and the National Institutes of Health (CDC-NIH)

Biosafety in Microbiological and Biomedical Laboratories (BMBL), 6th edition serves as a guidance document for standard microbiological practices, safety equipment, and lab design. This document is used to develop University policy and was used in the development of this manual.

Specialized Regulations and Guidelines

Select Agent Regulations

The Department of Health and Human Services (HHS) 42 CFR Part 73 and US Department of Agriculture (USDA) 7 CFR Part 331 & 9 CFR Part 121 regulations for the Federal Select Agent Program provides oversight for the possession, use, and transfer of select agents and toxins which have the potential to pose a severe threat to public, animal or plant health or to animal or plant products.

Shipping Regulations

The International Air Transport Association (IATA) and Department of Transportation (DOT) provide oversight for the shipment of biological materials internationally and within the United States respectively. The IATA Dangerous Goods Regulations (DGR) provide guidance on the air transport of dangerous goods, including infectious substances. Additionally, the IATA Infectious Substances Shipping Guidelines include information on the transport and shipping of biological materials via various modes of transport including sea, road, rail, and through mail and courier systems.

DOT's Hazardous Materials Regulations (HMR; 49 C.F.R., Parts 171-180) provide requirements for the transportation of infectious substances air, highway, rail, or water within the United States.

Infectious Waste Disposal

The disposal of infectious waste at the University is primarily regulated by the South Carolina Department of Environmental Services (SCDES). SCDES's Infectious Waste Program regulates the generators and transporters of biohazardous waste. At USC, a Biological and Infectious Waste Management Plan was developed to provide guidance on disposal of all USC biological waste.

SECTION 3: RISK ASSESSMENT AND MANAGEMENT

The Principal Investigator/Laboratory Manager is responsible for identifying biological hazards in the laboratory. A risk assessment should be performed to evaluate the possibility, severity, and consequences of an incident occurring. A five-step approach can be used to assess potential hazards in a laboratory. A risk assessment should be conducted:

- during conceptual design of a new project
- after the project has started
- when staff start working on a new project
- when there are changes in agents, procedures or equipment used
- when new materials (biological, chemical, radioactive, or a combination of these hazards) are being used
- after a near miss, incident, or accident
- when changes in regulations occur
- when there is a gain of knowledge in field

A biological risk assessment should consider:

- agent characteristics
- procedure hazards
- host factors
    - Training and experience of lab personnel working with the agent
    - Health status of lab personnel (*e.g.,* immunocompetence, pregnancy)
- procedural setting (*e.g.,* laboratory, field)

RISK ASSESSMENT PROCESS

1. Identify agent hazards and perform an initial assessment of risk. Agent hazard characteristics include:
    - capability of causing disease in a susceptible human host
    - virulence (measured by severity of disease)
    - availability of preventive measures and treatments
    - routes of transmission of lab infection
    - infective dose
    - stability in the environment
    - host range and endemic nature
    - reports of lab-associated infections (LAIs)
    - availability of preventative measures or treatments
2. Identify laboratory procedure hazards. Procedure hazards can include:
    - scale up activities (high concentration, large suspension volume)
    - aerosol generation
    - sharps usage
    - work with novel isolates or unknowns in environmental or clinical samples
3. Decide the appropriate biosafety level and select additional precautions indicated by the risk assessment

4. Evaluate the proficiencies of staff regarding safe practices and the integrity of safety equipment
5. Review the risk assessment with a biosafety professional, subject matter expert, and the IBC

Figure 1: Chain of Infection - The spread of an infection is described as a chain. Breaking any of the six points on the chain will prevent an infection from occurring.

SPECIAL CONSIDERATIONS

HEALTH STATUS & PREGNANCY

An individual's health status may affect their susceptibility to infection and ability to receive immunizations or other therapeutic interventions. Personnel are encouraged to consult with University Health Services for counseling and guidance. This guidance may be particularly valuable to determine susceptibility to infectious agents for individuals of reproductive age and/or those with conditions that may predispose them to an increased risk of infection (*e.g.,* medical immunosuppressive agents).

MINORS IN THE LABORATORY

In some instances, individuals under the age of 18 may encounter situations where exposure to biological hazards may be involved. Exposure to biohazards of those under 18 years of age requires

special attention because, from a regulatory standpoint, these individuals are considered minors. A minor's exposure to biohazards may occur in teaching laboratories or in an advanced placement summer science learning program. Principal Investigators with minors under their direction must be aware of the University policies and procedures established for the safety of individuals under 18 years of age.

It is the responsibility of the Principal Investigator to fully adhere to all regulations applicable to the safe use of any biohazards under their direction, with special attention given to the consideration of minors. No minor shall be permitted to work with open containers or dispersible forms of biohazardous agents. It is strongly recommended that minors not work directly with any potentially infectious materials. No minor shall work in the vicinity of any source of biohazards without the immediate and constant supervision of an adult who is familiar with all applicable safety practices pertaining to the minor's laboratory work.

SECTION 4: CLASSIFICATION OF BIOHAZARDS

The Principal Investigator must make an initial risk assessment based on the Risk Group of an agent. Agents are classified into four Risk Groups according to their relative pathogenicity for healthy adult humans by the following criteria:

- **Risk Group 1** agents are not associated with disease in healthy adult humans.
- **Risk Group 2** agents are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available.
- **Risk Group 3** agents are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available (high individual risk but low community risk).
- **Risk Group 4** agents are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available (high individual risk and high community risk).

The *NIH Guidelines* contain a list of agents classified by Risk Group. Also refer to CDC/NIH Biosafety in Microbiological and Biomedical Laboratories, Section VIII for additional information.

BIOSAFETY LEVELS

Four biosafety levels have been designated in the CDC/NIH *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) and in the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acids Molecules* Appendix G, which give detailed requirements for each level. The levels have been established based on the Risk Group of the infectious agent and the activities to be performed. It is important to note that the Principal Investigator is responsible for selecting and applying the recommended biosafety level for the work conducted. The investigator's unique knowledge and judgment of the agent to be used is critical in assessing the associated risks of exposure. A complete description of biosafety level criteria can be found on the EH&S Biosafety website.

Each biosafety level consists of a combination of prescribed lab procedures and safety equipment:

- **Biosafety Level 1 (BSL-1):** The practices and equipment utilized in a Biosafety Level 1 facility are appropriate for work with defined and characterized strains of viable microorganisms not known to cause disease in healthy adult humans. Examples of these types of microorganisms include *Bacillus subtilis*, and K-12 derivatives of *Escherichia coli.*
- **Biosafety Level 2 (BSL-2):** The equipment, practices, and facilities used in Biosafety Level 2 laboratories are established for a broad range of indigenous moderate risk agents. Examples include *Salmonellae*, herpes simplex virus, and *Staphylococcus aureus*. The primary hazards to workers associated with these agents are accidental autoinoculation, ingestion, and skin or mucous membrane exposure. Processes possessing the ability to produce aerosols must be conducted in primary containment devices.
- **Biosafety Level 3 (BSL-3):** Biosafety Level 3 facilities are established for the use of indigenous or exotic agents that possess the potential for infection by aerosol, and the results of such infection may have serious or lethal consequences. Typical examples of agents designated as requiring Biosafety Level 3 facilities include *Mycobacterium tuberculosis*, St. Louis encephalitis virus, and *Coxiella burnetii*.
- **Biosafety Level 4 (BSL-4):** Level 4 facilities are designed for work with dangerous and exotic agents, which pose a high risk to individuals to contract a life-threatening disease. In these facilities, all manipulations are of high risk, and the procedures and safety equipment are designed to prevent exposure.

SECTION 5: BIOLOGICAL SAFETY REQUIREMENTS

IBC REVIEW AND APPROVAL

An IBC Protocol must be submitted to the Institutional Biosafety Committee (IBC) for any work with recombinant or synthetic nucleic acid molecules, human/animal/plant pathogens, human-derived materials, and HHS/USDA regulated select agents and toxins.

The Committee may specify further precautions for certain types of operations and projects. To assist in observing safety precautions and to satisfy itself that adequate measures of safety are being practiced, the Biological Safety Officer serves as a liaison between the Committee and the individual researchers.

Protocols are approved for a period of three years, after which the protocol must be renewed. Principal Investigators are required to submit a Protocol Renewal prior to the expiration date for any protocol that will be continued beyond the expiration date of the initial application approval. Principal Investigators wishing to modify an approved protocol must submit an amendment. All amendments must be approved prior to initiating the proposed changes.

Authorization to use recombinant or synthetic nucleic acids or other biohazards can be suspended, limited, or cancelled by the IBC, pending a formal investigation, when sufficient cause exists. Examples of sufficient cause include: serious or repeated failure to comply with NIH, CDC, or other applicable regulations; repeated failure to comply with the USC Biosafety Manual, USC policies, and recognized good biosafety practices; or repeated failure to submit required documentation in a timely manner.

Refer to the IBC Charter for complete review and approval procedures.

BIOLOGICAL SAFETY TRAINING

Biological safety training requirements are dependent on the biological materials used in the laboratory. This manual does not provide detailed training regarding specific laboratory tasks and their associated biosafety procedures. The lab supervisor is responsible for providing training for lab personnel to understand their duties, potential hazards, risk assessments of hazards/experiments, precautions to minimize exposures, and proper procedures if an incident occurs. The supervisor is also responsible for ensuring lab personnel demonstrate proficiency in practices and techniques for experiments involving potentially infectious materials and for ensuring a lab-specific biosafety manual is accessible in the lab. The Biological Safety Program provides training on the general principles and practices of biosafety.

Training Course	Requirement	Frequency	Delivery
Laboratory Biosafety Level 2 (BSL-2)	All personnel working in a BSL-2 lab, including the Principal Investigator	Every 2 years	Classroom
Bloodborne Pathogens (BBP) for Researchers	Lab personnel working with human-derived materials such as human blood, body fluids, unfixed tissue or organ, human cell lines, or other potentially infectious materials	Annually	Online

NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acids	All Principal Investigators of laboratories conducting research involving recombinant or synthetic nucleic acid molecules	Every 3 years	Online
Shipping Biological Materials	Personnel shipping biological materials (*e.g.,* infectious substances, exempt patient specimens, genetically modified organisms, dry ice) to locations outside the University	Every 2 years	Online

The biosafety training schedule and guidance are posted on the Biological Safety Training webpage. A certificate is provided at the completion of each course and must be kept in lab records at least 3 years.

LABORATORY SAFETY INSPECTIONS- BIOLOGICAL SAFETY

Laboratory safety inspections are performed annually in all research labs. These inspections are used to evaluate the implementation of proper lab safety principles and practices, to identify hazards used, and document any safety deficiencies. Lab inspections also promote compliance with applicable research safety regulations, standards, and guidelines. During inspections, inspectors will provide guidance or recommendations to improve safety and compliance and serve as a technical resource to advise on lab safety issues. It is essential for a member of the laboratory to be present during each inspection to explain your research, verify your lab's hazards, show us around the lab, and provide you the opportunity to ask questions. Other safety walk-throughs pr unannounced inspections are conducted when needed.

To prepare for the annual lab safety inspection, it is recommended that labs perform self-inspections. Self-inspection checklists can be found on the EH&S website.

A link to the inspection report in the EH&S Research Safety Management System (RSMS) will be sent to the Principal Investigator and/or lab safety contact following the completion of each inspection. This report will include a description of any deficiencies identified and the appropriate corrective actions. The Principal Investigator is responsible for implementing all corrective actions documented in their lab inspection report within the two weeks following the inspection. The Biosafety Officer is available to answer questions or provide technical advice regarding research safety procedures, laboratory security, compliance requirements, or other biological safety issues.

GENERAL LAB REQUIREMENTS

In all biological research or teaching laboratories the following rules must be followed:

- Gloves must be worn when handling hazards to protect hands from exposure to these materials
- Eating or drinking, applying of cosmetics, and handling contact lenses is not allowed in lab areas where experiments are conducted, or any hazardous materials are used or stored.
- Food for human consumption is not allowed in any lab freezer or refrigerator that is used for research purposes.
- Closed toed shoes and long pants or equivalent for leg covering must be worn.
- Personal protective equipment used while in the lab must be removed before leaving to prevent contamination of non-lab areas.

- Mouth pipetting is prohibited. Mechanical pipetting devices must be used.
- All procedures should be performed to minimize the creation of splashes and/or aerosols.
- All disposable sharps must be placed in a conveniently located biohazard sharps container. Sharps containers must not be overfilled to prevent potential injury from protruding sharps.

The USC's lab research is primarily at BSL-1 and BSL-2, while animal work is at ABSL-1 and ABSL-2.

BIOSAFETY LEVEL 1 (BSL-1)

Biosafety Level 1 is suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment. The laboratory is not necessarily separated from the general traffic patterns in the building. Work is generally conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is neither required nor generally used. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by a scientist with general training in microbiology or a related science.

Details of the practices, containment, and facility requirements can be found in the BSL-1 Criteria.

BIOSAFETY LEVEL 2 (BSL-2)

Biosafety Level 2 builds upon Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It differs from BSL-1 in that: (i) laboratory personnel have specific training in handling pathogenic agents and are directed by competent scientists; (ii) access to the laboratory is limited when work is being conducted; (iii) extreme precautions are taken with contaminated sharp items*; and (iv) certain procedures in which infectious aerosols or splashes may be created are conducted in biosafety cabinets or other physical containment equipment.

***Note**: Plastic Pasteur pipets should replace glass Pasteur pipets when working with concentrated viruses and viral vectors.

Details of the practices, containment, and facility requirements can be found in the BSL-2 Criteria.

ENHANCED BIOSAFETY LEVEL 2 (BSL-2+)

Biosafety Level 2+ containment is used when working with microorganisms in a BSL-2 facility where BSL-3 work practices and procedures are used. While it is not a recognized containment level in the CDC's *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) or the *NIH Guidelines for Recombinant and Synthetic Nucleic Acid Molecules*, the BMBL states that after determining the appropriate biosafety level, additional precautions can be required as indicated by a risk assessment. Each decision to use BSL-3 work practices in a BSL-2 facility must be guided by the risk assessment process and in consultation with the Biosafety Officer and the Institutional Biosafety Committee (IBC).

BSL-3 work practices may include: all procedures with potential to generate aerosols or droplets done in a biosafety cabinet, enhanced PPE (disposable lab coats, double gloves, face shield, respiratory protection – determined by risk assessment), use of sealed rotor heads or safety cups when centrifuging specimens, special exposure protocols, and documented lab-specific training for work being conducted.

BIOSAFETY LEVEL 3 (BSL-3)

Biosafety Level 3 is applicable to clinical, diagnostic, teaching, research, or production facilities where work is performed with indigenous or exotic agents that may cause serious or potentially lethal disease through inhalation route exposure. BSL-3 builds on BSL-2 work practices stipulating that all procedures involving the manipulation of infectious materials must be conducted within a biosafety cabinet, other physical containment devices, or by personnel wearing appropriate personal protective equipment. Additionally, BSL-3 laboratories have special engineering and design features including but not limited to: access to the lab being restricted to entry between two self-closing doors, the sink having to be hands-free or automatic, and a ducted air ventilation system being required. The university has special notification and approval requirements for any lab considering plans to conduct BSL-3 research.

Details of the practices, containment, and facility requirements can be found in the BSL-3 Criteria.

ANIMAL BIOSAFETY LEVEL 1 (ABSL-1)

Animal Biosafety Level 1 (ABSL-1) is suitable for animal work involving well-characterized agents that are not known to cause disease in healthy adult humans, and that are of minimal potential hazard to laboratory personnel and the environment.

Details of the practices, containment, and facility requirements can be found in the ABSL-1 Criteria.

ANIMAL BIOSAFETY LEVEL 2 (ABSL-2)

Animal Biosafety Level 2 is suitable for work involving animals infected with agents associated with human disease and posing a moderate hazard to personnel and the environment. It addresses hazards from ingestion as well as from percutaneous and mucous membrane exposure. ABSL-2 builds upon the practices, procedures, containment equipment, and facility requirements of ABSL-1. ABSL-2 requires a biosafety cabinet or other physical containment equipment be used for procedures involving infectious materials that may generate aerosols or splashes.

Details of the practices, containment, and facility requirements can be found in the ABSL-2 Criteria.

ANIMAL BIOSAFETY LEVEL 3 (ABSL-3)

Animal Biosafety Level 3 involves practices suitable for work with laboratory animals infected with indigenous or exotic agents, agents that present a potential for aerosol transmission and agents causing serious or potentially lethal disease. ABSL-3 builds upon the standard practices, procedures, containment equipment, and facility requirements of ABSL-2. The University has special notification and approval requirements for any lab considering plans to conduct ABSL-3 research.

Details of the practices, containment, and facility requirements can be found in the ABSL-3 Criteria.

PLANT CONTAINMENT

The principal purpose of plant containment is to avoid the unintentional transmission of a recombinant or synthetic nucleic acid-containing plant genome, including nuclear or organelle hereditary material or release of recombinant or synthetic nucleic acid molecule-derived organisms associated with plants.

The containment principles are based on the recognition that the organisms that are used pose no health threat to humans or higher animals (unless deliberately modified for that purpose), and that the containment conditions minimize the possibility of an unanticipated deleterious effect on organisms and ecosystems outside of the experimental facility, *e.g.,* the inadvertent spread of a serious pathogen from a

greenhouse to a local agricultural crop or the unintentional introduction and establishment of an organism in a new ecosystem.

Four biosafety levels, referred to as Biosafety Level (BL) 1 - Plants (P), BL2-P, BL3-P, and BL4-P, are established in Appendix P-II, *Physical Containment Levels* of the *NIH Guidelines*. The selection of containment levels required for research involving recombinant or synthetic nucleic acid molecules in plants or associated with plants is specified in Appendix P-III, *Biological Containment Practices* of the *NIH Guidelines*. These biosafety levels are described in Appendix P-II, *Physical Containment Levels* of the *NIH Guidelines*. This appendix describes greenhouse practices and special greenhouse facilities for physical containment.

BL1-P through BL4-P are designed to provide differential levels of biosafety for plants in the absence or presence of other experimental organisms that contain recombinant or synthetic nucleic acid molecules. These biosafety levels, in conjunction with biological containment conditions described in Appendix P-III, *Biological Containment Practices*, provide flexible approaches to ensure the safe conduct of research.

For experiments in which plants are grown at the BL1 through BL4 laboratory settings, containment practices shall be followed as described in Appendix G, *Physical Containment* of the *NIH Guidelines*. These containment practices include the use of plant tissue culture rooms, growth chambers within laboratory facilities, or experiments performed on open benches. Additional biological containment practices should be added by the Greenhouse Director or Institutional Biosafety Committee as necessary (see Appendix P-III, *Biological Containment Practices*), if botanical reproductive structures are produced that have the potential of being released.

BIOLOGICAL CONTAINMENT PRACTICES FOR RESEARCH INVOLVING PLANTS

Appropriate selection of the following biological containment practices may be used to meet containment requirements for a given organism. This list is not exhaustive; there may be other ways of preventing effective dissemination that could possibly lead to the establishment of the organism or its genetic material in the environment resulting in deleterious consequences to managed or natural ecosystems.

Biological Containment Practices (Plants)

Effective dissemination of plants by pollen or seed can be prevented by one or more of the following procedures: (i) cover the reproductive structures to prevent pollen dissemination at flowering and seed dissemination at maturity; (ii) remove reproductive structures by employing male sterile strains, or harvest the plant material prior to the reproductive stage; (iii) ensure that experimental plants flower at a time of year when cross-fertile plants are not flowering within the normal pollen dispersal range of the experimental plant; or (iv) ensure that cross-fertile plants are not growing within the known pollen dispersal range of the experimental plant.

Biological Containment Practices (Microorganisms)

Effective dissemination of microorganisms beyond the confines of the greenhouse can be prevented by one or more of the following procedures: (i) confine all operations to injections of microorganisms or other biological procedures (including genetic manipulation) that limit replication or reproduction of viruses and microorganisms or sequences derived from microorganisms, and confine these injections to internal plant parts or adherent plant surfaces; (ii) ensure that organisms, which can serve as hosts or promote the transmission of the virus or microorganism, are not present within the farthest distance that the airborne virus or microorganism may be expected to be effectively disseminated; (iii) conduct

experiments at a time of year when plants that can serve as hosts are either not growing or are not susceptible to productive infection; (iv) use viruses and other microorganisms or their genomes that have known arthropod or animal vectors, in the absence of such vectors; (v) use microorganisms that have an obligate association with the plant; or (vi) use microorganisms that are genetically disabled to minimize survival outside of the research facility and whose natural mode of transmission requires injury of the target organism, or assures that inadvertent release is unlikely to initiate productive infection of organisms outside of the experimental facility.

Biological Containment Practices (Macroorganisms)

Effective dissemination of arthropods and other small animals can be prevented by using one or more of the following procedures: (i) use non-flying, flight-impaired, or sterile arthropods; (ii) use non-motile or sterile strains of small animals; (iii) conduct experiments at a time of year that precludes the survival of escaping organisms; (iv) use animals that have an obligate association with a plant that is not present within the dispersal range of the organism; or (v) prevent the escape of organisms present in run-off water by chemical treatment or evaporation of run-off water.

SECTION 6: HAZARD COMMUNICATION- SIGNS AND LABELS

The universally accepted biohazard warning symbol must be used to notify workers about the presence of infectious agents. It is the responsibility of the Principal Investigator to ensure that all necessary postings are installed and properly maintained. The warning symbol must be removed when the biohazardous agent is no longer in use or present. The biohazard symbol included on postings should be orange or red in color with a contrasting background. Generally, the location of the posting is predicated by how access is gained to the biohazard area.

In addition, postings should also be displayed in other areas such as biosafety cabinets, freezers, or other specially designated work and storage areas. Universal biohazard labels must be affixed to containers of regulated waste, refrigerators and freezers containing blood or other infectious materials. Labels must be affixed to other containers used to store, transport, or ship blood or other potentially infectious materials. Individual containers of blood or other potentially infectious materials that are placed in a labeled container during storage, transport, shipment, or disposal are exempted from these labeling requirements.

Laboratory equipment that will be permanently removed from the lab (sent to surplus, disposed of, etc.) must be thoroughly decontaminated using appropriate disinfectants, the biohazard label removed, and the equipment decontamination form attached to the outside.

All BSL-2 laboratories at USC must post the BSL-2 signage at the entrance door to the laboratory when infectious agents are present. This signage provides information about access and entry/exit requirements as well as any additional information regarding biohazard communication for the laboratory. The BSL-2 signage is a fillable document that must contain the contact information for the PI including their name, office number, and emergency contact phone number. Additionally, biohazardous materials must be listed and the biosafety level assigned to your lab indicated on the Laboratory Hazard Notice.

ABSL-2 signage must be posted on procedure room doors in which work involving laboratory animals infected with pathogenic agents is conducted. This signage provides information about PPE requirements, entry and exit procedures, as well as additional information regarding biological agents in use and other biohazard communication for the room. The ABSL-2 sign must contain biological agent information and the responsible investigator contact information for each agent used.

SECTION 7: PRIMARY CONTAINMENT- BIOSAFETY CABINET

Biosafety cabinets (BSCs) are used at the University of South Carolina as a primary means of containment for working safely with infectious microorganisms. BSCs act as primary barriers to prevent the escape of biological aerosols into the laboratory environment. This is an important function, because many laboratory techniques (*e.g.,* pipetting, vortexing, sonicating) are known to produce inadvertent aerosols that can be readily inhaled by the laboratory worker. Proper maintenance of BSCs used for work at all biosafety levels cannot be over emphasized. A BSC must be routinely inspected and tested by trained personnel, following strict protocols, to verify that it is working properly. BSCs are only one part of an overall biosafety plan which requires consistent use of good microbiological practices, appropriate containment equipment, and proper facility design.

BSCs are designed, in varying combinations, for:

- Personnel Protection: Protects personnel from harmful agents inside the BSC.
- Product Protection: Protects the work, product, experiment, or procedure performed in the BSC from contaminants in the lab environment or from cross contamination in the cabinet.
- Environmental Protection: Protects the environment from contaminants inside the BSC.

There are three broad types of BSCs, each with varying degrees of protection:

- Class I BSCs provide personnel and environmental protection, but not product protection. It is similar in air movement to a chemical fume hood but has a HEPA filter in the exhaust system to protect the environment. Class I BSCs are often used specifically for purposes that do not require product protection, such as to enclose equipment (*e.g.,* centrifuges, harvesting equipment or small fermenters), or for procedures with potential to generate aerosols (*e.g.,* cage dumping, culture aeration or tissue homogenization).

- Class II BSCs provide personnel, environmental and product protection. There are 4 types of Class II BSCs (A1, A2, B1, B2). Airflow is drawn into the front grille of the cabinet, providing personnel protection. In addition, the downward laminar flow of HEPA-filtered air provides product protection by minimizing the chance of cross-contamination across the work surface of the cabinet. In general, Class II BSCs are appropriate for work with agents assigned to biosafety levels 1-3. Class II BSCs provide the microbe-free work environment necessary for cell culture propagation and may be used for the formulation of nonvolatile antineoplastic or chemotherapeutic drugs. However, this type of BSC must not be used for work involving volatile chemicals or gases. The Class II BSC is the most common type used at USC.

- Class III BSCs are designed for work with highly infectious microbiological agents and provides maximum protection for the environment and the worker. It is a gas-tight enclosure with a non-opening view window. Both supply and exhaust air are HEPA filtered, and these cabinets are not exhausted through the general lab exhaust system.

SELECTION AND INSTALLATION

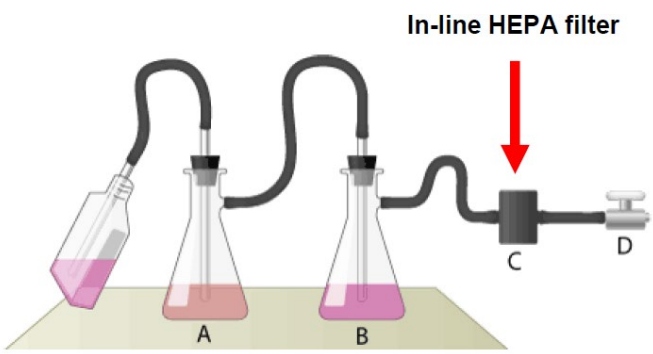
The Class II, Type A2 biosafety cabinet is the most common BSC used in USC research labs. This type of BSC provides personnel, product, and environmental protection from hazardous particulates such as

biological agents that require containment at Biosafety Level 1, 2 or 3. HEPA filtered exhaust air from a Class II, Type A2 BSC can be safely re-circulated back into the laboratory environment if the cabinet is tested and certified at least annually and operated according to manufacturer's recommendations. Proper selection and installation of BSCs must be in accordance with the most recent edition of Primary Containment of Biohazards: Selection, Installation and Use of Biological Safety Cabinets available in the BMBL in Appendix A.

USE OF ULTRAVIOLET (UV) LAMPS

According to the CDC/NIH – "Ultraviolet (UV) lamps are not recommended in BSCs nor are they necessary. If installed, UV lamps must be cleaned weekly to remove any dust and dirt that may block the germicidal effectiveness of the ultraviolet light. The lamps should be checked weekly with a UV meter to ensure that the appropriate intensity of UV light is being emitted. UV lamps must be turned off when the room is occupied to protect eyes and skin from UV exposure, which can burn the cornea and cause skin cancer. If the cabinet has a sliding sash, close the sash when operating the UV lamp." Some BSC manufacturers no longer include UV lamps in their Class II BSCs, unless specifically requested by the customer. The use of UV lamps can sometimes result in a false perception about their effectiveness. The ESCO Global has published a technical paper titled UV Lamps in Laminar Flow and Biological Safety Cabinets that is available for reference online at http://escolifesciences.us/resources/pdf/white-papers/uv-lamps-laminar-flow-and-biological-safety-cabinet.pdf. The University encourages labs not to use UV lamps in their biosafety cabinets. Any lab planning to use a UV light in their BSC must send a written justification to the Biosafety Officer and Institutional Biosafety Committee prior to use. If the lab has an IBC protocol, the IBC will determine if UV lamp use in the BSC will be approved for the project.

PROTECTION OF VACUUM LINES



CDC/*NIH Guidelines* for Biosafety Level 2 (BSL-2) state that vacuum lines should be protected with High Efficiency Particulate Air (HEPA) filters, or their equivalent. Filters must be replaced as needed. Liquid disinfectant traps may be required.

CDC/NIH Primary Containment for Biohazards: Selection, Installation and Use of Biological Safety Cabinets, 2nd Edition One method to protect a house vacuum system during aspiration of infectious fluids: The left suction flask (A) is used to collect the contaminated fluids into a suitable decontamination solution; the right flask serves as a fluid overflow collection vessel. A glass sparger in flask B minimizes splatter. An in-line HEPA filter (C) is used to protect the vacuum system (D) from aerosolized microorganisms.

USE OF BIOSAFETY CABINETS

A properly certified and operational biosafety cabinet is an effective engineering control which must be used in concert with appropriate work practices, procedures, and other administrative controls to reduce the risk of exposure to infectious microorganisms. The following work practices and procedures should be in place when working in a BSC.

Preparing for Work in a Class II BSC

- Place necessary materials in the BSC before beginning work to minimize the number and extent of air curtain disruptions compromising the fragile air barrier of the cabinet. Moving arms in and out slowly, perpendicular to the face opening of the cabinet will reduce the risk. Other personnel activities in the room (*e.g.,* rapid movements near the BSC face, walking traffic, open/closing doors) may also disrupt the cabinet air barrier.
- Lab coats should be worn buttoned over street clothing; latex, vinyl, nitrile, or other suitable gloves are worn to provide hand protection.
- Before beginning work, the investigator should adjust the stool/chair height so that his/her face is above the front opening.
- The front grille must not be blocked with toweling, research notes, discarded plastic wrappers, pipetting devices, or other materials. All operations should be performed on the work surface at least four inches in from the front grille.
- Extra supplies should be stored outside the cabinet. Only the materials and equipment required for the immediate work should be placed in the BSC. Clutter in the BSC can cause disruptions to the airflow in the cabinet.
- BSCs are designed for 24-hour per day operation. Some researchers prefer continuous operation to help control the laboratory's level of dust and other airborne particulates.
- If the cabinet has been shut down, the blowers should be operated at least four minutes before beginning work to allow the cabinet to "purge".
- The work surface, the interior walls, and the interior surface of the window should be wiped with 70% ethanol (EtOH) or other appropriate disinfectant.
- The surfaces of all materials and containers placed into the cabinet should be wiped with 70% ethanol to reduce the introduction of contaminants into the cabinet.

Placement of Materials in the BSC

- All materials should be placed as far back in the cabinet as practical, toward the rear edge of the work surface and away from the front grille of the cabinet.
- The biohazard collection bag should not be taped to the outside of the cabinet. Upright pipette collection containers should not be used in BSCs nor placed on the floor outside the cabinet. The frequent inward/outward movement needed to place objects in these containers is disruptive to the integrity of the cabinet air barrier and can compromise both personnel and product protection. Only horizontal pipette discard trays containing a chemical disinfectant should be used within the cabinet.
- Potentially contaminated materials should not be brought out of the cabinet until they have been surface decontaminated.

Operations in a Class II BSC

- The workflow should be from "clean to dirty". Materials and supplies should be placed in the BSC in such a way as to limit the movement of "dirty" items over "clean" ones.
- Opened tubes or bottles should not be held in a vertical position. Investigators working with Petri dishes and tissue culture plates should hold the lid above the open sterile surface to minimize direct impaction of downward air. Items should be recapped or covered as soon as possible.
- Open flames are not required in the near microbe-free environment of a biosafety cabinet.

- Aspirator bottles or suction flasks should be connected to an overflow collection flask containing disinfectant, and to an in-line HEPA or equivalent filter. This combination will provide protection to the central building vacuum system or vacuum pump.
- Inactivation of aspirated materials can be accomplished by placing sufficient chemical decontamination solution into the flask to inactivate the microorganisms as they are collected.
- Contaminated items should be placed into a biohazard bag, discard tray, or other suitable container prior to removal from the BSC.
- Two or more users should never use a BSC concurrently. BSCs are designed for single operator use. Multiple users increase the risk of cross contamination and loss of containment due to airflow disruptions.

Cabinet Surface Decontamination

- With the cabinet blower running, all containers and equipment should be surface decontaminated and removed from the cabinet when work is completed.
- Investigators should remove their gloves and gowns in a manner to prevent contamination of unprotected skin and aerosol generation and wash their hands as the final step in safe microbiological practices.

CERTIFICATION OF BIOSAFETY CABINETS

1. General Certification Requirements:
    a. All Class II BSCs must be tested and certified at least annually to ensure continued proper operation.
    b. All Class II BSCs will be tested and certified in accordance with specifications in NSF Standard 49 Annex N-5 and the manufacturer's specifications.
    c. The operational integrity must be validated by certification before a newly installed BSC is used and after a BSC has been repaired or relocated.
    d. After a BSC has been certified, a label will be prominently affixed to the front of the BSC, displaying the date of certification and name of the certifier.
    e. One copy of the certification report will be provided to the laboratory staff.
    f. NSF accredited field certifiers will be used to test and certify BSCs.
2. Performance Testing Requirements for Class II BSCs.
    Class II BSCs are the primary containment devices that protect the worker, product, and environment from exposure to microbiological agents. BSC operation, as specified by NSF/ANSI 49 - 2020, Annex N-5 needs to be verified at the time of installation and, as a minimum, annually thereafter. The purpose and acceptance level of the operational tests ensure the balance of inflow and exhaust air, the distribution of air onto the work surface, and the integrity of the cabinet and the filters. Other tests check electrical and physical features of the BSC. All Class II BSCs will be tested and certified as per NSF/ANSI 49 - 2020 specifications by using the following tests:
    - Downflow Velocity Profile Test
    - Inflow Velocity Test
    - Airflow Smoke Patterns Test
    - HEPA Filter Leak Test
    - Cabinet Integrity Test (A1 Cabinets only)
    - Electrical Leakage and Ground Circuit Resistance and Polarity Tests
    - Lighting Intensity Test

- Vibration Test
- Noise Level Test
- UV Lamp Test

3. Gas Decontamination (Reference NSF/ANSI 49 Annex I-2)
    a. BSCs that have been used for work involving infectious materials must be decontaminated before HEPA filters are changed or internal repair work is done.
    b. BSCs must be decontaminated prior to decommissioning and salvaging.
    c. Before a BSC is relocated, a risk assessment considering the agents manipulated within the BSC must be performed to determine the need and method for decontamination.
    d. The most common decontamination method uses formaldehyde gas, although more recently, hydrogen peroxide vapor and chlorine dioxide gas have been used successfully.
    e. In most instances where BSC decontamination is necessary, either depolymerized paraformaldehyde or chlorine dioxide gas should be used. Prior to decontamination with an alternative method (such as Vapor-Phase Hydrogen Peroxide), cycle parameters and validation of those parameters must be developed. When considering decontamination methods, many factors must be considered, such as safety, cycle time, effectiveness, equipment costs, and NSF requirements.
    f. Lab personnel should contact Environmental Health and Safety for guidance when determining the need for gas decontamination.
4. Certification of Biosafety Cabinets Used for Work Not Requiring Personnel Protection

As previously described, a Class II biosafety cabinet provides personnel, product, and environmental protection. Occasionally a laboratory will use a Class II BSC for work that does not require personnel protection (*e.g.,* using BSL-1 agents, PCR work). In these unique situations, a request is sometimes made for a BSC used for this purpose to be exempt from the annual certification. There are multiple justifications for requirements to annually certify all Class II BSCs using a qualified vendor, even when a BSC is used for experiments that do not require personnel protection:

    a. According to CDC/NIH: “A BSC must be routinely inspected and tested by trained personnel, following strict protocols, to verify that it is working properly. This process, referred to as certification of the BSC, should be performed at least annually.”
    b. A Class II BSC should be selected for protection of the worker, product, and environment. The certification performance testing verifies that a Class II BSC provides all three types of protection. Therefore, if a Class II BSC does not pass certification, it will not reliably provide all three types of protection and would not be beneficial for safety or research purposes. The Class I BSC provides personnel and environmental protection, but no product protection. Work that does not require personnel protection may not need to be conducted in a BSC. For instance, PCR can be safely performed in a PCR enclosure or cabinet that protects the sample from contamination. A PCR cabinet is not considered safety equipment, and therefore EH&S and the IBC does not require annual certification (even if recommended by the manufacturer).
    c. EH&S is responsible for promoting the proper maintenance of safety equipment (*e.g.,* BSCs, chemical fume hoods, eyewashes, safety showers) in labs to ensure this safety equipment functions according to its design specifications. EH&S is available to provide guidance on the selection, installation and use of appropriate safety equipment based on

a risk assessment. Once a BSC has been installed, it must be certified annually based on its Class and Type.

d. If a BSC does not pass annual certification, then a sign must be prominently displayed on the front of the sash that indicates – "This Biosafety Cabinet Did Not Pass Certification – DO NOT USE!" When the maintenance or repairs have been completed and the BSC passes certification, the sign can be removed from the sash and the BSC can be used. This procedure is consistent with numerous other academic research institutions that require annual certification of all BSCs to ensure proper operation, regardless of the intended use.

BSC MOVING, LIFESPAN, AND DECOMMISSIONING

Researchers occasionally need to move an installed biosafety cabinet (BSC) to another location in their lab or to another laboratory. A BSC must not be moved without first consulting with the Biosafety Office.

The current lifespan of a BSC is approximately 15 years. After 15 years, replacement parts may or may not be available due to electrical or mechanical changes at the factory or industrial part suppliers. BSCs have evolved through the years with many improvements in containment, ergonomics, serviceability, and energy efficiency. These issues should be considered when making decisions on BSC repair versus replacement (NSF/ANSI 49 Annex I-1 – Section I-1.9).

No BSC should be sent to a landfill or a recycling facility as a BSC, it should be disassembled per requirements contained in the NSF/ANSI 49-2020 Annex I-1 (Section I-1.10). Labs must consult with the Biosafety Office prior to decommissioning a BSC.

BSC PROGRAM ADMINISTRATION

1. Coordination of certification.
   Primary responsibility for the coordination of annual BSC certifications is held by the following:
   a. USC-Columbia – USC Environmental Health and Safety
   b. USC School of Medicine (SOM) – USC School of Medicine Facilities Management
2. Funding:
   a. USC Environmental Health and Safety provides funding for annual certification of BSCs in laboratories at the USC-Columbia campus. The Principal Investigator that owns the BSC must provide funds for expenses associated with relocations and/or any necessary BSC maintenance or repairs. If a BSC does not pass the annual certification, then the sash must be closed, and a sign must be prominently posted to ensure the BSC is not used until it has been properly certified.
   b. The USC School of Medicine (SOM) provides funding for annual certification of BSCs in research laboratories at the USC School of Medicine. The SOM may charge back individual Principal Investigators or departments for expenses associated with the annual certification, relocations, and/or necessary maintenance or repairs.
3. BSC Inventory:
   a. The Biological Safety Program maintains an inventory of BSCs for USC-Columbia and the USC School of Medicine. Each BSC's class and type, certification date, serial number, and location are included in these records. Certification dates are annually reported to EH&S so these records can be updated.
   b. All new BSC installations should be reported to EH&S to include in the inventory records.

c. EH&S will share this inventory with the designated certification vendor to ensure all BSCs are certified annually, and certification records are accurately maintained.

RESPONSIBILITIES

1. School of Medicine, Research Safety Coordinator
    a. Coordinates annual certification for BSCs at the USC School of Medicine (SOM).
    b. Ensures BSC certification vendor receives timely payment for services provided in SOM labs.
    c. Reports to EH&S any BSC maintenance/repairs, gas decontamination post-repair certifications, and BSC relocations or new installations.
2. Environmental Health and Safety
    a. Maintains an updated inventory record of all BSCs in laboratories at USC Columbia and USC School of Medicine Columbia.
    b. Coordinates with the certification vendor to ensure all BSCs are certified annually.
3. Principal Investigators/BSC Users
    a. Ensure the implementation of appropriate work practices when using a BSC, consistent with this program and the USC biological safety training.
    b. Promptly report any problems or questions with the selection, installation, or use of BSCs to the Biological Safety Officer (BSO). The BSO can provide guidance or assist with coordinating maintenance or repairs. The PI must ensure their BSCs pass certification. If a BSC does not pass certification, the PI must ensure it is not used until it is certified.
    c. Report the relocation or new installation of BSCs to the Biological Safety Officer before the relocation or installation. Any BSC that has been relocated or is a new installation must be certified prior to use. Proper reporting will also ensure the BSC certification inventory accurately reflects the location of each cabinet requiring annual certification.

SECTION 8: PERSONAL PROTECTIVE EQUIPMENT

Personal protective equipment (PPE) is worn to minimize exposure to hazardous materials and to prevent the spread of contamination. When working with biohazardous materials, PPE provides a barrier against skin, mucous membrane, or respiratory exposure to human pathogens. A properly maintained biosafety cabinet (BSC) or physical containment device should be used whenever possible, in addition to proper PPE. A BSC is especially important when conducting procedures with a potential for creating infectious aerosols or splashes or when high concentrations or large volumes of infectious agents are used. If it is not possible to perform a procedure within a biosafety cabinet or other physical containment device, a combination of PPE and administrative controls should be used, based on a risk assessment.

Examples of commonly used PPE in laboratories include lab coat, gloves, and safety glasses/googles. Other types of PPE should be worn when working with biological agents based on a risk assessment.

Personal protective equipment that should be provided at no cost and readily accessible to lab personnel includes, but is not limited to the following:

- Lab coat
- Gloves
- Face and Eye protection
- Other (*e.g.,* surgical mask with face shield, gowns), as required based on a risk assessment

It is the responsibility of the Principal Investigator (PI) or the department to provide employees with PPE. The Principal Investigator is also responsible for ensuring that the protective equipment is being used correctly. Individuals must use appropriate PPE as indicated by their PI and/or EH&S Research Safety. Accommodations will be made for individuals determined to be unable to use certain protective devices.

Prior to use, all PPE should be inspected for defects such as holes, rips, or any other defects that would compromise the integrity of the equipment. PPE with defects should be discarded and new PPE donned if found to be free of defects.

All personal protective equipment must be removed before leaving the work area. Remove PPE contaminated by any potentially infectious materials in such a way as to avoid contact with the outer surface. When personal protective equipment is removed, it must be placed in an appropriately designated area or container for storage, washing, decontamination, or disposal.

<u>Lab Coat</u>

A risk assessment should be done to determine the type of coat needed for lab work. For most work with biological materials, a fluid resistant/barrier, cotton, or cotton/polyester blend lab coat is sufficient. Disposable lab coats must be treated as biohazardous waste after use and discarded as such. Non-disposable lab coats should not be taken home for laundering. Lab coats should be at least knee length, have full length sleeves with fitted wrist cuffs, and able to button entirely (snap closures are preferred over buttons). Choose a lab coat that is the appropriate size (not too big or small, sleeve length reaches wrist, etc.).

<u>Gloves</u>

Gloves must be worn to protect hands from exposure to hazardous materials. Disposable gloves should never be washed or disinfected for reuse. Gloves must be replaced as soon as possible when visibly soiled, torn, punctured, or when their ability to function as a barrier is compromised. Disposable gloves must be treated as biohazardous waste after use. Alternatives to latex gloves must be available in the laboratory for use by individuals with a latex allergy.

Utility gloves may be disinfected for reuse if the integrity of the glove is not compromised. However, they must be discarded if they are cracked, peeling, discolored, torn, punctured, or exhibiting any sign of deterioration.

Face and Eye Protection

Masks and eye protection must be worn whenever splashes, sprays, droplets, or aerosols of potentially infectious materials may be generated and there is a potential for eye, nose, or other mucous membrane exposure. The type of face or eye protection should be chosen based on several factors, including but not limited to the nature of the hazard, the procedures to be performed, and other PPE used.

Respirators

Respirators include N95s that are worn to protect individuals from exposure to airborne potentially infectious agents that cannot otherwise be controlled using other engineering controls. Respirators must not be used in the laboratory without prior approval of EH&S. Principal Investigators and Supervisors are not authorized to select or recommend the use of respiratory protection, regardless of the type. Call EH&S (803-777-5269) or email Leigh Ann Broadway (lw32@mailbox.sc.edu) if you feel respiratory protection is required for your research, and to receive any required respirator fit-testing, medical clearance, and training. The justification and approval for use of respirators must also be documented in a biosafety protocol that has been approved by the IBC. Surgical face masks, used for mucous membrane protection, are not considered respirators and should not be used in situations where respiratory protection is required. All respirator users must be enrolled in the USC Respiratory Protection Program.

SECTION 9: LAB DESIGN- BIOLOGICAL SAFETY

Some laboratory activities conducted at the University of South Carolina may create a potential for occupational exposure to biohazardous materials. Biohazardous materials are defined as infectious agents, or materials produced by living organisms that may cause disease in other living organisms. While the laboratory procedures identified as good microbiological techniques are helpful in minimizing potential occupational exposure to biohazardous materials, containment of these agents using good facility design is also extremely important. The CDC and NIH document *Biological Safety in Microbiological and Biomedical Laboratories 6th Edition* (BMBL) provides guidance for the appropriate containment when conducting work involving biological hazards. The four biosafety levels are designated in ascending order by degree of protection provided to personnel and the environment. Special microbiological practices and facility design features serve to enhance worker safety and environmental protection, while also addressing the risk of handling agents requiring increasing levels of containment.

At a minimum, design for laboratories working with biological materials should include the following elements:

- Laboratories should have doors for access control.
- Laboratories must have a sink for hand washing.
- The laboratory should be designed so that it can be easily cleaned and decontaminated. Carpets and rugs in laboratories are not appropriate.
- Laboratory furniture must be capable of supporting anticipated loads and uses.
- Spaces between benches, cabinets, and equipment should be accessible for cleaning.
- Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
- Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.
- Laboratories windows that open to the exterior should be fitted with screens.

Additional design requirements may apply in Biosafety Level 2 (BSL-2) laboratories. More detailed Lab Design and Construction guidelines can be found on the New Laboratory page of the EH&S Chemical & Lab Safety section. The intent of these guidelines is to provide architects and engineers with a working knowledge of the facility design parameters required for the construction of facilities, and to provide for containment of biological hazards. All biological research laboratory design plans must be submitted to the Biological Safety Officer for review and guidance before a facility is renovated or new construction.

Biosafety Cabinets (BSCs):

HEPA filtered exhaust air from a Class II, Type A2 BSC can be safely re-circulated back into the laboratory environment if the cabinet is tested and certified at least annually and operated according to manufacturer's recommendations. BSCs can also be connected to the laboratory exhaust system by either a thimble (canopy) connection or a direct (hard) connection. This would allow for 100% of the filtered exhaust air to be discharged out of the laboratory. Provisions to assure proper safety cabinet performance and air system operation must be verified. The expense for installation and maintenance of a total-exhaust BSC is much higher, and therefore should only be selected and installed when justified based on a risk assessment of the research conducted in the lab (*e.g.,* cell culture work with infectious

agents and volatile toxic chemicals). The USC Biological Safety Officer (BSO) must be notified for further guidance prior to ordering and installing a total-exhaust cabinet (i.e., Class II, Type B2). The BSO should also be notified when a researcher is considering the installation of a any type of new BSC. Additional approval from the IBC may be required for installing any BSC besides a Class II, Type A2.

Utility services needed within a BSC must be planned carefully. Protection of vacuum systems must be addressed. Electrical outlets inside the cabinet must be protected by ground fault circuit interrupters (GFCI) and should be supplied by an independent circuit. When propane or natural gas is provided, a clearly marked emergency gas shut-off valve outside the cabinet must be installed for fire safety. The use of compressed air within a BSC must be carefully considered and controlled to prevent aerosol production and reduce the potential for vessel pressurization.

Certain considerations must be met to ensure the maximum effectiveness of biosafety cabinets as primary barriers used during manipulations of infectious microorganisms. Whenever possible, adequate clearance should be provided behind and on each side of the cabinet to allow easy access for maintenance and to ensure that the cabinet air re-circulated to the laboratory is not hindered. A 12- to 14-inch clearance above the cabinet may be required to provide for accurate air velocity measurement across the exhaust filter surface and for exhaust filter changes. BSCs should be located away from doors, windows that can be opened, and heavily traveled laboratory areas. The ideal location for the biosafety cabinet is remote from the entry (i.e., the rear of the laboratory away from traffic), since people walking parallel to the face of a BSC can disrupt the air curtain. Open windows, air supply registers, portable fans or laboratory equipment that creates air movement (*e.g.,* centrifuges, vacuum pumps) should not be located near the BSC. Similarly, chemical fume hoods must not be located close to a BSC.

All BSCs installed at USC should meet NSF Standard 49 requirements. The selection of cabinets should be based on an evaluation of the work to be performed and the specific safety requirements necessary to protect personnel, research, and the environment. Further guidance on the proper selection and installation of BSCs can be obtained from Appendix A—Primary Containment for Biohazards: Selection, Installation, and Use of Biological Safety Cabinets in the BMBL.

<u>Autoclaves:</u>

A method for decontaminating all laboratory wastes should be available in the facility. For maximum flexibility, autoclave space is recommended on each floor, or at a minimum in a convenient location in each building, where microbiological research is performed. Actual installation of autoclaves and their use are an operational decision. The architects and engineers should review the requirements of the building personnel and EH&S Research Safety when designing and specifying autoclave space.

Autoclave space should be finished with epoxy coatings and should not have a suspended, acoustical ceiling. This area should be thoroughly caulked and sealed to promote cleanliness and reduce pest harborage.

The space should have adequate exhaust capacity to remove heat, steam, and odors generated by using the autoclave(s). The autoclave space should operate at negative pressure to surrounding areas.

SECTION 10: BIOLOGICAL AND INFECTIOUS WASTE MANAGEMENT

DEFINITION OF INFECTIOUS WASTE

According to the SC Department of Environmental Services (SCDES), an infectious waste is any used material which is: generated in the health care community in the diagnosis, treatment, immunization, or care of human beings; generated in embalming, autopsy, or necropsy; generated in research pertaining to the production of biologicals which have been exposed to human pathogens; generated in research using human pathogens and which is listed in the categories below:

a. **Sharps**
   - Any discarded article that may cause puncture or cuts, including but not limited to: needles, syringes, Pasteur pipettes, lancets, broken glass or other broken materials, and scalpel blades.

b. **Microbiologicals**
   - Specimens, cultures, and stocks of human pathogenic agents, including but not limited to: waste which has been exposed to human pathogens in the production of biologicals; discarded live and attenuated vaccines; and discarded culture dishes/devices used to transfer, inoculate, and mix microbiological cultures.

c. **Blood and Blood Products**
   - All waste unabsorbed human blood, or blood products, or absorbed blood when the absorbent is supersaturated, including but not limited to: serum, plasma and other components of blood, and visibly bloody body fluids such as suctioned fluids, excretions, and secretions.

d. **Pathological Waste**
   - All tissues, organs, limbs, products of conception, and other body parts removed from the whole body, excluding tissues which have been preserved with formaldehyde or other approved preserving agents, and the body fluids which may be infectious due to bloodborne pathogens. These body fluids are: cerebrospinal fluids, synovial fluid, pleural fluid, peritoneal fluid, pericardial fluid, amniotic fluid, semen, and vaginal/cervical secretions.

e. **Contaminated Animal Waste**
   - Animal carcasses, body parts and bedding when the animal has been intentionally exposed to human pathogens in research or the production of biologicals.

f. **Other Waste**
   - Any other material designated by written generator policy as infectious, or any other material designated by a generator as infectious by placing the material into a container labeled infectious. Any solid waste which is mixed with infectious waste becomes designated as infectious and must be so managed.

g. **Infectious Waste Residues Resulting from Discharges**
   - Any residue or contaminated soil, water, or other debris resulting from the cleanup of a spill of any infectious waste.

WASTES NOT CLASSIFIED AS INFECTIOUS WASTE

The following are excluded from the definition of infectious waste:

a. **Hazardous waste** which is managed pursuant to the Hazardous Waste Management Regulations, R. 61-79

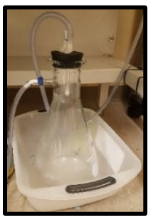
b. **Radioactive material** which is managed pursuant to the Department Regulation 61-63, Radioactive Material (Title A).
c. **Mixed waste** containing regulated quantities of both RCRA hazardous waste and source, special nuclear, or byproduct material subject to the Atomic Energy Act of 1954, as amended, are to be managed pursuant to all applicable regulations.
d. **Infectious wastes generated in a private residence** except when determined by the Commissioner to be an imminent or substantial hazard to public health or the environment.
e. **Etiologic agents or specimens being transported for purposes other than disposal to a laboratory** consistent with shipping and handling requirements of the U.S. Department of Transportation, U.S. Department of Health and Human Services, and all other applicable requirements.

INFECTIOUS WASTE MANAGEMENT

SHARPS WASTE

1. Sharps include any device or item capable of cutting or piercing the skin or a biohazard waste autoclave bag (*e.g.,* needles, syringes, Pasteur pipettes, pipette tips, scalpel and razor blades, blood vials, glass slides).
2. All sharps whether contaminated or not must be placed and maintained in rigid, leak-resistant and puncture-resistant biohazard sharps containers which are secured tightly to preclude loss of the contents.
3. Do not over-fill sharps containers. Once a container is ¾ full, the sharps container should be closed and placed in a large biohazard box or red wheeled cart (depending on campus location) for pickup by USC's infectious waste vendor.
4. Needles **must not** be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
5. Disposable sharps are recommended whenever possible. Non-disposable sharps **must be** placed in a hard walled container for transport to an area for decontamination (preferably autoclaving).

LIQUID INFECTIOUS WASTE



1. All liquid infectious waste (*e.g.,* human blood and body fluids, liquid culture media from infected cells, viral supernatant) must be placed, stored, and maintained before and during transport in a rigid or semi-rigid, leak-resistant container which is impervious to moisture.
2. Liquid infectious waste must be decontaminated prior to disposal. If using bleach, waste can be disposed, using large amounts of water, down the lab sink into the sanitary sewer system.

a. For chemical decontamination of liquid infectious waste, if using a bleach solution (final concentration of 10%) a minimum of a 30-minute contact time is required.

Note: Liquid biological waste containing a different disinfectant than bleach or any other chemical constituent must be disposed as chemical waste. Bleach is not compatible with many other chemicals such as ammonia and acids. **Consult with EH&S with questions regarding proper disposal of any mixed waste.**

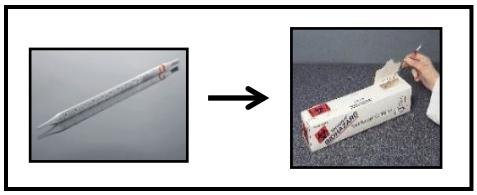
3. For stream sterilization (autoclaving), liquid waste should be collected in an appropriate container for autoclaving and the standard operating procedures for operating an autoclave for liquids should be followed. It is not recommended to autoclave large volumes of liquids.

4. Use either chemical decontamination OR stream sterilization but NOT both.

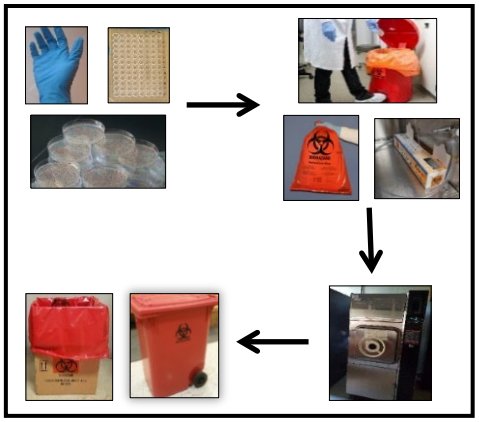
SOLID INFECTIOUS WASTE

1. All solid infectious waste (plastic consumables, gloves, etc.) must be placed, stored, and maintained before and during transport in a rigid or semi-rigid, leak-resistant container which is impervious to moisture.
2. Containers must have sufficient strength to prevent bursting and tearing and withstand handling, storage, and transfer without impairing the integrity of the container.
3. Reusable or disposable containers are acceptable. Reusable containers must be properly disinfected after each use. Containers should be kept closed when not actively adding waste to the biohazard bag.

4. Most types of solid infectious waste (i.e., waste not capable of puncturing an autoclave bag) should be collected in a **red or orange** color biohazard autoclave bag with sufficient strength to prevent tearing. Dispose of all materials with the Universal Biohazard symbol as infectious waste.



5. There may be alternative containers that are more specifically designed for disposal of some types of solid infectious waste. For instance, collection of serological pipets, swabs, and larger objects may be disposed in a collection container at the point of generation that is constructed of sturdy, plastic-lined paperboard for leak-resistance. These containers can be autoclaved and have a re-closable flap.



6. Infectious waste must be contained in containers that are appropriate for the type and quantity of waste generated.
7. Biohazard bags must be autoclaved. Place bags in an autoclave-safe tray prior to autoclaving. After autoclaving, discard the red or orange bag in a large biohazard box with a red bag liner or a red biohazard waste wheeled cart for pickup by USC's infectious waste vendor (the type of container depends on location and building).

8. Infectious waste generated in the lab must be autoclaved and disposed on a regular basis.
9. Do NOT autoclave infectious waste that is contaminated with hazardous chemicals. NEVER dispose of infectious waste in the regular waste stream.

ANIMAL INFECTIOUS WASTE

1. Waste generated from research animals (*e.g.,* animal carcasses, body parts, blood, bedding) that may be contaminated with zoonotic infectious agents or human pathogens during research must be treated as infectious waste.
2. Larger tissues, organs, and animal carcasses should be collected in red or orange biohazard bags but should NOT be autoclaved.
3. All biohazard bags containing animal carcasses or body parts should be placed in the freezers located in the DLAR (animal) facility.
4. Animal perfusion liquids must be collected in a hazardous waste carboy labeled as “Perfusion Liquid Waste Only”. This waste must be picked up as hazardous chemical waste due to the presence of paraformaldehyde and potentially other hazardous chemicals. Solid perfusion waste must be collected separate from the liquid waste.

BIOSAFETY LEVEL 1 WASTE MANAGEMENT

BSL-1 Microbiological Waste (Including RG1 Agents or BSL-1 Recombinant DNA)

According to the *NIH Guidelines* for Research Involving Recombinant or Synthetic Nucleic Acid Molecules, the following requirements apply to non-exempt BL1 waste:

- All contaminated liquid or solid wastes are decontaminated before disposal (Appendix G-II-A1c).

According to Biosafety in Microbiological and Biomedical Laboratories, 6th Edition, the following requirements apply to Biosafety Level 1 (BSL-1) microbiological waste:

- Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method, consistent with applicable institutional, local, and state requirements. (BMBL, 6th Edition; Section IV - Part A.15)

The following procedures should be used for disposal of Biosafety Level 1 microbiological waste:

- BSL-1 solid microbiological waste (including Risk Group 1 agents and BSL-1 recombinant DNA) must be collected and autoclaved in clear biohazard bags and then discarded in the red wheeled carts on the USC Columbia campus. (Note: Red or orange biohazard bags still should not be used in BSL-1 labs since these labs do not generate infectious waste.)
- In BSL-2 labs that generate some BSL-1 waste, it is acceptable for these labs to dispose of all solid microbiological waste as BSL-2 infectious waste in situations when it is difficult for the lab to properly segregate BSL-2 infectious waste from BSL-1 non-infectious waste in the lab.
- BSL-1 liquid microbiological waste (including Risk Group 1 agents and BSL-1 recombinant DNA) must be decontaminated using bleach (final concentration of 10% bleach for at least a 30-minute contact time) prior to sanitary sewer disposal.

INSECTS USED FOR RESEARCH

All insects (transgenic and wild-type insects) used in research must be rendered non-viable before disposal to protect the environment outside the research laboratory. All vials or bottles that may contain live insects must be sealed to prevent the escape of any insects. The preferred method for the termination of insects is to place the sealed primary bottles or vials containing the insects in a -20 °C freezer until the insects are no longer viable. Then bottles should be collected in a black bag and disposed of in an outside dumpster. Vials or bottles containing insects that are no longer needed for research must not be viable at the time they are collected in the bags and disposed in the dumpster. In situations that require removing the insects from the primary container for dissection or similar procedures, it is acceptable to place small insects (*e.g.,* fruit flies) no longer required for research in ethanol for termination as an alternative to placing them back in the primary container for freezer storage.

TRANSGENIC PLANT MATERIALS

According to the *NIH Guidelines*, the following requirements apply to the disposal of transgenic plants, including seeds, soil, and other transgenic plant materials used for research at BL1 containment:

- Experimental organisms shall be rendered biologically inactive by appropriate methods before disposal outside of the greenhouse facility (Appendix P-II-A-1c.1).
    - This principle is based on the recognition that the organisms that are used pose no health threat to humans or higher animals. The intent is to minimize the possibility of an unanticipated deleterious effect on organisms and ecosystems outside of the experimental facility, *e.g.,* the inadvertent spread of a pathogen or the unintentional introduction and establishment of an organism in a new ecosystem.
- Disposal of BSL1 transgenic plant materials should be done following the same procedures described above for other BSL-1 recombinant DNA waste, with the following considerations:
    - Autoclave transgenic plant materials at 121°C for a minimum of 60 minutes.
    - Soil from transgenic plant research should be double-bagged and transported in a durable leak-proof container to the autoclave. Autoclavable bags should be filled no more than 1/2 full and 250 ml of water should be added to facilitate steam penetration for effective decontamination. If the soil is already moist, extra water does not need to be added to the bag before autoclaving. The final weight after autoclaving should be no more than 15 lbs. An autoclave-safe tray with a solid bottom and walls must be used to contain the contents and prevent soil from spilling.
    - Reusable horticulture supplies used with transgenic plants should be decontaminated.
    - Any soil or other plant materials that are **NOT** used for transgenic plant research should be discarded in durable trash bags and disposed by lab personnel in an outside dumpster. Custodial Services will not dispose of soil or other plant materials from research labs.

INFECTIOUS WASTE TREATMENT

- A written quality assurance plan must be implemented when conducting any onsite treatment
- Stream sterilization (autoclaving) – The following waste will be autoclaved prior to disposal:

    - Solid infectious waste that is not capable of puncturing the biohazard autoclave bags (*e.g.,* culture plates and stocks)
    - Infectious waste that must be stored longer than 72 hours at room temperature
- Compactors or grinders will not be used to process infectious waste.
- Infectious waste will be picked up by USC's infectious waste vendor for incineration.

SPILL RESPONSE

Laboratories conducting experiments involving biological hazards such as microorganisms, human-derived materials, and recombinant or synthetic nucleic acid molecules must have plans for handling accidental spills.

The following items should be conveniently accessible in any lab using potentially infectious materials, and all lab personnel must know the location of these materials:

1. Gloves (latex or nitrile)
2. Lab coat or disposable gown
3. Safety glasses or goggles
4. Disinfectant solution*
5. Tongs, forceps, dustpan, broom
    - A mechanical device must be used to remove sharps without using gloved hands
6. Absorbent materials (*e.g.,* paper towels)
7. Signage to post at lab entrance for controlling access ("Biohazard Spill – Do Not Enter")
8. Biohazard bags for collecting all contaminated materials generated during the cleanup, and a puncture-resistant biohazard sharps container if spill involves contaminated sharps
9. A copy of all applicable biological spill procedures

* A freshly prepared 10% bleach solution is effective for the decontamination of most biological spills. Some laboratories have the potential for spills involving agents or materials that may be resistant to a 10% bleach disinfectant. In these cases, it is important for the lab to use an effective disinfectant. A list of selected EPA-registered disinfectants is available online at https://www.epa.gov/pesticide-registration/selected-epa-registered-disinfectants.

PROCEDURES FOR SPILLS OF BIOLOGICAL MATERIALS REQUIRING BSL-1 OR BSL-2 CONTAINMENT:

1. Alert people in the immediate area that a spill occurred (avoid spreading spilled material)
2. Put on appropriate personal protective equipment (*e.g.,* gloves, lab coat, safety glasses)
3. Cover the spill with absorbent material (*e.g.,* paper towels)
4. Carefully soak the paper towels and spilled material with disinfectant (avoid splashing)
5. Allow a 20-minute disinfectant contact time

6. Wipe down any contaminated equipment with disinfectant
7. Remove broken glass or other sharps with a brush and dustpan, tongs, or forceps
    - Place contaminated sharps in a puncture-resistant biohazard sharps container
8. Use absorbent material to wipe up the spill
9. Clean the area once more with absorbent material and disinfectant solution
10. Place contaminated disposable materials in a leak-proof biohazard bag for autoclaving, and properly decontaminate any non-disposable materials prior to reuse
11. Remove gloves and thoroughly wash hands
12. Notify lab personnel when the clean-up has been completed

NOTE: If an exposure occurs it must receive immediate attention before cleaning up the spill, and personnel must follow the approved procedures for post-exposure evaluation and follow-up.

PROCEDURES FOR SPILLS INSIDE A BIOSAFETY CABINET (BSC):

1. Follow above procedures for spills of materials requiring BSL-1 or BSL-2 containment
2. If material has spilled into the catch basin below the work surface: a) close the drain valve; b) flood the drain with disinfectant (volume equal to quantity in basin; c) wait 20 minutes (disinfectant contact time); d) absorb remaining liquid with paper towels
3. After the completion of clean-up, the cabinet should run for 10 minutes before use

PROCEDURES FOR SPILLS INSIDE A CENTRIFUGE:

1. Turn off the centrifuge & wait 20 minutes before opening lid to allow aerosols to settle
2. Follow above procedures for spills of materials requiring BSL-1 or BSL-2 containment
3. Remove buckets and rotors and move them to closest biosafety cabinet before opening
4. Disinfect interior of centrifuge & disinfect buckets/rotor per manufacturer’s instructions

NOTE: When centrifuging high concentrations or large volumes of infectious agents, these materials may be centrifuged in the open laboratory using sealed rotor heads or centrifuge safety cups.

SECTION 11: RECOMBINANT DNA AND SYNTHETIC NUCLEIC ACID MOLECULES

The *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* (*NIH Guidelines*) detail safety practices and containment procedures for basic and clinical research involving recombinant or synthetic nucleic acid molecules (r/s NA), including the creation and use of organisms and viruses containing recombinant or synthetic nucleic acid molecules. Additionally, the *NIH Guidelines* address the possibility that genetic modification could increase an agent's pathogenicity or affect its susceptibility to antibiotics or other effective treatments. USC's Institutional Biosafety Committee (IBC) provides local review and oversight for research involving r/s NA, and other potentially infectious or hazardous biological materials.

All projects, both NIH and non-NIH funded, involving recombinant or synthetic nucleic acid molecules conducted at or sponsored by USC must comply with the *NIH Guidelines*. Non-compliance could result in the suspension, limitation, or termination of financial assistance for the non-compliant NIH-funded research project and of NIH funds for other recombinant or synthetic nucleic acid molecule research at USC. Non-compliance could also result in a requirement for prior NIH approval of any or all projects involving recombinant or synthetic nucleic acid molecules at USC.

Authorization to use recombinant or synthetic nucleic acids or other biohazards can be suspended or terminated by the Institutional Biosafety Committee when sufficient cause exists. Examples of sufficient cause may include:

- Serious or repeated failure to comply with NIH, CDC, or other applicable biosafety regulations
- Repeated failure to comply with the USC Biosafety Manual, USC policies, and recognized good biosafety practices
- Repeated failure to submit required documentation or responses to the IBC in a timely manner.

RESPONSIBILITIES (SECTION IV)

NIH Office of Science Policy

The OSP serves as source of information on recombinant or synthetic nucleic acid molecule activities and provide advice to research institutions, Biological Safety Officers, Principal Investigators, Federal agencies, and state and local governments.

Primary Responsibilities

- Reviewing and approving experiments in Section III-B-1 of the *NIH Guidelines*
- Publishing in the Federal Register, as needed
- Reviewing and approving the membership of an institution's Institutional Biosafety Committee

USC

All institutions conducting or sponsoring recombinant or synthetic nucleic acid research which is covered by the *NIH Guidelines* is responsible for ensuring that the research is conducted in full conformity with the provisions of the NIH Guidelines. USC must:

- Establish and implement policies that provide for the safe conduct of recombinant or synthetic nucleic acid molecule research and that ensure compliance with the *NIH Guidelines.*
- Establish an Institutional Biosafety Committee.
- Appoint subject matter experts to the IBC if plant and animal research subject to the *NIH Guidelines* is conducted.
- Ensure that when the institution participates in or sponsors recombinant or synthetic nucleic acid molecule research involving human participants: (i) the IBC has adequate expertise and training (using ad hoc consultants as deemed necessary) and (ii) no human gene transfer experiment will be initiated until IBC approval has been obtained and all other applicable institutional and regulatory authorization(s) and approvals have been obtained.
- Assist and ensure compliance with the *NIH Guidelines* by Principal Investigators conducting research.
- Ensure appropriate training for the IBC Chair and members, Biological Safety Officer, and other containment experts (when applicable), Principal Investigators, and laboratory staff regarding laboratory safety and implementation of the *NIH Guidelines*.
- Determine the necessity for health surveillance of personnel involved in connection with individual recombinant or synthetic nucleic acid molecule projects; and if appropriate, conduct a health surveillance program for such projects. The institution shall establish and maintain a health surveillance program for personnel engaged in large-scale research or production activities involving viable organisms containing recombinant or synthetic nucleic acid molecules which require BL3 containment at the laboratory scale. The institution shall establish and maintain a health surveillance program for personnel engaged in animal research involving viable r/s NA molecule-containing microorganisms that require BL3 or greater containment in the laboratory.
- Report any significant problems, violations of the NIH Guidelines, or any significant research-related accidents and illnesses to the NIH OSP.

Institutional Biosafety Committee

The IBC is responsible for ensuring that all research with r/s NA molecules conducted at or sponsored by USC is conducted in compliance with the *NIH Guidelines*. The IBC's responsibilities at USC include:

- Reviewing recombinant or synthetic nucleic acid molecule research conducted at or sponsored by USC for compliance with the *NIH Guidelines* as specified in Section III of the *NIH Guidelines*
- Review of r/s NA must include:
    - independent assessment of the containment levels required by the *NIH Guidelines* for the proposed research
    - assessment of the facilities, procedures, practices, and training and expertise of personnel involved in r/s NA research
    - for r/s NA research involving human research participants assessment focused on biosafety issues (*e.g.,* administration, shedding)
- Notifying the Principal Investigator of the results of the IBC's review and approval
- Lowering containment levels for certain experiments as specified in Section III-D-2-a
- Setting containment levels as specified in Sections III-D-4-b
- Periodically reviewing r/s NA research conducted at USC to ensure compliance with the *NIH Guidelines*

- Adopting emergency plans covering accidental spills and personnel contamination resulting from recombinant or synthetic nucleic acid molecule research
- Reporting any significant problems with or violations of the *NIH Guidelines* and any significant research-related accidents or illnesses to the appropriate institutional official and NIH OSP

Biological Safety Officer

The primary duties of the Biological Safety Officer (BSO) include:

- Periodic inspections to ensure that laboratory standards are rigorously followed
- Reporting to the IBC and the institution any significant problems, violations of the *NIH Guidelines*, and any significant research-related accidents or illnesses of which the BSO becomes aware
- Developing emergency plans for handling accidental spills and personnel contamination and investigating laboratory accidents involving recombinant or synthetic nucleic acid molecule research
- Providing advice on laboratory security
- Providing technical advice to Principal Investigators and the IBC on research safety procedures
- Ensure required biosafety training is provided to laboratory personnel. This training includes, but is not limited to, Biosafety Level 2 and Bloodborne Pathogens training

Principal Investigator

The Principal Investigator is responsible for full compliance with the *NIH Guidelines* in the conduct of r/s NA research and is responsible for ensuring that the reporting requirements are fulfilled and will be held accountable for any reporting lapses.

<u>General Responsibilities:</u>

- No initiation or modification of r/s NA research which requires IBC approval prior to initiation
- Report any significant problems, violations of the *NIH Guidelines*, or any significant research-related accidents and illnesses to the Biological Safety Officer
- Report any new information bearing on the *NIH Guidelines* to the IBC
- Be adequately trained in good microbiological techniques
- Adhere to IBC approved emergency plans for handling accidental spills and personnel contamination
- Comply with shipping requirements for r/s NA molecules

<u>Responsibilities of the Principal Investigator Prior to Initiating Research:</u>

- Make available to all laboratory staff the protocols that describe the potential biohazards and the precautions to be taken
- Instruct and train laboratory staff in: (i) the practices and techniques required to ensure safety, and (ii) the procedures for dealing with accidents
- Inform the laboratory staff of the reasons and provisions for any precautionary medical practices advised or requested

<u>Responsibilities of the Principal Investigator During the Conduct of the Research:</u>

- Supervise the safety performance of the laboratory staff to ensure that the required safety practices and techniques are employed
- Investigate and report any significant problems pertaining to the operation and implementation of containment practices and procedures in writing to the Biological Safety Officer
- Correct work errors and conditions that may result in the release of recombinant or synthetic nucleic acid molecule materials
- Ensure the integrity of the physical containment (*e.g.,* biosafety cabinets) and the biological containment (*e.g.,* purity and genotypic and phenotypic characteristics)
- In the event of a significant research-related incident, the IBC may suspend, limit, or terminate a Principal Investigator's authorization to use biological materials pending a formal investigation. The IBC may take further actions deemed appropriate if a Principal Investigator has repeated compliance violations that are not corrected, any serious safety violations, or multiple less serious safety concerns are identified that create a significant risk to laboratory workers, other persons, or the environment.
- The IBC may review and approve new or significantly amended biosafety policies
- Open IBC meetings to the public when possible and consistent with protection of privacy and proprietary interests
- Make meeting minutes available to the public upon request

INCIDENT REPORTING

All incidents involving recombinant or synthetic nucleic acids must be reported to the Biological Safety Officer (BSO) as soon as possible. Complete the Template for Reporting Incidents Subject to the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* and email it to the BSO at smiths69@mailbox.sc.edu. The Biological Safety Officer will investigate the incident and, along with the IBC, will determine whether the incident meets the NIH reporting requirements.

Certain types of incidents must be reported to the NIH on a more expedited basis. Examples include spills or accidents occurring in Biosafety Level (BL) 2 laboratories resulting in an overt exposure or spills and accidents that result in environmental release or exposures of animals or laboratory workers to organisms containing recombinant or synthetic nucleic acid molecules in Animal Biosafety Level 2 (BL2-N) laboratories must be immediately reported to the NIH.

SECTION 12: HUMAN DERIVED MATERIALS- BLOODBORNE PATHOGENS

In 1991, the Occupational Safety and Health Administration (OSHA) established the regulatory standard **29 CFR 1910.1030**, which is referred to as the Bloodborne Pathogens Standard. This regulation applies to all personnel with an occupational exposure to blood or other potentially infectious materials. Human blood or certain other body fluids may contain pathogenic microorganisms that cause disease in humans. These pathogens include, but are not limited to, hepatitis B (HBV) or human immunodeficiency virus (HIV). An individual is considered to have an occupational exposure if they have a reasonably anticipated skin, eye, mucous membrane, or parenteral contact with blood or other potentially infectious materials (OPIM) that may result from the performance of their job duties.

This OSHA regulation requires the University to establish a written Exposure Control Plan designed to eliminate or minimize employee exposure. The regulations also require the implementation of additional provisions designed to prevent exposures. These provisions include, but are not limited to, providing personal protective equipment, procedures for disposal of regulated waste, procedures for management of contaminated sharps, availability of containment equipment, training, hepatitis B vaccination, and post-exposure medical evaluation and follow-up.

The term “Universal Precautions” is an approach to infection control that refers to the concept of treating all human blood and certain human body fluids as if they are known to be infectious for HIV, HBV, and other bloodborne pathogens. It is especially important that employees strictly follow "Universal Precautions" any time they may reasonably anticipate contact with these potentially infectious materials.

SCOPE AND APPLICATION

The scope of personnel impacted by bloodborne pathogens is described in OSHA’s Bloodborne Pathogens Standard (29 CFR 1910.1030). According to OSHA, this section applies to all occupational exposure to blood or other potentially infectious materials as a result of the performance of an individual’s job duties. These other potentially infectious materials would include the following human body fluids: semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, pericardial fluid, peritoneal fluid, amniotic fluid, and any body fluid that is visibly contaminated with blood. These infectious materials would also include any unfixed human tissue or organ, and HIV-containing cell or tissue cultures, and HIV or HBV containing culture medium or other solutions.

At the University of South Carolina, anyone conducting research involving human blood, body fluids, or unfixed tissues are personnel with a potential occupational exposure and would be required to comply with the OSHA Bloodborne Pathogens Standard.

POLICY ON ESTABLISHED HUMAN AND NON-HUMAN PRIMATE CELL LINES

BACKGROUND

In 1994, OSHA issued an interpretation of the applicability of the BBP Standard towards human cell lines. According to the interpretation, human cell lines* are considered to be potentially infectious and within the scope of the BBP Standard unless the specific cell line has been characterized to be free of hepatitis viruses, HIV, Epstein-Barr virus, papilloma viruses and other recognized bloodborne

pathogens.[2] In addition, the BMBL recommends that human and other primate cells should be handled using Biosafety Level 2 (BSL2) containment and practices.[3]

The American Type Culture Collection (ATCC) note in their frequently asked questions section that all adventitious agents may not be detected through viral testing. For this reason, they strongly recommend that all human and other primate cell lines be handled at the same biosafety level as a cell line known to carry HIV or hepatitis virus. [4]

USC POLICY

In consideration of the applicable regulatory interpretation and consensus guidelines, the USC Institutional Biosafety Committee has adopted the following requirements for handling established human and non-human primate cell lines:

All human and non-human primate cells, including well established cell lines, must be handled in compliance with the OSHA Bloodborne Pathogens Standard and handled at Biosafety Level 2 (BSL-2). Animal Biosafety Level 2 (ABSL-2) containment and practices may be required when these materials are used in animal experiments.

* **OSHA Human Cell Line Definition:**

A human cell line is defined as *in vitro,* or animal passaged (*e.g.,* nude mouse) cultures or human cells that fulfill traditional requirements of a cell line designation. That is, the cells are immortalized cells, transformed by spontaneous mutation or natural or laboratory infection with an immortalizing agent such as Epstein-Barr virus (EBV). EBV is a bloodborne pathogen. It should be noted that human cervical carcinoma cells or other transformed human cell lines like HeLa cells are sometimes adulterated with laboratory pathogens accidentally introduced by cultivation with other cell cultures, or physically contaminated by other cell cultures handled in the same lab. In order to handle human HeLa cells, without having to comply with the requirements of the bloodborne pathogens standard (BPS), human HeLa cells should be documented to be pure HeLa cells and shown to be free of bloodborne pathogens by testing.

PROGRAM DESCRIPTION

Exposure Control Plan

Each laboratory or department must develop a written site-specific Exposure Control Plan. This plan indicates all job classifications in which all employees in those job classifications have occupational exposure; a list of job classifications in which some employees have occupational exposure; and a list of all tasks and procedures in which occupational exposure occurs. A copy of this plan must be accessible

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[2] OSHA Letter of Interpretation https://www.osha.gov/laws-regs/standardinterpretations/1994-06-21

[3] Biosafety in Microbiological and Biomedical Laboratories (BMBL), 6th Ed.- BSL-2 Criteria, ABSL-2 Criteria, Working with Human, NHP and Other Mammalian Cells and Tissues (pgs. 37-43, 78-87, 466-468) https://www.cdc.gov/labs/BMBL.html

[4] ATCC Testing cell lines for viruses before use. Should I test ATCC cell lines for viruses before using them in the lab? https://www.atcc.org/support/technical-support/faqs/testing-cell-lines-for-viruses-before-use

to all employees included in the plan. The Exposure Control Plan should also include procedures for the evaluation of circumstances surrounding exposure incidents. The plan must be reviewed and updated at least annually and whenever necessary to reflect new or modified tasks and procedures which affect occupational exposure.

An Exposure Control Plan template for research laboratories is available on the EH&S Biosafety website.

TRAINING

All employees with occupational exposure must participate in a bloodborne pathogens training program. This training is provided at no cost to the employee and during working hours. Training must be provided initially at the time workers are assigned tasks involving exposure and at least annually thereafter. Additional training should be provided when changes in tasks or procedures affect the employee's occupational exposure. Training for laboratory personnel is provided by the EH&S Biological Safety Program, and training requirements for academic programs are determined by each academic program:

- Where to access a copy of the regulations and an explanation of its contents
- Explanation of the epidemiology and symptoms of bloodborne diseases
- Explanation of bloodborne diseases and their modes of transmission
- Explanation of the departmental Exposure Control Plan and where to obtain a copy
- Explanation of methods to recognize activities that may involve exposure
- Explanation of engineering controls, work practices, and personal protective equipment.
- Information on the hepatitis B vaccine
- Information on the actions to take and persons to contact during an emergency or exposure incident
- Information on the post-exposure evaluation and follow-up provided by the University
- Explanation of signs and labels where blood or other infectious materials are present
- An opportunity for questions and answers

It is the responsibility of the supervisor to ensure that workers attend initial and annual training. The department must maintain attendance records. Training records should include the dates of training sessions, summary of training, names and qualifications of person conducting training, and names and job titles of personnel attending training. Training records should be maintained for 3 years from the date training occurred. The training courses and schedule are available on the Biosafety training website.

VACCINATIONS

Hepatitis B vaccination is made available at no cost and within 10 working days of initial assignment to all employees who have occupational exposure. Although this vaccination is recommended, an individual may choose not to accept it by signing a declination statement. If the employee initially declines vaccination, but later decides to accept it, the University will make the vaccination available at that time. The Center for Health and Well-Being will administer the vaccination series.

ROLES AND RESPONSIBILITIES

DEPARTMENT

- Identify individuals with occupational exposure and notify EH&S.
- Develop a written departmental (or laboratory-specific) Exposure Control Plan.
- Review and update the plan on an annual basis or as exposure conditions change.
- Ensure employees attend the required initial and annual Bloodborne Pathogens Training.
- Provide necessary personal protective equipment (PPE) and engineering controls.
- Ensure appropriate laboratory facilities are available to personnel performing work involving blood or other potentially infectious materials.
- Maintain a current written Exposure Control Plan and training records.

SUPERVISORS

- Ensure all personnel with occupational exposure receive training, are provided and use the appropriate personal protective equipment (PPE) and adhere to "Universal Precautions".
- Laboratory supervisors must assure that employees demonstrate proficiency in standard microbiological practices, and in practices and operations of the facility before being allowed to work with HIV or HBV.
- Ensure that laboratory employees have prior experience in the handling of human pathogens or tissue cultures before working with HIV or HBV.
- Provide lab-specific training to employees that have no prior experience in handling human pathogens; initial work activities should not include infectious agents; a progression of work activities should be assigned as techniques are learned and proficiency is developed.

INDIVIDUALS

- Attend Bloodborne Pathogens Training and understand the exposure risks associated with the performance of job duties.
- Notify EH&S of hepatitis B vaccination status (*e.g.,* request vaccination, submit signed vaccination declination form).
- Utilize appropriate work practices, engineering controls, and personal protective equipment.
- Adhere to "Universal Precautions" when in contact with blood or other potentially infectious materials.
- Immediately report any exposure incidents to supervisory personnel and seek the necessary post-exposure medical evaluation and follow-up.

EH&S

- Develop policies/plans to promote compliance with the OSHA Bloodborne Pathogens Standard.
- Provide guidance regarding appropriate engineering and work practice controls.
- Provide guidance on the selection and use of personal protective equipment.
- Provide guidance on proper management of contaminated sharps.
- Coordinate removal of all regulated infectious waste from campus by an approved vendor.
- Provide appropriate warning signs and labels for containers and appliances being used to store regulated waste, blood, or other potentially infectious materials.

- Investigate exposure incidents and provide follow-up recommendations for necessary work practice modifications to improve safety minimize the risk of similar future incidents.
- Perform inspections of departments with laboratory personnel having occupational exposure.
- Review and update the Bloodborne Pathogens Exposure Control Plan Template annually.

OCCUPATIONAL HEALTH

- Maintain a copy of the Bloodborne Pathogens Standard (29 CFR 1910.1030).
- Provide medical surveillance, including administration of hepatitis B vaccine.
- Provide medical evaluation and follow-up for exposure incidents.
- Document the route(s) of exposure, and circumstances under which any exposure incident occurs.
- Provide identification and documentation of the source individual following exposures.
- Coordinate collection and testing of blood for HBV and HIV serological status (when applicable).
- Collect blood from exposed employees as soon as feasible and test the blood after consent is obtained.
- Administer post-exposure prophylaxis, when medically indicated, as recommended by the U.S. Public Health Service.
- Provide any employee involved in an exposure incident with a copy of the evaluating healthcare professional's written opinion within 15 days of the completion of evaluation.
- Inform exposed individuals about any medical conditions resulting from exposure to blood or other potentially infectious materials which require further evaluation or treatment.
- Provide counseling and evaluation of reported occupational illnesses.
- Maintain an accurate record for each employee with occupational exposure.
- Conduct initial and annual Bloodborne Pathogens Training.

SECTION 13: BIOLOGICAL TOXINS

Toxins are poisonous substances that are produced by organisms but are not living themselves. Because toxins are non-living, they do not replicate, are not infectious, and are difficult to transmit from person to person. Common biological toxins include tetrodotoxin, staphylococcal enterotoxins, diphtheria toxin, and cholera toxin. The use of regulated biological toxins must be approved by the IBC.

Prior to working with toxins, biosafety considerations should include:

- **Physical state:** dry (freeze-dried) toxins are easier to aerosolize than other preparations which can present a higher risk to the user. Work using dry toxins should be avoided when possible.
- **Toxin amount:** for some toxins, exposure to small quantities can result in death or serious health consequences. Toxins that have a low LD_{50} (median lethal dose) should be handled with extra precautions.
- **Routes of exposure:** the routes of exposure depend on the physical state and diluents used. The main routes of exposure in the laboratory include ingestion, eye or mucous membrane exposure, inhalation, and needle-sticks. The use of needles and other sharps in the manipulation of biological toxins should be minimized due to the increased exposure risk.
- **Containment:** Procedures involving toxins should be confined to biosafety cabinets, fume hoods, glove boxes, or other approved containment devices to minimize aerosol production and exposure. A laminar flow hood ("clean bench") must never be used for work with biological toxins.
- **Decontamination and inactivation procedures:** some toxins are highly resistant to standard decontamination and inactivation procedures and an alternative must be used. Many toxins are susceptible to inactivation with dilute sodium hydroxide (NaOH) at concentrations of 0.1-0.25N, and/or sodium hypochlorite (NaOCl) bleach solutions at concentrations of 0.1-0.5% (w/v).

Risk assessments should be conducted, and standard operating procedures (SOPs) developed for the use of biological toxins. Lab-specific training must be conducted prior to work with toxins and this training documented for all users. Training should include the following elements:

- The theory and practice of the toxins to be used
- An emphasis on the nature of the practical hazards associated with laboratory operations
- How to handle transfers of liquids containing the toxin
- Where to place waste solutions and contaminated materials or equipment
- How to decontaminate work areas after routine operations and after spills

SECTION 14: BSL-3 RESEARCH AND SELECT AGENTS AND TOXINS

Research involving select agents or agents requiring BSL-3 or ABSL-3 containment is more challenging and expensive to establish and maintain than any other type of research at the university. Select agent and BSL-3/ABSL-3 research that is conducted at or sponsored by the university must be reviewed and approved by the IBC (and IACUC for ABSL-3) prior to initiating the research.

NOTIFICATION AND APPROVAL PROCESS:

1. A research questionnaire [pdf] must be submitted to the Institutional Biosafety Committee (IBC) Chair and the university's Research Safety Bureau Chief at least three months before any faculty member submits a grant for this type of research, or any department hires a new faculty member planning to conduct select agent, BSL-3, or ABSL-3 research.
2. The PI must prepare and submit a BSL-3 research proposal to the Biosafety Officer. This proposal must be submitted prior to pursuing funding for BSL-3 research. The PI must describe any anticipated future research activities that may impact the BSL-3 facility design plan and require future approval prior to use. A description of how the BSL-3 laboratory will fulfill regulatory requirements will also be required.
    a. Proposals to conduct research involving select agents will require additional descriptions of how the BSL-3 facility will fulfill regulatory requirements for the Select Agent Program. The college(s) with research faculty using the BSL-3 facility must appoint a Responsible Official (RO) with the authority and responsibility to ensure compliance with the select agent regulations.
3. The university will appoint a committee of subject matter experts (e.g., biosafety, facilities, security) to evaluate the feasibility and resources required for the planning, design, renovation/construction, management, operations, and safety/compliance oversight for a new high containment facility. This evaluation will be based on the research proposal packet submitted.
    a. The PI will be consulted as necessary during the review process. Other stakeholders may be involved to determine if the proposed BSL-3 facility supports the university's strategic research mission and to assess other risk management issues.
4. Following completion of the assessment process, the committee will prepare a feasibility report on the proposed BSL-3 facility plans.
5. Finally, if plans for the design and construction of a new BSL-3/ABSL-3 facility are endorsed by the BSL-3 Committee, IBC, and senior administrators, then the department or college will need to hire an experienced full-time BSL-3 Facility Manager to oversee the design, construction, commissioning, and operations of the high containment facility.

Please contact Mark Robbins, the Research Safety Bureau Chief, with any questions regarding BSL-3 or select agents research, and contact Shayne Barlow, the Director of DLAR and attending veterinarian, with questions on ABSL-3 research. For more details on the requirements for select agents & BSL-3/ABSL-3 research, please visit the Select Agent and Biosafety Level 3 Research website.

FEDERAL SELECT AGENT PROGRAM

The Federal Select Agent Program oversees the possession, use and transfer of biological select agents and toxins, which have the potential to pose a severe threat to public, animal or plant health or to animal

or plant products. The Federal Select Agent Program is jointly comprised of two regulatory oversight agencies:

- Centers for Disease Control and Prevention/Division of Select Agents and Toxins
- Animal and Plant Health Inspection Service/Agriculture Select Agent Services

The Federal Select Agent Program website contains a complete list of select agents and toxins

PERMISSIBLE TOXIN AMOUNTS

The following toxins are not regulated if the amount under the control of a principal investigator, treating physician or veterinarian, or commercial manufacturer or distributor does not exceed, at any time, the amounts indicated in the table below.

HHS Toxins [§73.3(d)(7)]	Amount
Abrin	1000 mg
Botulinum neurotoxins	1 mg
Short, paralytic alpha conotoxins	100 mg
Diacetoxyscirpenol (DAS)	10,000 mg
Ricin	1000 mg
Saxitoxin	500 mg
Staphylococcal Enterotoxins (Subtypes A, B, C, D, and E)	100 mg
T-2 toxin	10,000 mg
Tetrodotoxin	500 mg

All labs using toxins on the select agent list must maintain a biological toxins inventory. This inventory form is intended to carefully track the receipt, use, and storage of biological toxins to ensure the lab does not exceed the permissible toxin amount.

DUE DILIGENCE

“Due diligence” must be conducted and documented when transferring HHS biological toxins to other individuals. To minimize the risk of individuals stockpiling select toxins in amounts below the permissible amount, the Federal Select Agent Program developed the “due diligence” provision to ensure that all individuals use best practices when handling, storing, and transferring select agents and toxins. Sections 42 CFR §§ 73.3(d)(7)(i) and 73.16(l) of the Select Agent Regulations require the transferor to use due diligence when transferring an amount of an HHS select toxin otherwise excluded under the provisions of §73.3(d). This provision requires a transferor to take reasonable actions to ensure that the recipient is eligible to receive the select toxin and has a legitimate reason to handle or use the toxin. To document that due diligence has been performed, complete the university’s due diligence form and email the completed form to the Biological Safety Officer.

The transferor must also immediately report to the Federal Select Agent Program if they detect a known or suspected violation of Federal law or become aware of suspicious activity related to the select toxin. If violations are suspected, follow the reporting instructions on Federal Select Agent Program website.

SECTION 15: TRANSPORTING BIOHAZARDOUS MATERIALS

Biohazardous materials should be transported in such a way as to minimize exposure to infectious agents. When transporting materials, a secondary (outermost) container must be used. The outer container should be labeled with the contents. Biohazardous materials must never be left unattended during transport (in hallways, between buildings, in cars, etc.) and must always be under the supervision of lab personnel trained in proper spill and emergency procedures.

TRANSPORTATION IN SAME BUILDING (BETWEEN ROOMS, FLOORS)

For biohazardous materials transported between rooms or floors of the same building, a leak proof primary receptacle (vial, Eppendorf tube, etc.) and a sturdy, leak proof outer container must be used and labeled with a biohazard symbol if the materials are potentially infectious. When possible, potentially infectious substances should be moved using a wheeled cart. If a wheeled cart is not available or is not feasible to use, utilize the “one glove rule”- wear a glove on the hand that is carrying the biohazardous material and use the ungloved hand to touch common surfaces (door handles, elevator buttons, etc.). If a service/freight elevator is available, it should be used instead of a public elevator when transporting biohazardous materials.

TRANSPORTATION BETWEEN BUILDINGS ON SAME CAMPUS

For biohazardous materials being transported on foot to a different building, the materials must be packaged as indicated above. A wheeled cart is highly recommended when transporting between buildings. When using a wheeled cart, ensure that the materials are properly secured on the cart to prevent the containers from falling and the contents from spilling. If there is a potential for biological samples to spill during transport, then proper spill clean-up materials should be readily accessible.

TRANSPORTATION BETWEEN CAMPUSES OR FROM OTHER LOCATIONS

If biohazardous materials must be transported between campuses using a vehicle, the materials must be packaged in a way as to avoid escape of the material from the packaging. The outer container must be sturdy enough to keep the materials contained in the event the container is dropped or damaged. Public transportation, mopeds, scooters, bicycles, and motorcycles must never be used to transport biohazardous materials between campuses or on public roads. Biohazardous materials must never be left unattended during transport and must always be under the supervision of lab personnel trained in proper spill and emergency procedures. A spill kit must be present in the vehicle. Any individual(s) transporting samples must have knowledge of the emergency procedures in case of an accident or spill.

Patient specimens

Infectious or potentially infectious patient (human or animal) specimens transported by personal or university vehicle are considered materials of trade according to the Hazardous Materials Regulations (HMR). Materials of Trade are hazardous materials other than a hazardous waste, that are carried on a motor vehicle for purposes that include being carried by a private motor carrier to directly support a principal business that is not transportation (e.g., academic and laboratory research, pest control, plumbing, painting) (49 CFR 171.1). Because patient specimens are being transported for research purposes, transport directly from hospitals or clinics to university laboratories is a function not subject to the requirements of the HMR.

Infectious substance Material of Trade (49 CFR 173.6) is defined as: Division 6.2 material, other than a Category A infectious substance, contained in human or animal samples (including, but not limited to, secreta, excreta, blood and its components, tissue and tissue fluids, and body parts) being transported for research, diagnosis, investigational activities, or disease treatment or prevention, or is a biological product or regulated medical waste. When transported by motor vehicle in conformance with section 173.6 (Materials of Trade Exceptions), a material of trade is only subject to the requirements referenced in the following section:

- The material must be contained in a combination packaging (refer to the CDC schematic for packaging UN3373 Category B). For liquids, the inner packaging must be leakproof, and the outer packaging must contain sufficient absorbent material to absorb the entire contents of the inner packaging. For sharps, the inner packaging (sharps container) must be constructed of a rigid material resistant to punctures and securely closed to prevent leaks or punctures, and the outer packaging must be securely closed to prevent leaks or punctures. For solids, liquids, and sharps, the outer packaging must be a strong, tight packaging securely closed and secured against shifting, including relative motion between packages, within the vehicle in which it is being transported.
- Other than a regulated medical waste, the amount of Division 6.2 material in a combination packaging must conform to the following limitations:
    - One or more inner packagings, each of which may not contain more than 0.5 kg (1.1 lbs) or 0.5 L (17 ounces), and an outer packaging containing not more than 4 kg (8.8 lbs) or 4 L (1 gallon)
    - A single inner packaging containing not more than 16 kg (35.2 lbs) or 16 L (4.2 gallons) in a single outer packaging.
- For a regulated medical waste, a combination packaging must consist of one or more inner packagings, each of which may not contain more than 4 kg (8.8 lbs) or 4 L (1 gallon), and an outer packaging containing not more than 16 kg (35.2 lbs) or 16 L (4.2 gallons).
- The operator of a motor vehicle that contains a material of trade must be informed of the presence of the hazardous material (including whether the package contains a reportable quantity) and must be informed of the requirements of this section.

Patient specimens must be transported in such a way as to minimize exposure to infectious agents (packages must always remain upright and secured in the vehicle). The inner or outer container must be labeled with the contents and contact information for the lab. The secondary container is required by the Bloodborne Pathogens Standard to be labeled with the biohazard symbol. All outer labeling must be included on the package per DOT/IATA requirements for “Biological Substance, Category B”. Biohazardous materials must never be left unattended during transport and must always be under the supervision of lab personnel trained in proper spill and emergency procedures.

If dry ice will be used, the packaging must be constructed in a way to allow for the release of carbon dioxide gas and the outside must be labeled “Dry ice”. The package should be located as far away from the passengers as possible, and windows must be opened to allow for adequate ventilation of the cabin.

It is strongly recommended that only university vehicles be used in the transport of patient specimens. Consult with your Department Chair for guidance on the availability of a university vehicle and request approval from your Chair before using a personal vehicle for transporting specimens. If a personal vehicle will be used, it must be exclusively used to transport patient specimens during the transport operation (no stops in between). If an employee is approved by their Chair or supervisor to use their own vehicle, they are encouraged to speak with their auto insurance carrier for liability and coverage questions.

All spills or exposures during transport must be reported immediately to the supervisor and Environmental Health & Safety using the Laboratory Incident Report Form. A spill kit must be present in the vehicle. The person transporting patient specimens must have knowledge of the emergency procedures in case of an accident or spill.

SECTION 16: SHIPPING BIOLOGICAL MATERIALS

Shipping potentially infectious biological materials is strictly regulated and requires shipping training. If you will be shipping biological materials and will require shipping training, contact the Biological Safety Officer at smiths69@mailbox.sc.edu to receive access to the online training.

To complete the training required for shipping biological materials:

1. Contact the Biological Safety Officer for the link directly to the Internet Shipping Training. You will be provided a unique Activation Code that can be used to access the training program.
2. Select the training subjects that are applicable to the type of biological materials you will be shipping. You must select the appropriate subjects for your shipment.
3. Complete the applicable subjects included in the Online Shipping Training.
4. Complete the test to verify understanding of the regulations and print your certificate as a record to document your successful completion of training.
5. Notify the Biological Safety Officer that you have completed the required sections of the training program. (Note: Your training certificate must be signed by the Biological Safety Officer)

SHIPPING TRAINING PROGRAM

This training contains separate sections that cover the requirements for shipping the following materials:

- Category A Infectious Substances
- Category B Infectious Substances
- Exempt Human and Animal Specimens
- Genetically Modified (Micro)Organisms
- Dry Ice
- Dry and Wet Liquid Nitrogen Shippers
- Excepted Quantities

The training program will allow you to select specific training subjects based on the type of samples you are shipping, which ensures you to only receive training on subjects applicable to you. Please allocate sufficient time to complete all required sections of the training program prior to the date you plan to prepare your shipment. Contact the Biological Safety Officer at least 2 weeks prior to the shipping date.

RECORD OF TRAINING

After the training test is completed, it is scored automatically. You will be able to print a certificate as your record of training, documenting your test score, the specific areas tested on, as well as the test and expiration date of the certificate. USC requires a test score of at least 80% to receive a certificate and prior to shipping your samples (you may take retake the test as many times as needed). Training certificates must be made available for review upon request by USC staff or national authorities.

SECTION 17: PERMITS FOR BIOLOGICAL MATERIALS

The import, interstate movement, and export of biological materials is highly regulated and may require permits. If a permit is required for the transport of materials, it is the responsibility of the Principal Investigator to submit for application and obtain the necessary permits. Required permits should be acquired prior to requesting specimens. A copy of any required permits should be submitted to the Institutional Biosafety Committee (IBC) as part of the IBC protocol review process.

CDC Import Permit

A Center for Disease Control (CDC) Import Permit is required for the importation of biological materials that could cause disease in humans (human pathogen). The purpose of the CDC Import Permit Program (IPP) is to prevent the introduction and spread of human pathogens into the United States. The IPP ensures that the importation of human pathogens is monitored and that facilities receiving permits have appropriate biosafety measures in place to work with the imported agents or infectious materials. Materials requiring import permits include:

- 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)

Anyone needing to import infectious biological agents, infectious substances, or vectors will likely need to obtain a CDC import permit. The CDC provides an IPP eTool to help importers determine if a CDC permit is required, and also instructions on how to apply for an import permit. An inspection may be conducted before a permit is issued. The IBC protocol containing the permit will be evaluated to assess the applicant's facility safety measures to mitigate the risk of accidental release of a human pathogen.

U.S. Department of Agriculture (USDA) APHIS Permits

The USDA's Animal and Plant Health Inspection Service (APHIS) issues permits for the import, transit and release of regulated animals, animal products, veterinary biologics, plants, plant products, pests, organisms, soil, and genetically engineered organisms. ePermits is a web-based system for users to submit import/interstate movement/transit/release permit applications, track applications, apply for renewals and amendments, and receive copies of their permits.

Veterinary Services (VS) Permit (9 CFR, Part 122)

In general, a USDA VS permit is needed for materials derived from animals or exposed to animal-source materials. Materials which require a permit include:

- Certain live animals and animal products (*e.g.,* sperm and embryos)
- Animal tissues, blood, cells, or cell lines of livestock or poultry origin
- RNA/DNA extracts
- Hormones and enzymes
- Monoclonal antibodies for in vivo use in non-human species, certain polyclonal antibodies, and antisera
- Bulk shipments of test kit reagents
- Microorganisms including bacteria, viruses, protozoa, and fungi.
    - Partial list of VS-regulated livestock and poultry pathogens

Plant Protection and Quarantine (PPQ) Permits (7 CFR, Part 330)

A PPQ permit is required for the importation, interstate movement, and environmental release of:

- Plant pests including plant feeding insects, mites, snails, slugs, and plant pathogenic microorganisms (bacteria, viruses, fungi, etc.)
- Biological control organisms of plant pests and weeds
- Bees
- Parasitic plants
- Federally listed noxious weeds

A PPQ permit is also required for the importation and interstate movement of soil or other potentially infected host material for the purpose of isolating or culturing microorganisms from those materials. These materials include:

- Plant material
- Insects/arthropods
- Environmental samples (water, dust, sediments, etc.)

PPQ 526

The PPQ 526 permit is the primary permit for the movement or release of the above listed organisms and materials. The average application processing time is eighty (80) days. The processing time depends on the complexity of the request. A facility inspection may be required before a PPQ 526 Permit is issued. During an inspection, the PPQ inspector reviews the facility to determine if the facility and equipment are sufficient for containment of the organism(s).

Biotechnology Regulatory Services (BRS) Permit (7 CFR, Part 340)

A BRS permit is required for the importation, interstate movement, or environmental release of organisms developed using genetic engineering that may pose a plant pest risk, including plants, insects, or microbes.

Exports

Export control laws must be followed when shipping biological materials outside the United States. The university's Office of Research Compliance can assist researchers to comply with export control laws, but responsibility ultimately resides with the researcher. Refer to the Office of Research Compliance Export Control Regulations and Research website for more information.

SECTION 18: LAB BIOSECURITY

Biosecurity ensures the safe use and security of biological materials in laboratories. Lab biosecurity involves security measures taken to prevent the loss, theft, or intentional misuse of biological materials and research-related information, especially that pertaining to high-consequence biological agents and toxins (i.e., Select Agents). Biosecurity is accomplished by limiting access to facilities, research materials and information. Laboratories should always take steps to ensure microorganisms and valuable biological materials are secured and protected from unauthorized access, loss, theft, misuse, and intentional release.

Security measures that labs should take include:

1. Preventing unauthorized personnel from accessing lab areas
    - Lab doors should not be propped open and should be locked when unoccupied
2. Developing a lab response action plan that addresses:
    - Accidental release/theft or other potential emergency situations
    - Identify the individuals that:
        - are responsible for the agents
        - have access to the agents
        - are responsible for maintaining inventory logs
    - Material accountability procedures that prevent illicit removal of biological materials. Some examples include creating electronic data and record keeping procedures and conducting proper receipt and transfer of biological agents.
    - Procedures for how to handle breaches or near-breaches
3. Performing risk assessments to identify biological materials needing additional security provisions for their protection that may not be sufficiently covered through standard biosafety practices.
4. Regularly updating biological agent inventories with storage locations
5. Documenting internal and external transfers of biological agents within and between facilities
6. Documenting the inactivation and/or disposal of biological agents
7. Ensuring that electronic information pertaining to biological materials is safe from theft or misuse

DUAL USE RESEARCH OF CONCERN (DURC)

Certain types of life science research have the potential to be used for dual purposes: both benevolent and nefarious purposes. Using a broad definition, several types of biological research can be considered dual use research. Dual use research *of concern* is a subset of dual use research that the National Science Advisory Board for Biosecurity (NSABB) defines as “life sciences research that, based on current understanding, can be reasonably anticipated to provide knowledge, information, products, or technologies that could be directly misapplied to pose a significant threat with broad potential consequences to public health and safety, agricultural crops and other plants, animals, the environment, materiel, or national security.” The NSABB provides advice, guidance, and recommendations related to biosecurity and dual use biological research, including help informing U.S. policy regarding this type of research. The *United States Government Policy for Oversight of Life Sciences Dual Use Research of Concern* and the *United States Government Policy for Institutional Oversight of Life Sciences Dual Use Research of Concern* were developed to address the practices and procedures required to effectively identify and evaluate DURC. The scope of these policies is limited to research involving the use of 15 select agents and toxins.

Ultimately, oversight for DURC is a shared responsibility between the Principal Investigator, IBC, Biosafety Program, institution, and federal government. Principal Investigators must notify the IBC prior to conducting any research that could be considered DURC. While the scope of the DURC polices is limited to specific select agents and toxins, the U.S. Government recognizes that research outside the scope could also constitute DURC. Therefore, any biological research that could be considered dual use research of concern must be reviewed and approved by the IBC prior to conduct of this research. While research may be identified as DURC, it does not mean that it will not receive IBC approval and cannot be conducted.

APPENDICES

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APPENDIX A: ABBREVIATIONS/ACRONYMS

ABSA = American Biological Safety Association (Association for Biosafety and Biosecurity)

ABSL = Animal Biosafety Level

BSC = Biosafety Cabinet

BSL = Biosafety Level

BSO = Biological Safety Officer

CDC = Centers for Disease Control

DOT = Department of Transportation

EH&S = Environmental Health & Safety

HBV = Hepatitis B Virus

HEPA = High-efficiency Particulate Air

HHS = United States Department of Health and Human Services

HIV = Human Immunodeficiency Virus

IATA = International Air Transport Association

IBC = Institutional Biosafety Committee

NIH = National Institutes of Health

NSABB = National Science Advisory Board for Biosecurity

OSHA = Occupational Safety and Health Administration

OSP = Office of Science Policy

PCR = Polymerase chain reaction

PI = Principal Investigator

PPE = Personal protective equipment

r/s NA = Recombinant or synthetic nucleic acids

SCDES = South Carolina Department of Environmental Services

DPH = South Carolina Department of Public Health

SOM = School of Medicine

USC = University of South Carolina

USDA = United States Department of Agriculture

APPENDIX B: DEFINITIONS

Biohazard: microorganisms (bacteria, fungi, viruses, Rickettsiae, Chlamydiae and parasites), cells, plants, or animals into which recombinant DNA has been inserted; microorganisms or other agents that are actual or potential pathogens for humans, animals, or plants; select agents or toxins identified by the U.S. Departments of Health and Human Services (HHS) and of Agriculture (USDA)

Biosafety: the discipline addressing the safe handling and containment of infectious microorganisms and hazardous biological materials. The practice of safe handling of pathogenic micro-organisms and their toxins in the biological laboratory is accomplished through the application of containment principles and the risk assessment.

Biosecurity: measures taken to prevent unauthorized possession, loss, theft, misuse, diversion, and accidental or intentional release of high-consequence biological agents and toxins, and critical relevant biological materials and information within and from laboratories.

Decommissioning*:* the process of removing all hazards and decontaminating existing laboratory space and equipment prior to vacating a laboratory room.

Decontamination: the removal or inactivation of biological agents by physical or chemical means

Disinfection: chemical inactivation or destruction of viruses and vegetative bacteria but NOT bacterial spores

Laboratory*:* one or more room/s assigned to a PI, where research or teaching is conducted and where hazardous chemicals, biological materials, and/or radiological agents are used and/or stored.

Sterilization: a process, physical or chemical, that destroys or eliminates all forms of life, including spores

BLOODBORNE PATHOGENS STANDARD DEFINITIONS

Blood: human blood, human blood components and products made from human blood

Bloodborne Pathogens: pathogenic microorganisms that are present in human blood and can cause disease in humans. These pathogens include, but are not limited to, hepatitis B virus (HBV) and human immunodeficiency virus (HIV)

Contaminated: the presence or the reasonably anticipated presence of blood or other potentially infectious materials on an item or surface

Contaminated Sharps: any contaminated object that can penetrate the skin including, but not limited to, needles, scalpels, broken glass, and broken capillary tubes

Engineering Controls: controls (*e.g.,* sharps disposal containers, self-sheathing needles, safer medical devices, such as sharps with engineered sharps injury protections and needleless systems) that isolate or remove the bloodborne pathogens hazard from the workplace

Exposure Incident: a specific eye, mouth, other mucous membrane, non-intact skin, or parenteral contact with blood or other potentially infectious materials resulting from the performance of an employee's duties

Personal Protective Equipment: specialized clothing or equipment worn by an employee for protection against a hazard

Source Individual: any individual, living or dead, whose blood or other potentially infectious materials may be a source of occupational exposure to the employee

Universal Precautions: all human blood and certain human body fluids are treated as if they are known to be infectious for bloodborne pathogens

Work Practice Controls: controls that reduce the likelihood of exposure by altering the way a task is performed (*e.g.,* prohibiting recapping of needles by a two-handed technique)

Disinfectants destroy microorganisms by coagulating or denaturing proteins, injuring cell membranes and stopping normal enzymatic reactions. The range of susceptibility of microorganisms to disinfectants is relatively broad. A table summarizing the properties of commonly used chemical disinfectants is included below. Vegetative bacteria, fungi, and lipid-containing viruses are highly susceptible to all the agents listed. Non-lipid containing viruses are moderately resistant to these agents, while spore forms are the most resistant.

The EPA maintains lists of Selected EPA-Registered Disinfectants. These lists should be used as a reference point when selecting an appropriate disinfectant for biological agents used in the lab.

Disinfectant	Commonly Available	Preparations Effective Concentrations of Active Agents	Applications	Notes
Chlorine compounds	5.25% sodium hypochlorite (household bleach) *	0.5% (1:10 dilution, 5000 ppm); solution stable (~24hrs) if made with deionized water and protected from heat and light	Biohazard spills, contaminated instruments and glassware, liquid waste	-Broad spectrum with activity against non-lipid viruses, some bacterial spores -Highly reactive with organic matter; evolution of chlorine gas -For liquid waste add to a final conc. of 0.5% in a chemical fume hood
		0.05% (1:100 dilution, 500 ppm); solution unstable, make fresh dilutions as needed	Cleaned work surfaces; avoid use in biosafety cabinets	
Alcohols (ethyl or isopropyl)	95-100%	70-90%	Work surfaces, equipment surfaces, animal injection sites	-Flammable -Activity reduced by presence of organic matter
Phenolic compounds	Various concentrates and ready-to-use solutions or sprays ('Lysol' concentrate is 5-6% o-benzyl-p-chlorophenol)	0.2-3%	Biohazard spills, contaminated instruments and glassware, liquid waste	-Dilute with deionized water -For liquid waste add to a final conc. of at least 1% (collect as chemical hazardous waste)
Quaternary ammonium compounds	Various concentrates and ready-to-use solutions or sprays (benzalkonium chloride, Hyamine 3500)	0.1-2%	Work surfaces, equipment surfaces, contaminated glassware	-Often mixed with alcohols - Dilute with deionized water -Activity reduced by presence of organic matter and anionic detergents

* Household bleach is more effective when diluted. A 1:10 dilution is recommended for routine disinfection in most labs (*e.g.,* using a 1L container, thoroughly mix 900mL of deionized water with 100mL bleach. Bleach degrades over time and 10% bleach is potent for about 24hrs, so a 20% bleach solution is recommended when prepared weekly in a spray bottle used for routine disinfection. The bottle must be labeled with the date the solution was prepared and the concentration. A 20% bleach solution (1:5 dilution) is recommended when used to decontaminate large amounts of organic materials. Since bleach is corrosive, when using as a disinfectant on a stainless-steel surface (*e.g.,* in a biosafety cabinet) it is important to wipe down the metal surface with sterile water or ethanol after using the bleach solution.

AUTOCLAVE

Autoclaves use high pressure and temperature to achieve effective sterilization and caution must be used when operating them. All users must be trained prior to use and this training must be documented and records maintained with other lab training documentation. Laboratories and departments are encouraged to use USC's Autoclave Safety Training Guide when developing a training program for their autoclaves. This guide provides general information for the safe use of autoclaves. Since there are multiple brands of autoclaves on campus that have different program settings and operating procedures, users must receive specific training for the autoclave(s) they will be using. The Department Chair is responsible for notifying Principal Investigators if hands-on autoclave training should be provided by each Principal Investigator for their lab staff or if a departmental trainer will conduct the training.

Potential Hazards:

- Physical hazards: hot temperature and steam and boil over of hot liquids could cause burns. Pressurized vessels that are closed tightly could result in an explosion of the vessel potentially causing serious injury to people in the vicinity or damage to the autoclave

Exposure Hazards: exposure to infectious agents that have not been properly decontaminated through spills or splashes of liquids.

Safety Considerations:

- Equipment-specific training is required prior to use of autoclaves (there are many types of autoclaves on campus. Controls for different brands of autoclaves may have unique characteristics for loading, load sizes, and cycle types and settings. For this reason, it is important to receive hands-on training for the autoclave you will be using.
- Appropriate PPE must be worn. This includes autoclave safe gloves, lab coat, and eye protection.
- If a cycle has just completed, care should be taken when opening the autoclave as the escaping steam can cause serious burns.
- Incidents involving the autoclave resulting in injury or an exposure must be immediately reported to the PI/lab supervisor. Additionally, a laboratory incident report form must be filled out and sent to EH&S.

CENTRIFUGE

Centrifuges use centrifugal force to separate liquid and solid media suspensions based on differences in particle size and density. There are 3 types of centrifuges: microcentrifuge (~15,000 rpm), low/high speed (2,000-20,000 rpm), and ultracentrifuge (~120,000 rpm). All pose a hazard if not used correctly.

Potential Hazards:

- Physical hazard**:** mechanical failure (i.e., rotor damage, mechanical stress, etc.) can result in bodily injury or property damage
- Exposure hazard: individuals can potentially be exposed to infectious agents, hazardous chemicals, or radioactive substances

Safety Considerations:

- Lab-specific training is required prior to use of a centrifuge regardless of type.
- An SOP must be developed, printed out, and placed in lab safety binder for high-speed centrifuges/ultracentrifuges.
- Know your centrifuge! - review the centrifuge manual; keep the manual near the centrifuge
- Incidents involving the centrifuge resulting in injury or an exposure must be immediately reported to the PI/lab supervisor. Additionally, a laboratory incident report form must be filled out and sent to EH&S.
- Appropriate PPE must be worn.
- Regularly inspect the rotors and the inside of the centrifuge for any damage such as cracks, dents, or anything that could impair the function of the centrifuge.
- Before leaving the centrifuge, make sure that it is operating properly.
- Use sealed rotor heads and safety cups when infectious agents are centrifuged outside a BSC. Sealed rotor heads and safety cups should only be opened in a BSC.
- Never open a centrifuge before it has stopped spinning. After the centrifuge has stopped spinning, it should not be opened immediately in case of a spill inside and the potential aerosolization of its contents.

CRYOSTAT/MICROTOME

Cryostats and microtomes are used to thinly section tissues. Cryostats use a subzero temperature environment where tissues can be cut in temperatures as low as -30°C. Cryostats are also referred to as freezing microtomes.

Potential Hazards

- Sharps hazard: cutting blade is extremely sharp and can cause severe injury if handled incorrectly
- Cold hazard: subzero temperatures can result in cold burns/frostbite
- Biological hazard: individuals can potentially be exposed to infectious agents or recombinant DNA materials if cutting unfixed infectious tissues
- Ergonomics: repetitive motions and static/awkward postures can potentially lead to musculoskeletal disorders

Safety Considerations

- Lab-specific training is required prior to use of cryostats or microtomes.
- An SOP must be developed, printed out, and placed in lab safety binder.
- Know your cryostat/microtome! - review the cryostat/microtome manual; keep the manual nearby.
- Incidents involving the cryostat/microtome resulting in an injury or an exposure must be immediately reported to the PI/lab supervisor and as soon as possible to EH&S.
- Appropriate PPE must be worn.
- Blades must be handled with forceps, similar mechanical devices, or cut resistant gloves.

- In a cryostat, when handling the blade or specimen, the wheel safety lock must be in the locked position and the knife guard/anti-roll plate should be used to cover the cutting edge.
- Disposable blades must be disposed of in a biohazard sharps container. The sharps container should be located near the cryostat/microtome.
- Prior to cryostat cleaning, the wheel must be locked, and the blade removed
- Metal parts of a cryostat should not be touched with bare hands as low temperature can result in cold related skin injuries.
- Short stretch breaks are recommended when working for an extended period at a cryostat/microtome.

LAMINAR FLOW HOODS (CLEAN BENCHES)

Laminar flow hoods work by passing HEPA filtered air across the work surface to protect samples from airborne contamination. Horizontal laminar flow hoods provide filtered air from a HEPA filter that is situated behind the work surface while filtered air is directed towards the work surface from the top of the hood in vertical laminar flow hoods.

Safety Considerations:

- Due to the directionality of airflow in laminar flow hoods, only product protection is provided and not personnel protection.
- Clean benches can only be used for providing a sterile environment for non-biohazardous and non-toxic materials (*e.g.,* media preparation).
- Biohazardous materials or toxic materials and radionuclides must be handled in Class I/II biosafety cabinets or chemical fume hoods, respectively.

NOTE: Clean benches are not considered safety equipment. Therefore EH&S and the IBC do not require annual certification (although labs are encouraged to follow manufacturer certification recommendations).

APPENDIX E: CONTROLLED SUBSTANCE USE IN BIOLOGICAL RESEARCH

When using DEA controlled substances in biological research, laboratories must comply with federal and state regulations as well as institutional policies. Every person involved in using controlled substances must be registered with the U.S. Drug Enforcement Administration (DEA) and the South Carolina Department of Public Health (DPH). DPH's Bureau of Drug Control is responsible for enforcing the SC Controlled Substances Act which establishes the requirements for the use of controlled substances in South Carolina. The SC Controlled Substances Act requires that every person or entity who engages in controlled substances activity in South Carolina:

- **Obtain an annual registration from DPH**
- Register with the **U.S. Drug Enforcement Administration (DEA)** prior to engaging in such activity (Section 44-53-290).

PI Roles and Responsibilities

- Applying for and maintaining DPH and DEA permits
- Ensuring lab's compliance with federal and state regulations and institutional policies
- Properly securing DEA controlled substances
- Restricting access to only authorized users
- Maintain all required documentation
- Properly dispose of all expired, unused, or unwanted DEA controlled substances
- Promptly report missing or stolen materials to the appropriate authorities

Researcher Roles and Responsibilities

- Understanding and following DPH and DEA regulations
- Follow proper procedures for use, storage, and inventory tracking of DEA controlled substances

Secure Storage Requirements

Controlled substances must be kept and maintained in a secure location. Controlled substances should be stored in a securely locked safe or cabinet.

Record Keeping

Drug inventory and usage records must be maintained. Inventories and records for scheduled substances must be kept separate from other lab documents. All records relating to controlled substances must be maintained for the required amount of time established by the DEA and DPH.

APPENDIX F: FIELD RESEARCH- BIOSAFETY CONSIDERATIONS

Field research encompasses a broad range of research projects that can take place in a wide variety of locations. Below are recommendations for work that will involve animals or biological hazards.

Animals/Wildlife

Dangerous wildlife and pests exist worldwide. If handling wildlife is part of a research project, care must be taken to handle animals in a manner that avoids potential attacks or exposure to diseases. For questions regarding field research involving animals or IACUC approval requirements, consult with Shayne Barlow (barlows@mailbox.sc.edu, 777-8106).

To avoid pests in living spaces/quarters, food items should be stored in closed, plastic or other durable, leak-poof containers. Garbage or anything that could be a source of food for vermin or other pests should be kept away from campsites/living areas. The handling of sick or dead wildlife should be avoided.

Biological Hazards

There is potential for exposure to biological hazards in most outdoor areas. Biological hazards include harmful microorganisms that can cause vector-borne, zoonotic, and waterborne illnesses. Care should be taken in areas that have a high potential for vector-borne illnesses like Zika and West Nile fevers. Appropriate clothing and PPE should be worn in a way to avoid exposure to infected insects or animals (*e.g.,* pants tucked into boots, long sleeves to cover arms, etc.). Insect repellant should be considered as well. Avoid drinking untreated/unfiltered water that could harbor harmful bacteria or other microorganisms that may cause illness. If you have questions pertaining to biological hazards, contact the Biological Safety Officer, Sherika Smith, at smiths69@mailbox.sc.edu or 777-1625.

APPENDIX G: LAB CLOSE-OUT PROCEDURES FOR BIOLOGICAL MATERIALS USE

LABORATORY AND EQUIPMENT DECOMMISSIONING

This policy is applicable to all laboratories and auxiliary spaces serving as laboratories for users of biological materials. It provides measures for the removal of biohazards from laboratory spaces when the Principal Investigator (PI) is:

- leaving the University of South Carolina
- moving to another building on campus, or
- relocating to another laboratory within the same building
- disposing or transferring laboratory equipment that is no longer needed

When laboratories are vacated, all biological materials, in usable or waste form, must be disposed in a proper manner.

All laboratory equipment must be decontaminated before they are:

- placed back into service,
- transported and stored in another location, or
- disposed in a proper manner

Work surfaces and storage locations for biohazardous materials must be properly decontaminated.

PROCEDURES

1. PI will contact EH&S, preferably at least 4 weeks before the planned relocation date, by sending an email to Jocelyn Locke at 777-7650 or jlocke@mailbox.sc.edu or Sherika Smith at 777-1625 or smiths69@mailbox.sc.edu to request a decommissioning consultation.
2. Laboratory personnel must coordinate the removal of all biological agents. All equipment must be decontaminated and tagged with Equipment Decontamination Form. All biological samples and wastes must be properly labeled with wastes being decontaminated and disposed appropriately.
3. EH&S personnel will conduct a final inspection of the decommissioned laboratory. EH&S will notify, by email, the PI and Department Chair, of the closed-out status of the laboratory. If any non-conformances are identified, they must be addressed by the PI and a new inspection scheduled to complete the laboratory close-out process.

The status of the close-out process for the laboratory being decommissioned will be recorded in the EH&S Research Safety Management System.

RESPONSIBILITIES

Department Chair

- Notify Environmental Health & Safety (EH&S) when a PI plans to vacate a laboratory.
- Ensure PIs are aware of and follow the procedures contained in this policy.

- Be responsible for all costs associated with proper disposal and/or decontamination of hazardous materials or equipment that are remaining in the laboratory after the PI leaves the University.

Principal Investigator

- Notify EH&S of the plan to vacate the laboratory at least 4 weeks in advance and begin review of applicable items from the Laboratory Decommissioning Checklist.
- Before leaving, arrange for the transfer or disposal of biological materials. All biological waste must be decontaminated and disposed of in the appropriate receptacles. Refer to the Biological and Infectious Waste Management section for further guidance.
- Ensure that all laboratory rooms, storage areas, equipment and work surfaces are thoroughly cleaned before vacating the assigned space.
- Ensure that all laboratory equipment to be removed has a completed Equipment Decontamination Form attached to the equipment.
- Correct all non-conformances that remain after a decommissioning inspection by EH&S.

Environmental Health & Safety

- Provide guidance to lab personnel on how to perform activities listed on the Laboratory Decommissioning Checklist and verify each item is completed.
- Collect all biological wastes.
- Perform a decommissioning inspection as soon as applicable activities described in Laboratory Decommissioning Checklist have been completed, then notify the PI and Department Chair of any findings.

Facilities Management

- Ensure all equipment to be moved or repaired is tagged with Equipment Decontamination Form.
- Verify with EH&S that the lab space has been decommissioned before renovations begin.