Biological Safety Manual

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Environmental Health and Safety Office
April 2012

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Acronyms

ABSL	Animal Biosafety Level
ANSI	American National Standards Institute
APHIS	Animal and Plant Health Inspection Service
B Virus	Cercopithecine Herpesvirus
BMBL	Biosafety in Microbiological and Biomedical Laboratories
BSL	Biosafety Level
CDC	Centers for Disease Control and Prevention
CFR	Code of Federal Regulations
dBA	Decibel, A-weighted
DEQ	Department of Environmental Quality
DNA	Deoxyribonucleic Acid
DOT	Department of Transportation
EHS	Environmental Health and Safety Office
HBV	Hepatitis B Virus
HEPA	High Efficiency Particulate Air
HHS	Department of Health and Human Services
HIV	Human Immunodeficiency Virus
IACUC	Institutional Animal Care and Use Committee
IARC	International Agency for Research on Cancer
IATA	International Air Transport Association
IBC	Institutional Biosafety Committee
IRB	Institutional Review Board
LD	Lethal Dose
LS	Laboratory Supervisor
MSDS	Material Safety Data Sheet
MTA	Material Transfer Agreement
NHP	Non-human Primate
NIH	National Institutes of Health
NTP	National Toxicology Program
OSHA	Occupational Safety and Health Administration
OSP	Office of Sponsored Programs
OTT	Office of Technology Transfer
PPD	Purified Protein Derivative
PI	Principal Investigator
PLHCP	Physician or Other Licensed Healthcare Professional
PPE	Personal Protective Equipment
RG	Risk Group
RMW	Regulated Medical Waste
RNA	Ribonucleic Acid
RSC	Radiation Safety Committee
SIV	Simian Immunodeficiency Virus
SOP	Standard Operating Procedures
TB	Tuberculosis
USDA	United States Department of Agriculture

USDI	United States Department of Interior
UV	Ultraviolet
VAC	Virginia Administrative Code

Foreword

The Laboratory Safety group within the Environmental Health and Safety Office (EHS) has developed the *Biological Safety Manual* to provide guidance on the safe handling of biological materials, safety practices to minimize the risk of exposure to potentially infectious materials, and procedures for proper acquisition, use, storage, transfer, and disposal of biological materials. For laboratories operating at Biosafety Level (BSL) 3, an additional laboratory-specific operations and procedures manual is provided (e.g., *Biomedical Research Laboratory Biosafety Plan*). The recommendations and requirements provided in this manual are based on guidance from regulatory agencies and current professional standards. The *Biological Safety Manual* is intended for use with the *Laboratory Safety Manual*, which provides guidance relevant to all instructional and research laboratories at George Mason University. This manual supersedes all other documents regarding biological safety at George Mason University.

The *Biological Safety Manual* is reviewed annually by EHS and revised as necessary to reflect changes in George Mason University policies and government regulations.

Document History

Version	Date	Comments
1	July, 2006	Initial *Biological Safety Manual*
2	November, 2007	Annual review and update
3	April, 2012	Review and update

1.0 Introduction

Instructional and research laboratories contain hazards that must be properly managed in order to minimize the risk they pose to health, safety, and the environment. These hazards include exposure to hazardous substances (e.g., chemicals, biological material, and radioactive material) and physical hazards associated with equipment and instruments used by laboratory personnel. This manual addresses hazards associated with laboratory work involving biological material.

1.1 Biological Material

The term biological material, as used in this manual, is a general term referring to all prokaryotic and eukaryotic organisms (and their components), viruses, subviral agents, recombinant deoxyribonucleic acid (DNA), and biologically-derived toxins used in research and instructional laboratories. For the purposes of biosafety, it is useful to categorize biological material as *biohazardous* or *nonbiohazardous*, and to accurately assess the risks involved in working with each type biological material present in the laboratory.

1.1.1 Biohazardous Material

Biohazardous material includes all infectious agents, vectors known to carry and transmit infectious agents, infected or potentially-infected animals, infectious material, recombinant DNA, and biologically-derived toxins that present either a risk or a potential risk to the health of humans, animals, or plants either directly through infection or indirectly through damage to the environment.

- **Infectious agents** include human, animal, and plant pathogens (bacteria, parasites, fungi, viruses, subviral agents).

- **Infectious material** includes infectious agents and all biological material that contains, or has the potential to contain, infectious agents. Examples of infectious material include all human or nonhuman primate (NHP) material (e.g., blood and other body fluids, organs, tissues, cultured cells), infected animals and materials from infected animals, and environmental samples likely to contain infectious agents.

- **Recombinant DNA** molecules are considered biohazardous if they are nonexempt from the National Institutes of Health (NIH) *Guidelines for Research Involving Recombinant DNA* (NIH Guidelines). Examples include recombinant DNA that is formed by the deliberate transfer of a drug-resistance trait to microorganisms that are not known to acquire the trait naturally (if that transfer could compromise the use of the drug to control disease agents in humans, veterinary medicine, or agriculture); is designed for use in human gene transfer experiments; contains genes for the biosynthesis of toxic molecules lethal for vertebrates at a median lethal dose (LD_{50}) of less than 100 ng/kg body weight (e.g., microbial toxins such as the botulinum toxins, tetanus toxin, diphtheria toxin, and *Shigella dysenteriae* neurotoxin); is designed for the generation of transgenic plants or animals; or contains infectious DNA or ribonucleic acid (RNA) viruses or defective DNA or RNA viruses in the presence of helper virus. The NIH Guidelines are available on the EHS website.

- **Biologically-derived toxins** include all molecules produced by animals, plants, microorganisms or other agents that have a median lethal dose (LD_{50}) value of <50 mg/kg (oral administration in rats). Examples are bacterial exotoxin, plant lectins (ricin), and mycotoxins (aflatoxins, sterigmatocystin, luteoxkyrin, rugulosin, patulin, etc.).

1.1.2 Nonbiohazardous Material

Biological materials that are not normally infectious are considered nonbiohazardous. This includes nonpathogenic microorganisms, viruses, and subviral agents; plants and NHP animals (except those listed under biohazardous material above), biological material not likely to contain infectious agents, recombinant DNA molecules exempt from NIH Guidelines, environmental samples not likely to contain infectious agents, and biologically-derived, nontoxic molecules.

1.2 Regulations, Guidelines, and Permit Requirements

The procurement, handling, storage, and disposal of biological materials are regulated by federal, state, and local agencies. The following is a summary of regulations and guidelines from various government entities regarding the use of infectious agents, biologically-derived toxins, and recombinant DNA molecules in the United States.

- **United States Department of Agriculture (USDA)**
  - **Animal and Plant Health Inspection Service (APHIS):**
    - *Possession, Use, and Transfer of Biological Agents and Toxins; Final* Rule (Title 7 of the Code of Federal Regulations Part 331 (7 Code of Federal Regulations [CFR] Part 331), 9 CFR Part 121) – requirements as established by USDA, APHIS to control those agents and toxins that have been determined to pose a severe threat to human, animal, and plant health. Includes requirements regarding registration, security risk assessments, biosafety and security plans, training, transfers, and documentation.

    - *Organisms and Vectors* (9 CFR Part 122) – permit requirements for zoonotic agents imported into the United States and subsequently transported domestically.

    - *Introduction of Organisms and Products Altered or Produced Through Genetic Engineering Which are Plant Pests or Which There is Reason to Believe are Plant Pests* (7 CFR 340) - regulates, any living stage (including active and dormant forms) of insects, mites, nematodes, slugs, snails, protozoa, or other invertebrate animals, bacteria, fungi, other parasitic plants or reproductive parts thereof; viruses; or any organisms similar to or allied with any of the foregoing; or any infectious agents or substances, which can directly or indirectly injure or cause disease or damage in or to any plants or parts thereof, or any processed, manufactured, or other products of plants. The introduction into the United States of such articles also may be subject to other regulations promulgated under the Plant Protection Act (7 USC 7701- 7772) and found in 7 CFR parts 319, 330, and 360.

- **Department of Commerce**
  - **Bureau of Industry and Security:**
    - *Commerce Control List* (15 CFR Parts 730 – 774) – items including commodities, software, and technology that are regulated for export control purposes.

- **Department of Health and Human Services (HHS)**
  - **Centers for Disease Control and Prevention (CDC):**
    - *Foreign Quarantine* (42 CFR Part 71, specifically Part 71.54 etiologic agents, hosts and vectors) – regulatory guidance on permit requirements for the importation and subsequent domestic transportation of agents that cause disease in humans.

    - *Possession, Use and Transfer of Select Agents and Toxins* (42 CFR Part 73) – requirements regarding the possession or use in the United States, receipt from outside the United States, or transfer within the United States, of select agents and toxins. The requirements are designed to implement provisions of the Public Health Security and Bioterrorism Preparedness and Response Act of 2002 (Public Law 107–188). The act was designed to provide protection against the effects of misuse of select agents and toxins whether inadvertent or the result of terrorist acts against the United States homeland or other criminal acts.

  - **National Institutes of Health (NIH):**
    - *Biosafety in Microbiological and Biomedical Laboratories* (BMBL) (written in conjunction with the Centers for Disease Control and Prevention [CDC]) – combinations of standard and special microbiological practices, safety equipment, and facilities for biosafety levels and containment necessary for working with infectious agents.

    - *Guidelines for Research Involving Recombinant DNA Molecules* (NIH Guidelines) – guidelines for the safe conduct of research involving construction and handling of recombinant DNA and organisms containing recombinant DNA. NIH requires all institutions that receive NIH funding to ensure that research involving recombinant DNA (regardless of the source of funding for the research) complies with the NIH Guidelines.

- **Department of Labor**
  - **Occupational Safety & Health Administration (OSHA):**
    - *Bloodborne Pathogens* Standard (29 CFR 1910.1030) – regulatory guidance on the occupational health risks caused by exposure to human blood and other potentially infectious human materials.

- **Department of Transportation (DOT)**
  - **Research and Special Programs Administration:**
    - *Hazardous Materials Regulations* (49 CFR Parts 171-180) – documentation and packaging requirements for the transport of hazardous materials.

- **The National Research Council**
  - *Guide for the Care and Use of Laboratory Animals* (The Guide; written in conjunction with the Institute of Laboratory Animal Research, Commission on Life Sciences) – addresses areas that require policy attention: the role and function of the Institutional

Animal Care and Use Committee (IACUC), protocols for animal care and use, occupational health and safety, personnel qualifications, and other areas.

- *Occupational Health and Safety in the Care and Use of Research Animals* (written in conjunction with NIH) – an implementation handbook and companion to The Guide, identifies principles for building a program and discusses the accountability of institutional leaders, managers, and employees for a program's success and provides a detailed description of risks (e.g., physical and chemical hazards, allergens and zoonoses, and hazards from experiments).

- **Public Law 107–56 *USA PATRIOT Act of 2001*** – places restrictions on persons who possess select agents and provides criminal penalties for possession of such agents that cannot be justified for specified peaceful purposes.

- **Public Law 107-188 *Public Health Security and Bioterrorism Preparedness Response Act of 2002*** – designed to improve the ability of the United States to prevent, prepare for, and respond to bioterrorism and other public health emergencies.

- **Virginia Administrative Code (VAC)**
  - **Department of Environmental Quality (DEQ):**
    - *Regulated Medical Waste Management Regulations* (9VAC20-120) – outlines specific waste management procedures for treating, storing, disposing of, and handling of regulated medical waste (RMW).

1.3 Roles and Responsibilities

It is the responsibility of George Mason University employees, affiliates, students, and visitors to conduct laboratory work and activities in a manner that will not adversely impact themselves, other personnel, students, the surrounding community, George Mason University property, or the environment (University Policy Number 1406). The *Laboratory Safety Manual* outlines specific responsibilities for various George Mason entities with regard to laboratory safety and compliance. Responsibilities specific to biological safety are outlined in the following sections.

1.3.1 Vice President for Research and Economic Development

The University President delegates authority to the Vice President for Research and Economic Development, who is charged with overseeing all aspects of laboratory compliance with regard to environmental health and safety. A list of responsibilities is provided in *Laboratory Safety Manual*. Specific responsibilities with regard to biological safety are to:

- Approve and oversee all plans, policies, and procedures related to laboratory compliance with regard to environmental health and safety for instructional and research laboratories at George Mason University.
- Oversee EHS.
- Appoint members to the Institutional Biosafety Committee (IBC).
- Appoint Responsible Official and Alternate Responsible Official to oversee the *Biomedical Research Laboratory Select Agent Program*.

- When necessary, enforce sanctions regarding laboratory noncompliance as recommended by the IBC, Radiation Safety Committee (RSC), or EHS.

1.3.2 Environmental Health and Safety Office (EHS)

EHS partners with various George Mason officials, departments, and personnel to promote health, safety, environmental protection, and compliance with applicable local, state, and federal regulations. This mission is accomplished by establishing environmental, health, and safety policies and procedures; providing training; implementing preventive actions; and ensuring continuous improvement of George Mason's health and safety programs. The EHS Laboratory Safety Program provides services to assist laboratories in meeting regulatory requirements and minimizing the risks associated with laboratory hazards. A full list of EHS responsibilities is provided in the *Laboratory Safety Manual*.

1.3.2.1 Laboratory Safety Personnel

The Laboratory Safety group within EHS is responsible for administering the university's Biological Safety Program. Specific responsibilities with regard to biological safety are to:

- Maintain current knowledge of laboratory safety regulations and guidelines.
- Provide technical guidance in the development of policies and procedures regarding biological safety.
- Inspect biological laboratories to monitor compliance with laboratory safety policies and procedures.
- Provide biological safety training for laboratory personnel and other related training upon request.
- Serve as members of the IBC and coordinate IBC activities.
- Evaluate work area designs to determine suitability of a space for proposed work.
- Assist Principal Investigators/Laboratory Supervisors (PI/LS) in evaluating and controlling biohazards.
- Implement corrective actions as necessary.
- Manage biological waste generated in instructional and research laboratories.
- Continually evaluate and improve the university's Biological Safety Program.
- Notify the Office of Research Integrity and Assurance of any projects which may include use of animals or human subjects.
- Provide one officer-level employee to serve as a member of the IACUC.

1.3.2.2 Occupational Health and Safety Personnel

The Occupational Health and Safety group within EHS is responsible for supporting EHS-Laboratory Safety personnel in providing a safe and healthful laboratory environment. In addition to the specific responsibilities with regard to biological safety outlined below, the Director of Occupational Health and Safety serves as members of IBC and IACUC.

- Provide technical guidance in the development of policies and procedures regarding biological safety.
- Provide *Bloodborne Pathogens Safety Training* and *Respiratory Protection Training* to laboratory personnel.

- Administer the *Medical Surveillance Plan* for laboratory personnel to include, but not limited to, approving laboratory personnel for vaccination based on risk; selecting respiratory protection equipment and performing fit tests when needed; and initiating the *Animal Handler Questionnaire*.
- Coordinate with a physician or other licensed healthcare professional (PLHCP) to provide medical surveillance to laboratory personnel.
- Conduct exposure monitoring when necessary or upon request.
- Conduct root-cause analysis following laboratory and research animal-related injuries and accidents.

1.3.3 Office of Sponsored Programs (OSP)

OSP is the pre-award and post-award office for all externally sponsored projects at George Mason University. OSP provides assistance in proposal budget development and proposal submission. OSP also reviews and signs all proposals to external sponsors. All contracts with George Mason University which involve sponsored projects are reviewed, negotiated, and executed by OSP. Specific responsibilities of OSP are to:

- Notify EHS of awarded projects that involve laboratory hazards, as indicated on the *Proposal Approval Routing Form* (available on the OSP website).
- Include in all contractual agreements with other entities requirements for compliance with university, local, state, and federal rules and regulations for hazardous substances.
- Notify EHS of all Material Transfer Agreements (MTA) involving hazardous substances.

1.3.4 Office of Technology Transfer (OTT)

OTT advises faculty, students, and staff on issues related to intellectual property and provides the university's bridge to the George Mason Intellectual Properties, Inc., which manages the protection and commercialization of the university's patents, copyrights, and know-how.

1.3.5 Office of Research Integrity and Assurance

Office of Research Integrity and Assurance handles the administrative management for research with human subjects and the care and use of animals in research. This office also supports the Institutional Review Board (IRB), formerly the Human Subjects Review Board, and the IACUC. Specific responsibilities of the Office of Research Integrity and Assurance are to:

- Work with EHS and relevant safety committees on university policy and regulatory compliance regarding laboratory hazards.
- Notify EHS of any IRB or IACUC protocols that may include use of laboratory hazards.

1.3.6 Student Health Services

Student Health Services offers confidential health services that include components of diagnosis, treatment, self care, health promotion, and disease prevention. The goal of Student Health Services is to provide all currently enrolled students with high quality health care. In addition, Student Health Services provides professional consultation in the event of potentially hazardous exposures and administers various vaccinations to faculty, staff, and students as medically indicated.

1.3.7 Physician or Other Licensed Healthcare Provider (PLHCP)

A PLHCP is an individual whose legally-permitted scope of practice (i.e., license, registration, or certification) allows them to independently provide, or be delegated responsibility to provide, some or all of the health care services required in this Program. Specific responsibilities of PLHCP related to laboratory safety are to:

- Provide required medical surveillance to employees upon request by EHS, including necessary vaccinations.
- Maintain records of all medical surveillance and follow-up medical care.
- Provide medical care to employees in the event of an exposure, injury, or development of allergy symptoms.
- Fill out and return the *Student Medical Clearance* form as necessary.

1.3.8 George Mason University Veterinarian

The George Mason University Veterinarian is responsible for developing and implementing an effective program of veterinary care for research animals. Specific responsibilities of the Veterinarian related to biological safety are to:

- Provide guidance and training to university personnel on procedures for the safe handling, immobilization, sedation, analgesic, anesthesia, and euthanasia of laboratory and research animals.
- Serve on the IACUC and review proposed animal use.
- Provide guidance and training on animal husbandry and Animal Biosafety Level (ABSL) 2 and 3 standard practices.
- Provide veterinary guidance, training, and oversight on animal surgery procedures.
- Oversee all aspects of animal care and use.

1.3.9 Waste Management Facility Operator

George Mason University is required by DEQ to have Waste Management Facility Operators on staff for each campus where autoclaves are used to treat biohazardous waste. Currently, both the Fairfax and Prince William campuses have a Waste Management Facility Operator whose responsibilities are to:

- Receive Class I and Class III Waste Management Facility Operator training and pass the exam as required by Virginia Code 9VAC20-120.
- Maintain current Waste Management Facility Operator license for George Mason University. Continuing professional education or training may be required for license renewal.
- Notify EHS when changes to the license are necessary.
- Verify that waste autoclaves are functioning properly:
  - Perform indicator organism tests monthly on each waste autoclave and maintain indicator organism log. The Waste Management Facility Operator may assign this responsibility to a designee.
  - Verify that autoclave performance standards are being met as outlined in Virginia DEQ RMW Regulation 9VAC20-120.
- Maintain autoclave user logs according to 9VAC20-120.

- Coordinate maintenance when necessary.
- Maintain service contracts for autoclaves and forward annual certification documentation to EHS.
- See that un-autoclaved waste is not left unattended in unrestricted areas and that it is placed on carts provided by the EHS instead of the floor.

1.3.10 Institutional Biosafety Committee (IBC)

The IBC is an advisory committee at George Mason University dedicated to excellence in the science and practice of biological safety. Committee members review research proposals to assess potential risks and ensure proper practices and procedures are used to mitigate the risks.. Specific responsibilities of the IBC are to:

- Require that all instructional and research projects involving the acquisition, use, storage, or disposal of recombinant DNA comply with the NIH Guidelines, as well as all applicable federal, state, and local regulations and guidelines.
- Require that all instructional and research projects involving the acquisition, use, storage, or disposal of biohazardous materials comply with the BMBL as well as all applicable federal, state, and local regulations and guidelines.
- Require that all instructional and research projects involving the acquisition, use, storage, or disposal of biologically-derived toxins having an LD_{50} < 50mg/kg (oral administration in rats) comply with the BMBL as well as all applicable federal, state, and local regulations and guidelines.
- Advise EHS and the Vice President for Research and Economic Development on the development of policies and procedures concerning the use of biological materials in instructional and research laboratories.
- Advise EHS and Vice President for Research and Economic Development regarding training, experience and qualifications of individuals who work with or in the vicinity of recombinant DNA, biohazardous materials, and biologically derived toxins in instructional and research laboratories.
- Recommend to the Vice President for Research and Economic Development sanctions on any individual whom it determines has violated the terms of an approved protocol, has conducted projects subject to its authority without gaining appropriate IBC approval, or has otherwise violated any provision of applicable federal, state, and local regulations and guidelines, or institutional policies regarding subjects under its purview.

1.3.11 Institutional Animal Care and Use Committee (IACUC)

The IACUC is a federally-mandated committee, qualified through the experience and expertise of its members that oversees its institution's animal program, facilities, and procedures. All faculty, staff and students working with animals must submit an application to the IACUC prior to initiation of the work for review and approval.

Specific responsibilities of the IACUC related to biological safety are to:

- Review proposed animal use while considering biological safety requirements (e.g., training, vaccinations, personal protective equipment [PPE], etc.).
- Conduct semiannual inspections of facilities and animal use areas.

- Review and investigate concerns about animal care and use at George Mason University.
- Review and approve research by George Mason University employees being conducted at other institutions.
- Educate animal researchers with regard to the use of safe devices, zoonoses, PPE, etc., as needed.

1.3.12 Deans or Directors

Each Dean or Director has the responsibility for overseeing all instructional and research laboratory work and activities in their unit. Deans and Directors may delegate these responsibilities to departmental chairpersons. Specific responsibilities of Deans and Directors with regard to biological safety are to:

- Provide necessary resources for safety related items (e.g., PPE, flushing emergency showers and eyewashes).
- Sign Project Review Form approval letters as necessary to acknowledge the commencement or continuation of research and instructional projects in their unit.
- Carry out additional responsibilities outlined in the *Laboratory Safety Manual*.

1.3.13 Principal Investigator (PI)/Laboratory Supervisor (LS)

PI/LS are responsible for overseeing all laboratory work, activities, and employees in their laboratory. It is the responsibility of the PI/LS to ensure that George Mason University laboratory safety policies and procedures and good safety practices are followed, applicable safety regulations are considered, appropriate safety equipment is made available, and necessary safety training is completed. PI/LS who oversee research or instructional laboratories must attend safety training in order to become familiar with the laboratory safety program at the university. A full list of responsibilities is provided in the *Laboratory Safety Manual*. However specific responsibilities with regard to biological safety and safety in animal spaces are listed below.

- Ensure that personnel under their supervision attend safety training.
- Ensure that personnel under their supervision complete required medical surveillance.
- Provide IBC and IACUC with information on potential hazards associated with proposed research.
- Develop Supplemental Laboratory Standard Practices for all work areas where research animals are cared for or used.
- Ensure that all exposures, injuries, accidents, and suspected allergic symptoms resulting from the care or use of laboratory or research animals are reported using the *First Report of Accident* form.

1.3.14 Laboratory Personnel

A comprehensive list of responsibilities for PI/LS and other laboratory personnel is provided in the *Laboratory Safety Manual*. Individuals working in laboratories where biohazardous materials are used or stored are responsible for implementing appropriate measures to adequately contain materials and to prevent exposure to potentially infectious material. Laboratory personnel should be familiar with the contents of this manual, the *Laboratory Safety Manual*, relevant government regulations, and George Mason University policies and procedures.

Animal handling personnel include employees, affiliates, visiting researchers, contractors, students, and volunteers whose duties include working with or near laboratory or research animals.

1.3.14.1 Support Services Staff

Support services staff includes housekeeping, facilities personnel, security personnel, and police who work in areas containing hazardous materials, but who do not work directly with these materials. Specific responsibilities are to:

- Attend relevant safety training (e.g., Laboratory Safety Awareness for Police, Laboratory Safety Awareness for Facilities, Housekeeping Safety, Hazard Communication, etc.) and subsequent refresher training as required.
- Take precautions to avoid contact with hazardous material.
- Report unsafe conditions involving hazardous materials to their supervisor and EHS.
- Be familiar with and use proper PPE needed for safety.
- Request assistance from their supervisor or EHS when uncertain about the risks related to hazardous materials.

2.0 Assessment and Management of Risk

As with any laboratory procedure, a certain degree of risk is associated with work involving biological materials. Accurate assessment of the risk involved and the implementation of measures to effectively manage that risk are critical components of biosafety. In the context of biological research and instructional laboratories, risk assessment focuses primarily on the prevention of laboratory-associated infection. Risk management is the application of appropriate engineering controls, administrative controls, PPE, and safety practices to reduce or eliminate the potential for exposures.

The assessment and management of risk is an ongoing process and must be continually evaluated to reflect changes in the quantity or type of biological material present in the laboratory, types of procedures to be performed, and current recommendations or requirements from government agencies regarding safe laboratory practices.

2.1 Risk Assessment

Risk assessment is a process used to identify the hazardous characteristics of a known infectious or potentially infectious agent or material, the activities that can result in exposure to an agent, the likelihood that such exposure will cause laboratory associated infections, and the probable consequences of such an infection. The information identified by risk assessment will provide a guide for appropriate containment level, microbiological practices, safety equipment, and facility safeguards. The PI/LS is specifically and primarily responsible for assessing risk and applying appropriate management strategies. EHS personnel and the IBC can be of great assistance in the risk assessment process.

PI/LS must perform risk assessments that consider the types of hazards present in the laboratory, the risk of exposure to laboratory personnel, and the type of work to be performed. Prudent planning is a critical component of risk assessment. One recommended approach to performing a risk assessment is a five-step process outlined below. Examples of each step are provided for additional guidance.

1. Identify agent hazards and perform an initial risk assessment.
    - Pathogenicity and viability of any biological agents present.
    - Potential routes of exposure.
    - Availability of preventative measures and effective treatments for the disease.
    - Type and volume of biological materials.

2. Identify laboratory procedure hazards.
    - Potential for a harmful personal exposure to occur.
    - Potential for release of a hazardous substance to the environment.
    - Potential for production of harmful byproducts.

3. Make a final determination of the appropriate biosafety level and select additional precautions indicated by the risk assessment.
    - Appropriate PPE.
    - Precautions associated with generation of waste.

- Proper storage of materials, waste, and equipment.

4. Evaluate the proficiencies of staff regarding safe practices and the integrity of safety equipment.
   - Level of training and experience of personnel.
   - Use and condition of laboratory equipment.
   - Availability of safety equipment such as biosafety cabinets.
   - Appropriate response procedures in the event of an emergency.

5. Review the risk assessment with EHS.

Risk assessment can be qualitative or quantitative. In the presence of known hazards, quantitative assessments can be performed. In many cases, however, quantitative data will be incomplete or absent (e.g., investigation of an unknown agent or receipt of an unlabeled sample). Types, subtypes, and variants of infectious agents involving different or unusual vectors, the difficulty in measuring the amplification potential of an agent, and the unique considerations of genetic recombinants are a few of the challenges to be considered when assessing risk.

2.1.1 Risk Groups (RG)

The World Health Organization (WHO) has recommended RG classification for biological agents based on an agent's capability to infect and cause disease in a susceptible human or animal host, its virulence as measured by the severity of disease, and the availability of preventative measures and effective treatments for the disease. The four RG address the risk to both laboratory personnel and the community. In addition, the NIH Guidelines established a comparable classification system grouping human etiological agents into four RG on the bases of hazard (Table 1). RG classifications for specific infectious agents are listed on the EHS website. In the event that several RG classifications are listed for an agent, the highest RG number should be used for the risk assessment.

Table 1: Classification of Biological Agents by RG

RG Classification	NIH Guidelines for Research Involving Recombinant DNA Molecules, 2011	World Health Organization Laboratory Biosafety Manual, 3rd Edition, 2004
RG 1	Agents that are not associated with disease in healthy adult humans.	Agents unlikely to cause human or animal disease (No or low personnel and community risk).
RG 2	Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available.	Agents that can cause human or animal disease but are unlikely to be a serious hazard to laboratory personnel, the community, livestock, or the environment. Laboratory exposures may cause serious infection, but effective treatment and preventive measures are available and the risk of spread of infection is limited (moderate individual risk; low community risk).

RG Classification	NIH Guidelines for Research Involving Recombinant DNA Molecules, 2011	World Health Organization Laboratory Biosafety Manual, 3rd Edition, 2004
RG 3	Agents that are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available (high risk to laboratory personnel but low community risk).	Agents that usually cause serious human or animal disease but are not ordinarily spread from one infected individual to another. Effective treatment and preventive measures are available (high individual risk; low community risk).
RG 4	Agents that 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).	A pathogen that usually causes serious human or animal disease that can be readily transmitted from one individual to another, directly or indirectly. Effective treatment and preventive measures are not usually available (high individual and community risk).

Federally-licensed vaccines containing live bacteria or viruses are not subject to RG classifications. However, these classifications are applicable to cultures of the strains used for vaccine production, or further passages of the vaccine strains.

2.1.2 Laboratory Exposure

For an infectious agent to cause disease, it must first enter or invade the body in sufficient numbers. Individuals working with infectious material must be aware of possible routes of exposure and should implement procedures and practices that reduce their risk of exposure.

Common routes of exposure in the laboratory include inhalation, ingestion, and direct contact with open skin or mucous membranes.

- **Inhalation** of aerosolized infectious material is the most common cause of laboratory-associated infection. In addition, laboratory personnel may be exposed to animal allergens via the inhalational route. Aerosols can be produced during vortexing, sonication, homogenization, electroporation, pipetting, popping tube caps, flame-sterilizing instruments, flow cytometry, centrifugation, shaking or vigorously stirring cultures, changing animal bedding or handling infected animals.

- **Ingestion** of biohazardous material most often occurs as the result of poor hygiene and poor laboratory practices, such as eating, drinking, or smoking in the laboratory, transfer of material to mouth by contaminated hands or articles, or mouth pipetting.

- **Percutaneous exposure** may result from a needlestick puncture with a contaminated sharp object, bites from an animal or arthropod vector, or through wounds, abrasions, and eczema.

- **Permucosal exposure** can occur by touching mucous membranes with hands that have been in contact with contaminated surfaces (such as bench tops, phones, computers, etc.) or with hands that were not washed after working in a laboratory where biohazardous material is used.

2.1.3 Biological Laboratory Hazards

Biological laboratories inherently contain various hazards. The following sections outline specific hazards related to infectious material, sharps, hazards associated with research animals and animal laboratories. Additional laboratory hazards are outlined in the *Laboratory Safety Manual*.

2.1.3.1 Infectious Material

Infectious material includes infectious agents and all biological material that contains or has the potential to contain infectious agents. Examples of infectious material include all human or NHP material (e.g., blood and other body fluids, organs, tissues, cultured cells), infected animals and material from infected animals, and environmental samples likely to contain infectious agents. It is important to understand the potential for exposure to infectious material in biological laboratories.

- **Aerosols:** Aerosols are solid particles or liquid droplets, ranging in diameter from 0.01 to a few microns, suspended in a gaseous medium (e.g., air). Aerosols are a serious hazard because they are generated by a variety of laboratory procedures and equipment and are not easily detected by laboratory personnel. When inhaled, aerosols are retained in the lungs creating an exposure hazard for laboratory occupants and for individuals occupying adjacent spaces open to airflow from the laboratory. Procedures such as pipetting, homogenization, sonication, centrifugation, and vortexing are proven sources of aerosols.

- **Cell Culture:** Laboratory personnel who handle or manipulate human or animal cells and tissues are at risk for exposure to potentially infectious latent and adventitious agents that may be present in these cells and tissues. The CDC has documented cases of inadvertent transmission of disease to organ recipients, accidental transplantation of human tumor cells to healthy workers, and transmission of disease to laboratory personnel from poorly characterized human and animal cell lines. Because of the potential for human cell lines to harbor bloodborne pathogens, OSHA has included human cell lines under the Bloodborne Pathogens Standard.

- **Animal Bites and Scratches:** Personnel working with or near laboratory or research animals should be aware of the risk of animal bites or scratches. Animals respond to sights, smells, and loud or high-pitched noises. Animals may attempt to escape when approached by an animal handler or another animal and they may become aggressive. Improper handling of animals can also cause discomfort, pain, and provoke the animal to bite or scratch.

  Animal bites and scratches that cause minor discomfort are sometimes disregarded by animal workers unfamiliar with the diseases that may result. Even minor bites and scratches can result in infection and illness when not properly treated.

  Animal handlers must practice proper animal handling techniques and wear appropriate PPE. In addition, animal handlers must have a current tetanus vaccination and report all potential exposure incidents to PI/LS.

Due to their strength, dexterity, intelligence, and tenacity of NHP, those working with or near NHP should take additional precautions. Personnel working with or near NHP cannot wear loose clothing. They must also tie their hair back, and use a face shield for additional protection of the eyes, nose, and mouth. Prior to working with NHP personnel must know the location of the Monkey Bite Kit in case of an exposure incident.

2.1.3.2 Sharps

To lessen the risk of accidental infection the use of sharps should be avoided when alternate methods are available. Sharps are instruments or equipment capable of causing a puncture or cut, including needles, scalpels, razor blades, glass Pasteur pipettes, slides, and broken glassware. For animal handlers, injury could occur when restraining an animal during a procedure involving the use of needles.

2.1.3.3 Animal Allergens

Animal allergens present another hazard to personnel who work with or around laboratory and research animals. The amount of animal allergens a person is exposed to is directly proportional to the number of animals. Animal allergy symptoms may include but not be limited to rash, itchy eyes, runny nose, congestion, or difficulty breathing. If you experience any of these symptoms, immediately leave the animal space and notify your supervisor and fill out a *First Report of Accident* form. The following sections outline animal allergens that may be encountered at George Mason University.

- **Rodents (Mice, Rats, Guinea Pigs, and Rabbits):** Allergens are present in rat urine, saliva, dander, and contaminated litter. Personnel may develop symptoms due to inhalation exposure to allergenic aerosols. Procedures which are likely to release higher concentrations of aerosolized allergens include cage cleaning and handling the animals for shaving, injection, or collecting blood or urine samples.

- **Other Animals (Birds, Fish, and Insects):** Allergens are present in bird serum and droppings that contain serum, as well as fish and insect proteins. Personnel may develop symptoms due to inhalation exposure to aerosols.

2.1.3.4 Zoonoses

Zoonotic diseases are diseases that can be transmitted between animals and humans and vice versa. The following sections describe zoonotic diseases which may be encountered when working with or near laboratory and research animals. Proper PPE and work practices, including good hygiene must be used at all times when handling animals. More information regarding zoonoses is provided in *Animal & Vivarium Safety* training.

2.1.3.5 Physical Hazards in Animal Spaces

Physical hazards that may be encountered when working with or near laboratory or research animals include ergonomic hazards, slips, trips, falls, heat stress, splashes and noise.

2.1.3.5.1 Ergonomic Hazards

Animal-handling personnel may encounter ergonomic hazards over the course of their work. They may need to lift heavy loads, including feed bags, animals, and cages. When possible, these hazards should be eliminated by the use of materials handling equipment. Personnel should be physically capable of lifting the load and instructed in proper lifting techniques. Employees should never attempt to manually lift more than 50 pounds alone. Always use two people when lifting heavy loads. Items that weigh more than 25 pounds should not be stored above or below hip-height. For more information, see the *Material Handling Safety Guide*.

In addition to lifting, repetitive motion tasks may also place ergonomic stress on an animal handler. Tasks that might contribute to repetitive motion stress include: adjusting control knobs, opening and closing cage doors, moving small animals from cage to cage, and mopping floors. Personnel should stretch every 15 minutes to reduce stress caused by repetitive motions.

2.1.3.5.2 Slips, Trips, and Falls

Due to the use of cage wash and sterilization equipment, floors may be wet in areas where personnel work with or near research or laboratory animals. Personnel should wear appropriate footwear to prevent slips and falls. In addition, spills must be cleaned up as soon as possible.

Be aware when working in food storage and dispensing areas as these areas may have slip and trip hazards where food has been spilled and not swept up. Clean up all spilled food as soon as possible to prevent slip hazards.

2.1.3.5.3 Electrical Hazards

Due to the potential for electrical hazards in wet areas, ground fault circuit interrupters must be present on electrical outlets in areas where conditions may be wet.

2.1.3.5.4 Heat Stress

Steam may be used for sterilization of laboratory materials or equipment and for cage washing. As a result, animal handlers may be exposed to hot environments at times due to the higher-than-average temperature and humidity, as well as PPE they are required to wear. EHS will conduct an assessment, including exposure monitoring and identification of applicable control measures, upon request. Contact EHS if you are concerned about working in areas with elevated temperatures.

2.1.3.5.5 Noise

Personnel working with or near animals may be exposed to loud noise due to vibration, exhaust fans, and animals. George Mason University's *Hearing Conservation Program* has been developed to protect employees from exposure to excessive noise and complies with OSHA standard 29 CFR 1910.95. EHS administers the *Hearing Conservation Program* and conducts noise monitoring to evaluate occupational noise exposure and determine inclusion in the program as well as to evaluate the noise output from a source.

2.2 Risk Management

Risk management involves the use of measures designed to reduce potential exposure of laboratory personnel, the community, and the environment to hazards present in the laboratory. A comprehensive risk management program includes engineering, administrative, and physical

controls that reduce the duration, frequency, and severity of exposure to laboratory hazards. Administrative controls include written safety procedures and practices, training, documentation, access restrictions, and proper signage and labeling. Engineering controls include facility features such as laboratory design, ventilation systems, storage areas, and safety equipment. Physical controls are provided by PPE and good laboratory practices. Controls applicable to research and instructional laboratories are discussed in the remaining sections of this manual.

2.2.1 Biosafety Levels

The biosafety levels established by the CDC and NIH are an important tool in determining risk management strategies in biological laboratories. These levels indicate the type of laboratory facilities and practices required based on the type of material being used, the laboratory techniques employed, the safety equipment available for use with the material, and the design and construction of the facility in which the material is being manipulated. Selection of an appropriate biosafety level for work with a particular material or animal study depends upon several factors:

- RG, biological stability, and endemicity of the material.
- Possible routes of exposure.
- Nature and function of the laboratory.
- Procedures and manipulations that will be performed.
- Safety equipment and engineering controls available in the laboratory.
- Availability of effective vaccines or therapeutic measures.

The BMBL outlines criteria for four biosafety levels (BSL-1 through -4) and four animal biosafety levels (ABSL-1 through -4) and provides safety guidelines for each. A summary of each of the biosafety levels is provided below.

- **Biosafety Level 1 (BSL-1)** is assigned to work involving well-characterized, nonbiohazardous agents not known to consistently cause disease in immunocompetent adults, and present minimal potential hazard to laboratory personnel and the environment. These agents are of minimal potential hazard to laboratory personnel and the environment. Examples of such agents include: *Bacillus subtilis*, *Naegleria gruberi*, infectious canine hepatitis virus, and organisms exempt under the NIH Guidelines.

- **Biosafety Level 2 (BSL-2)** is assigned to work with infectious agents and materials that cause disease in humans with a varying degree of severity and are a moderate hazard to laboratory personnel and the environment. Examples of such agents include: Hepatitis B virus (HBV), Human Immunodeficiency Virus (HIV), *Salmonella spp.*, and *Toxoplasma spp*. All human and NHP material must be handled at BSL-2.

- **Biosafety Level 3 (BSL-3)** is assigned to work with indigenous or exotic agents that may cause serious or potentially lethal disease as a result of exposure by inhalation. *Mycobacterium tuberculosis*, St. Louis encephalitis virus, and *Coxiella burnetti* are representative infectious agents assigned to BSL-3. Use of a biosafety cabinet or other physical containment device is required for all procedures involving the manipulation of

infectious material in BSL-3 laboratories and special engineering and design features are required.

- **Biosafety Level 4 (BSL-4)** is assigned to work with dangerous and exotic agents that pose a high individual risk of aerosol-transmitted laboratory infection and life threatening disease. *Variola spp*. (causative agent of smallpox) and viruses that cause hemorrhagic fever are representative infectious agents assigned to BSL-4. BSL-4 laboratories must be located in a controlled area that is completely isolated from all other areas of the building, preferably in a separate building, and special engineering and design features are required. Laboratory staff must have specific and thorough training in handling extremely hazardous infectious agents. Laboratory staff must understand the primary and secondary containment functions of standard and special practices, containment equipment, and laboratory design characteristics. There are no BSL-4 laboratories at George Mason University.

- **Animal Biosafety Level 1 (ABSL-1)** is assigned for animal work that does not involve biological agents or involves well-characterized agents that are not known to cause disease in immunocompetent humans, and that are of minimal potential hazard to laboratory personnel and the environment.

- **Animal Biosafety Level 2 (ABSL-2)** is assigned for animal work with those agents associated with human disease that pose moderate hazards to personnel and the environment. ABSL-2 builds on the practices, procedures, containment equipment, and facility requirements of ABSL-1.

- **Animal Biosafety Level 3 (ABSL-3)** is assigned to animal work involving indigenous or exotic agents that present the potential of aerosol transmission and of causing serious or potentially lethal disease. ABSL-3 builds on the practices, procedures, containment equipment, and facility requirements of ABSL-2.

- **Animal Biosafety Level 4 (ABSL-4)** is assigned to animal work involving dangerous or exotic agents that pose a high risk of life-threatening disease, aerosol transmission, or related agents with unknown risk of transmission. There are no ABSL-4 laboratories at George Mason University.

2.3 Samples of Unknown Hazard

When assessing the risk associated with samples or agents of unknown hazard, such as field or environmental samples, PI/LS should consider the following:

- Type of sample (e.g., air, water, soil, plant, animal).
- Location from which the sample was taken and any known biohazardous agents endemic to that location.
- Potential for the samples to contain biohazardous or infectious material.
- Risks inherent to the procedures that are to be performed (e.g., isolation of microbes, aerosol production, likelihood of percutaneous or mucous membrane exposure, other hazardous materials involved).

In devising management strategies for the risk posed by samples of unknown hazard, PI/LS are encouraged to be conservative. BSL-2 and BSL-3 procedures and practices are designed to protect laboratory personnel from infectious material and should be implemented when the risk for exposure to an infectious agent is possible. EHS is available to assist PI/LS in devising appropriate risk management strategies for samples or agents of unknown hazard.

3.0 Engineering Controls

Engineering controls are facility features and equipment intended to reduce the likelihood or severity of an exposure. Safety equipment is a primary barrier intended to remove or minimize exposure to hazardous biological materials. Biological safety cabinets (BSCs) are the primary device used to provide containment of infectious aerosols or splashes during manipulation. Safety equipment includes personal protective equipment such as gloves, lab coats, and protective eyewear. Facility design and construction are secondary barriers designed for protecting the laboratory worker, persons outside the laboratory, and the public. Facility design features may include specialized ventilation systems, airlocks, or controlled access zones. PI/LS should consult with Facilities Management and EHS to select, acquire, and install appropriate engineering controls prior to the commencement of the work that requires these controls. This includes (but is not limited to) ventilation, room pressure controls, biosafety cabinets, chemical fume hoods, and shielding devices. In biological laboratories, engineering controls are used primarily for containment.

3.1 Facility Design

The *Laboratory Safety Manual* provides general design requirements for all George Mason University laboratories. Additional design requirements for biological laboratories are provided below.

3.1.1 BSL-1 Facility Requirements

In addition to facility requirements listed in the *Laboratory Safety Manual*, BSL-1 laboratories must meet the following requirements:

- The entrance to the laboratory must have a closable door.
- The laboratory must be designed for easy cleaning (no carpets or rugs).
- A sink for hand washing must be available within the laboratory.
- Work surfaces and bench tops should be impervious to water and resistant to moderate heat, organic solvents, acids, alkalis, and decontamination chemicals.
- Laboratory windows that open to the exterior should be fitted with screens.
- The laboratory should be equipped with nonfabric laboratory furniture that can be easily cleaned and decontaminated.

3.1.2 BSL-2 Facility Requirements

In addition to the facility requirements listed for BSL-1 laboratories, BSL-2 laboratories must meet the following requirements:

- Doors to the laboratory must be lockable to restrict access.
- A method for decontamination of infectious wastes must be available.
- Vacuum lines should be protected with HEPA filters.
- BSCs should be located away from doors and heavily traveled areas.
- The laboratory must be equipped with a biosafety cabinet located away from doors and heavily traveled areas.
- An eyewash must be readily available.

- BSL-2 laboratories should be designed to have 10 to 12 air changes per hour.
- Laboratory windows should not be operable.

3.1.3 ABSL-1 Facility Requirements

In addition to the facility requirements listed for BSL-1 laboratories, ABSL-1 laboratories must meet the following requirements:

- Animal facilities should be separated from areas that are open to unrestricted personnel traffic.
- External facility doors should be self-closing and self-locking.
- Doors to animal rooms should open inward, be self-closing, and kept closed when experimental animals are present.
- The animal facility should be designed, constructed, and maintained to facilitate cleaning and housekeeping. The interior surfaces (walls, floors, and ceilings) should be water-resistant.
- Windows are not recommended. Any windows must be resistant to breakage. Where possible, windows should be sealed.
- If floor drains are provided, the traps should always be filled with an appropriate disinfectant.
- Ventilation should be provided in accordance with the *Guide for Care and Use of Laboratory Animals*, latest edition. No recirculation of exhaust air should occur. It is recommended that animal rooms maintain negative pressure compared to adjoining hallways.
- The facility should have a hand washing sink.
- Cages are washed manually or in a cage washer. The mechanical cage washer should have a final rinse temperature of at least 180°F.
- Illumination is adequate for all activities, avoiding reflections and glare that could impede vision.

3.1.4 ABSL-2 Facility Requirements

In addition to the facility requirements listed for BSL-2 and ABSL-1 laboratories, ABSL-2 laboratories must meet the following requirements:

- Access to the facility is limited by secure locked doors.
- Exhaust air is discharged to the outside without being recirculated to other rooms. Ventilation should be provided in accordance with criteria from *Guide for Care and Use of Laboratory Animals*, latest edition. The direction of airflow in the animal facility is inward; animal rooms should maintain negative pressure compared to adjoining hallways.
- An autoclave should be available in the animal facility to decontaminate infectious waste.
- A handwashing sink should be in the animal room where infected animals are housed, as well as elsewhere in the facility.

3.2 Safety Equipment

Safety equipment designed to reduce the risk of exposure to biohazardous material is a critical component of biosafety. Such equipment includes emergency shower and eyewash stations, biosafety cabinets, and filtered vacuum traps. Additional safety equipment may be required depending on the substances used and procedures performed. PI/LS are responsible for working

closely with EHS so that safety equipment is available to laboratory personnel, routinely inspected, and repaired or replaced as necessary.

3.2.1 Emergency Showers and Eyewash Stations

An American National Standards Institute (ANSI) approved emergency shower and eyewash station must be available within in a 10-second walk from each area where flammable or corrosive substances are used, be clearly labeled, and easily accessible. All laboratory personnel must know the location of the nearest shower and eyewash stations and must be trained in their use. If an emergency shower or eyewash station is not available, contact EHS.

Emergency showers are designed to provide immediate response to exposures that cover a significant part of the body. Eyewash stations must be capable of being activated with one hand and maintain appropriate flow without the need for additional control. Eyewash units must be capable of providing 0.4 gallons of water per minute at 30 psi for a minimum of 15 minutes.

Emergency showers and eyewash stations must be installed, maintained, flushed, and tested in accordance with the ANSI Standard for Emergency Eyewash and Emergency Shower Equipment (Z358.1-1990). Both emergency showers and eyewash stations must be flushed every two weeks to verify that they are operating properly and the effluent is clear. Routine flushing is managed by each department. EHS provides protocols for flushing emergency showers and eyewash stations and distributes supplies necessary for the procedure to each department. Contact the Safety Liaison from your department or EHS for more information. Showers and eyewash stations are tested by EHS annually to certify that water pressure and flow rate are within acceptable parameters.

3.2.2 First Aid and Spill Supplies

All laboratories and laboratory support rooms should be equipped with first aid supplies to assist laboratory personnel in responding to minor injuries and spill supplies relevant to the activities of the laboratory. These supplies should be clearly marked, easily accessible, and located near the laboratory exit. All laboratory personnel must know the location of these supplies. Supplies should be routinely inspected and replaced as necessary. EHS will provide and restock spill kits upon request.

3.2.3 Biosafety Cabinets

Biosafety cabinets are the primary means of containment developed for working safely with infectious materials. Biosafety cabinets are designed to provide personnel and environmental protection as well as protection for work materials when appropriate practices and procedures are followed. Three kinds of biosafety cabinets, designated Class I, Class II, and Class III, have been developed to meet varying research and clinical needs. Class II and III biosafety cabinets use HEPA filters in the exhaust and supply systems. All biosafety cabinets at George Mason University are Class II or Class III cabinets.

Biosafety cabinets should be situated in an area of the laboratory where there is adequate clearance on either side of the cabinet for maintenance, and sufficient clearance above the cabinet to provide accurate air velocity measurement across the exhaust filter and for filter changes. Biosafety cabinets must not be situated near doors or in high traffic areas. Biosafety

cabinets are certified annually in accordance with federal and state regulations. If a cabinet fails to meet certification requirements, discontinue use and contact EHS immediately. The CDC guidebook on *Primary Containment for Biohazards: Selection, Installation and Use of Biosafety Cabinets* is available on the EHS website.

Many biosafety cabinets are equipped with germicidal UV lamps. EHS discourages the use of these lamps because the effectiveness of these lamps in sterilizing the biosafety cabinet is affected by many factors including the length of exposure, the distance of the lamp from the surface of the biosafety cabinet, the intensity of the lamp, and items left on the surface of the cabinet that block the UV rays.

3.2.3.1 Class II Biosafety Cabinets

Class II biosafety cabinets are partial barrier systems that rely on laminar flow movement of air to provide containment. If the air curtain is disrupted (movement of materials in and out of the cabinet, rapid or sweeping movement of the arms), the potential for containment release into the laboratory work environment is increased as is the risk of contaminating materials.

Class II biosafety cabinets provide personal, environmental, and product protection. Airflow is drawn into the front grill of the cabinet, providing personnel protection. In addition, the downward laminar flow of HEPA filtered air provides product protection by minimizing the likelihood of cross contamination across the work surface of the cabinet. Air exhausted from the cabinet is HEPA filtered to provide environmental protection. This air may be recirculated into the laboratory (Type A cabinets) or discharged from the building through a hard connection (Type B cabinets).

HEPA filters are effective at trapping particulates and infectious agents, but do not capture volatile chemicals or gasses. Volatile chemicals should not be used in biosafety cabinets that recirculate air to the laboratory, as the buildup of chemical vapors in the cabinet (by recirculated air) and in the laboratory (from exhaust air) could create health and safety hazards. PI/LS must contact EHS if their work requires the use of volatile chemicals, carcinogens, or radioactive materials in the biosafety cabinet.

Below are general guidelines to follow when using a Class II biosafety cabinet:

- After turning on the blower, wait 5 minutes before beginning work in the cabinet.
- Wear appropriate PPE. Gloves should overlap the lab coat or cuffs.
- Adjust stool height so that your face is above the front opening. Armpits should be at sash height.
- Place all work material into the biosafety cabinet before beginning work to minimize movement in and out of the cabinet. Place material as far back in the cabinet as practical.
- Wipe all material and equipment with 70% alcohol prior to placing in the cabinet to reduce the introduction of contamination.
- Never obstruct air intake grill, as this is where air is drawn into the cabinet. Disinfect all interior surfaces before and after working in the cabinet with a 1% bleach solution or other approved disinfectant. In an effort to reduce the corrosive effects of the bleach solution, follow with a wipe down of 70% alcohol.

- Keep laboratory door closed and traffic around the cabinet at a minimum while working in the cabinet.
- Wait several minutes after adding or removing items from the biosafety cabinet in order to allow the airflow to stabilize.
- Conduct all work 6 inches inside the work area.
- Organize the work areas and procedures to minimize cross-contamination.
- Do not use open flames in the cabinet. When deemed absolutely necessary, touch-plate micro burners equipped with a pilot light to provide a flame on demand may be used. Small electric furnaces are available for decontaminating bacteriological loops and needles, and are preferable to an open flame inside the biosafety cabinet. Disposable or recyclable loops should be used whenever possible.

3.2.3.2 Class III Biosafety Cabinets

Class III cabinets are designed for work with highly infectious agents and provide maximum protection for the environment and personnel. Class III cabinets have a gas-tight enclosure with a non-opening view window. Both supply and exhaust air are HEPA filtered. Exhaust air must pass through two HEPA filters or a HEPA filter and an air incinerator before discharge to the outdoors. Airflow is maintained by a dedicated independent exhaust system which keeps the cabinet under negative pressure. Long, heavy-duty rubber gloves are attached in a gas-tight manner to ports in the cabinet and allow for manipulation of the materials inside. Laboratory personnel must receive laboratory-specific training for use of Class III cabinets and must follow laboratory-specific Standard Operating Procedures (SOP) for experiments to be performed in the cabinet.

3.2.4 Negative Pressure Cage Rack

Ventilated cages must be under negative pressure with respect to the surrounding area and exhaust is filtered through a high efficiency particulate air (HEPA) filter which is designed to remove 99.97% of particles with an aerodynamic diameter of 0.3 microns. Animal cages must have cage filter tops to prevent contamination of the work area as well as the inside of the cage due to aerosol generation.

3.2.5 Downdraft Necropsy Tables

Downdraft necropsy tables are designed to capture chemical vapors generated during necropsy. Exhaust fans alongside the table produce downdrafts which draw clean air across the work surface. The effectiveness of downdraft tables can become compromised due to turbulence in the room, the size of the animal, and work practices. Use of downdraft tables must be evaluated by EHS prior to use.

3.2.6 Vacuum Line Chemical Traps and Filters

Chemical traps and filters prevent suction of material into the vacuum lines. A hydrophobic HEPA filter trap must be installed on all vacuum lines within the biosafety cabinet to keep infectious materials out of the lines and to prevent sample contamination.

Chemical traps should be used to decontaminate waste from cell culture or from work with live virus. For the latter, a chemical disinfectant trap within the biosafety cabinet is required. Figure 1 depicts a common setup of a chemical trap and filter system.

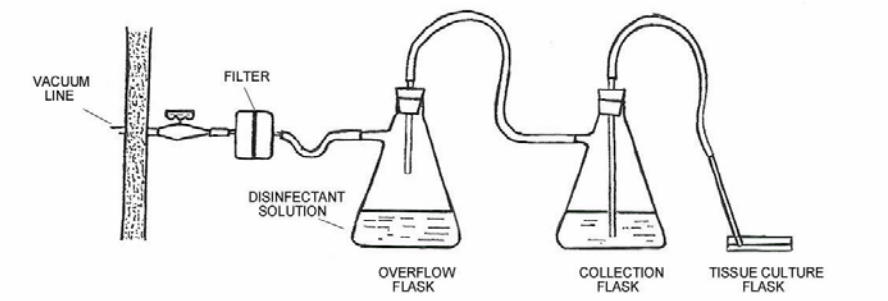

Figure 1. Design of Chemical Trap for Tissue Culture Procedures
[Source: NIH (1979) *Laboratory Safety Monograph- A Supplement to the NIH Guidelines for Recombinant DNA Research].*

When using a vacuum line chemical trap:

- Prior to the collection of the liquid BSL-2 waste, add 10% bleach to the vacuum flask to equal 10% of the maximum collection volume so that, when collection is complete, the final solution will contain 10% bleach. Maximum collection volume should be no more than two-thirds full.
- Vacuum line filters should be examined before each use and replaced if clogged or if liquid makes contact with the filter.
- Used filters must be discarded as biohazardous waste.

3.3 Laboratory Equipment and Material

PI/LS are responsible for maintaining laboratory equipment and providing training to laboratory personnel on the correct use of equipment. A routine inspection and maintenance program that includes necessary instrument calibration, certification, and maintenance procedures should be implemented for all equipment in the laboratory to identify worn parts, frayed wires, malfunctioning instruments, faulty safe guards, and other potential hazards. The *Laboratory Safety Manual* provides a general equipment safety checklist.

The use of laboratory equipment with biohazardous materials requires special attention to procedures which prevent contamination and the release of aerosols. All PI/LS must properly train all laboratory personnel on the safe use of these types of equipment. Additionally, all individuals who have access to areas where these items are used must be trained or made aware of any associated hazards.

3.3.1 Safety Procedures for Centrifugation of Biohazardous Materials

Aerosol production and leakage are the predominant hazards associated with the centrifugation of biohazardous materials. The following procedures should be employed to minimize these hazards.

- Check tubes, bottles, and rotors for cracks or deformities prior to use.
- Examine O-rings and replace if worn, cracked, or missing.

- Fill and decant all centrifuge tubes and bottles in the biosafety cabinet.
- Never overfill centrifuge tubes. Limit the volume of each centrifuge tube to 75% of the total volume of the tube.
- Wipe outside of tubes with disinfectant before placing in safety cups or rotors.
- Cap tubes before centrifugation.
- Balance buckets, tubes, and rotors properly.
- Always use sealed safety cups, safety buckets, or sealed rotors with O-rings as secondary containment.
- Check that the rotor is seated correctly.
- When using swinging bucket rotors, make sure that all buckets are hooked correctly and move freely.
- Do not exceed safe rotor speed.
- Close and secure the lid before starting the centrifuge.
- Do not leave the centrifuge unattended until full operating speed is achieved.
- Stop the centrifuge immediately if unusual noises or vibration occur.
- Never open a centrifuge until the rotor has stopped and you have allowed potential aerosols to settle.
- Wear appropriate PPE.
- Check inside the centrifuge for possible spills and leaks. Decontaminate the centrifuge and rotor thoroughly according to procedures.
- After the run, allow aerosols to settle before opening the centrifuge.
- Open safety buckets or rotors in a biosafety cabinet.
- Decontaminate safety buckets, rotors and centrifuge after each use.
- If a centrifuge malfunctions or a tube breaks, turn the centrifuge off immediately and, when feasible, unplug it. Allow aerosols to settle before opening the lid of the centrifuge.

3.3.2 Safety Procedures for Culture Plates, Tubes, and Bottles

While the risk of occupational exposure from opening plates, tubes, or bottles containing microbial cultures is relatively low, some laboratory-associated infections have been acquired in this manner. Therefore, the following precautions should be taken when conducting experiments involving culture plates, tubes, and bottles.

- Inoculating cultures:
  - When possible, use pre-sterilized disposable inoculating loops and needles.
  - If wire inoculating loops or needles are used, use electric or gas incinerators specifically designed to sterilize inoculating instruments instead of open flame sterilization.
  - Allow wire inoculating loops and needles to cool completely to prevent splatter and the release of aerosols.
- Opening and manipulating cultures:
  - Cultures, particularly those of spore-forming organisms (e.g., fungi, *Bacillus*, ssp.), should be manipulated in a biosafety cabinet.
  - To assure a homogenous suspension within a liquid culture, the culture should be gently swirled. Vigorous shaking may create heavy aerosol.
  - To reduce exposure to any aerosols produced, wait several minutes after resuspension before opening the liquid culture.

- Slowly open screw-capped bottles or tubes to prevent potential exposure to aerosol. Aerosols are created when a film forms between the rim of the bottle or tube and the liner of the cap.
- Open containers of lyophilized infectious material in a biosafety cabinet.
- Use vented plastic Petri dishes and properly dry dishes before inoculation to reduce the formation of a film between the rim and lid of inverted plates. This film is usually contaminated with microorganisms. Aerosols can be released when the lid is removed and the film is disrupted. Filter papers fitted into the lids reduce, but do not prevent dispersal. If plates are obviously wet, they should be opened in the biosafety cabinet.

3.3.3 Safety Procedures for Use of Microtome or Cryostat

Due to the extremely sharp blade and the nature of the material used with a microtome or cryostat, training is essential. It is the responsibility of PI/LS to train personnel in the proper use of these instruments. The following safety guidelines should be followed when using a microtome/cryostat.

- Treat all tissues, including frozen and fixed tissues, as potentially infectious.
- Wear appropriate PPE, such as a lab coat, face shield, safety glasses or goggles, surgical grade Kevlar gloves that provide dexterity and cut protection, examination gloves to protect against biohazards, and respirator (when required).
- Use engineering controls such as forceps, dissecting probes, or small brushes for manipulating samples or cleaning the instrument, while the blade is in place, or changing the blade.
- Use extreme caution when aligning blocks. Make sure the block holder is in locked position when loading/aligning blocks.
- Keep blocks wet to minimize airborne shavings during slicing.
- Dislodge stuck blocks using mechanical means such as forceps and/or dissecting probes.
- Use blade edge protectors/guards to cover blade edges that extend beyond the blade holder.
- When not in use, store blades in appropriate storage containers. Do not leave blades in the instrument.
- Do not move or transport the instrument with the blade in position.
- Avoid the use of freezing propellants with biohazardous materials as they may produce aerosols.
- Consider trimmings and sections of tissue to be contaminated and discard in the appropriate waste stream.
- Decontaminate equipment with an appropriate disinfectant after each use. When working with tissues that may contain agents that are resistant to disinfection (e.g., *M. tuberculosis*, prions, bacterial spores), additional decontamination procedures will be required. Contact EHS for assistance.

3.3.4 Safety Procedures for Mechanical Disruption of Biohazardous Material

The use of homogenizers, ultrasonic disrupters, grinders, or other equipment used for mechanical disruption can result in considerable aerosol production. The following safety procedures should be employed when using a mortar and pestle, ball mill, colloid mill, jet mill, homogenizer (tissue grinder), magnetic mixer, stirrer, sonic cleaning device, ultrasonic cell disintegrator (sonicator), shaker, vortexer, or other means of mechanical disruption with biohazardous material.

- Perform operations in a biosafety cabinet, unless the equipment has a special containment apparatus.
- Before use, inspect equipment to verify that it is functioning properly.
- During each use, place a towel or other splash-absorbing material wetted with disinfectant under the work area.
- When possible, use homogenizers and grinders designed to prevent the production and release of aerosols.
- After use, disinfect the equipment with the appropriate disinfectant.

3.3.5 Safety Procedures for Using Safety Blenders with Biohazardous Material

Safety blenders are designed to prevent leakage from the bottom of the blender bowl, provide a cooling jacket to avoid biological inactivation, and to withstand sterilization by autoclaving. Guidelines for the use of safety blenders are provided below.

- Use safety blenders designed to prevent leakage from the rotor bearing at the bottom of the bowl. In the absence of a leak-proof rotor, inspect the rotor for leakage prior to operation. A preliminary test run with sterile water, saline, or methylene blue solution is recommended prior to use.
- If the blender is used with infectious material, place a towel moistened with an appropriate disinfectant over the top of the blender and use the blender in a biosafety cabinet.
- Use unbreakable jars with the blender. Avoid using glass jars.
- Allow aerosols to settle before opening the safety blender bowl.
- Sterilize the device and residual contents promptly after use.

3.3.6 Safety Procedures for Lyophilizing Biohazardous Material

Depending on lyophilizer design, aerosol production may occur when material is loaded or removed from the lyophilizer unit. The following guidelines should be used when lyophilizing biohazardous material.

- Use polypropylene tubes instead of glass when possible.
- Load and unload sample material in a biosafety cabinet.
- Use a filter on the vacuum pump exhaust to prevent the release of biohazardous material.
- After lyophilization is complete, disinfect all surfaces of the unit.

3.3.7 Safety Considerations for Use of Miscellaneous Equipment with Biohazardous Material

- **Water baths:** Water baths should contain disinfectant. For cold water baths, 70% propylene glycol is recommended. Sodium azide, which poses an explosion hazard, should not be used.
- **Freezers and refrigerators:** Deep freeze, liquid nitrogen, dry ice chests, and refrigerators should be cleaned periodically, defrosted, and decontaminated.
- **Shakers and shaking incubators:** Shakers and shaking incubators should be examined carefully for potential breakage of flasks, test tubes, or bottles. Screw-capped durable plastic or heavy walled glass flasks should be used. These should be securely fastened to the shaker

platform. Fragile glass containers may be enclosed in a plastic bag with an absorbent material prior to placement in the shaker to contain spills of biohazardous material. Shakers and incubators should be routinely cleaned and decontaminated.

3.4 Housekeeping

Poor housekeeping leads to cluttered work areas which are difficult to decontaminate, increases the potential for accidents and physical injury, and increases the likelihood of experimental error and contamination.

Well-defined housekeeping procedures are an essential component of biosafety. A well-conceived and well-executed housekeeping program provides a clean, uncluttered work area that is free from physical hazards, contains all instruments and safety equipment necessary to perform experiments safely, and is easily decontaminated.

4.0 Administrative Controls

Administrative controls are precautionary measures implemented to reduce the risk of accidents in the laboratory through training, signage and labeling, record keeping, and medical surveillance. Administrative controls should be established prior to beginning a laboratory project or protocol.

4.1 Training

George Mason University laboratory personnel, students, support services staff, and visitors entering laboratories or laboratory support rooms are required to receive safety training commensurate with their level of participation in laboratory activities and the duties they are to perform. EHS provides training in general laboratory safety awareness, chemical safety, biosafety, radiation safety, and laser safety in accordance with relevant guidance and regulatory requirements. Personnel can view a complete list of training offered by EHS, and register for training through the EHS website (ehs.gmu.edu).

4.1.1 Biological Safety for BSL-2 Laboratories

All individuals working in or frequenting laboratories where infectious materials are used or stored must receive *Biological Safety for BSL-2 Laboratories* training before beginning work with infectious materials. This training reviews the principles of biosafety including risk assessment and management strategies, risk groups and biosafety levels safe laboratory practices, methods of disinfection and decontamination, waste management, and spill and exposure response. *Biological Safety for BSL-2 Laboratories* must be renewed annually and can be renewed by attending *BSL-2 Biosafety Refresher* within 12 months.

4.1.2 Bloodborne Pathogens

In accordance with the OSHA Bloodborne Pathogen Standard, training on risks associated with bloodborne pathogens, safe laboratory practices, medical waste management, and emergency procedures is provided annually to all individuals at occupational risk for exposure to bloodborne pathogens. Individuals working in or frequenting laboratories or clinical settings where bloodborne pathogens or other potentially infectious materials are present must receive bloodborne pathogens training before beginning work. Training must be renewed annually.

4.1.3 Autoclave Equipment

Training in the proper use of autoclaves is required for all personnel who use autoclaves to sterilize equipment or waste products. This training is provided as part of *Biological Safety for BSL-2 Laboratories* training. However, it is also provided separately upon request**.**

4.1.4 Animal & Vivarium Safety

In accordance with the NIH's Office of Laboratory Animal Welfare, OSHA, and National Research Council, training on risks associated with handling animals is provided monthly for individuals who work with or in the vicinity of laboratory animals. This training reviews the risks associated with handling animals in the research laboratory environment, vivarium facility safety procedures, zoonotic diseases, personal protective equipment, animal waste handling & disposal, and laboratory animal allergens. This training must be renewed annually. Once you

have taken this training course, you may substitute *Animal & Vivarium Safety Refresher* training to meet the annual requirement.

4.1.5 Robinson Ventilation

Due to the complexities of the air handling system and the mixed use of Robinson Hall, EHS has established this training session for those responsible for laboratory courses in Robinson Hall. This training includes means by which to optimize the air handling system and to verify that it is activated, and storage requirements and waste handling procedures for dissection. This training includes *Chemical Safety* training and therefore participants receive course credit for both Robinson Ventilation and *Chemical Safety* training when they attend this session. Training must be renewed annually.

4.1.6 BSL-3 Training

Working in a BSL-3 environment requires specialized training. The BRL Training Plan outlines specific training requirements for personnel who work in the containment suite of the BRL or provide support for the containment suite. The training outlined in this plan is provided in addition to other university training requirements and is not meant to replace or substitute for any other university training requirements.

4.1.7 Shipping Class 6.2 Dangerous Goods and Dry Ice Training

Comprehensive training is required for personnel who ship infectious materials, diagnostic specimens, and dry ice. Instructional or research personnel who plan to ship infectious materials or diagnostic specimens must complete this training. Training must be renewed every two years. Contact EHS for more information.

4.1.8 Laboratory Specific Training

In addition to training provided by EHS, laboratory personnel must receive training specific to the laboratory in which they work. This training, provided by the PI/LS, should be based on the *Supplemental Laboratory Safety Plan*, and must include such topics as risks associated with the hazardous substances and physical hazards present in the laboratory, proper use of instruments and safety equipment, laboratory procedures and protocols, as well as procedures specific to animal handling. To verify receipt of this training, laboratory personnel must sign a *Laboratory Training Signature Page* provided in the *Safety Records and Resources* binder. A blank copy of the *Laboratory Training Signature Page* is available on the EHS website.

4.2 Prior Approval

EHS requires that *Project Review Forms* be submitted for all projects involving biological materials before the project start so that the project can be reviewed by the relevant safety committees. The IBC reviews projects involving biohazardous materials or potentially biohazardous materials and requires that projects are conducted in a manner that promotes laboratory health and safety and is in accordance with pertinent regulations and policies. The IBC identifies hazards and assesses the risks associated with the project or activity; evaluates the adequacy of safety procedures, facilities, and equipment; and determines the need for immunizations or other preventive medical measures. As necessary, projects will also be reviewed by the RSC, IACUC, or IRB. Projects cannot begin until IBC approval has been obtained.

In addition to IBC approval, the use of select agents and toxins must be registered with the CDC and/or APHIS. PI/LS considering work with select agents and toxins must first contact EHS for the approvals, permits, clearances and other necessary paperwork. Be aware that government clearance can take as much as 8 months to complete in advance of any project.

To initiate the review process, PI/LS should submit a completed *Project Review Form* to EHS. The IBC will inform PI/LS when their project has been approved. The *Project Review Form* is available on the EHS website.

Approval for all projects expires 36 months after the date of approval. EHS will notify PI/LS regarding project renewal. *Project Review Forms* should be reviewed by PI/LS annually and be updated as frequently as necessary to reflect changes in experimental protocols, the type or amount of biological material used, changes in personnel, or changes in locations where the work will be performed.

4.3 Laboratory Security and Access

Laboratories contain hazardous substances that can pose a serious danger to public health if handled by untrained personnel or removed from the laboratory. In addition, laboratories contain expensive instruments and equipment that must be protected from unauthorized use, vandalism, and theft. Therefore, it is imperative that PI/LS implement appropriate security precautions to prevent unauthorized individuals from gaining access to laboratory materials and equipment. To secure the laboratory, PI/LS must:

- Identify potential security risks in the laboratory (e.g., laboratory doors left open, doors left unlocked when the laboratory is unattended, or unsecured hazardous substance storage areas).
- Develop and implement laboratory security procedures to prevent unauthorized entry to the laboratory and access to hazardous substances.
- Develop and implement laboratory access restrictions to protect the health and safety of individuals entering the laboratory. Laboratory hazards and access restrictions for all laboratories should be clearly indicated at the entrance to the laboratory.
- Train laboratory personnel to implement security procedures.

The following security procedures must be followed in all laboratories:

- Keep doors closed at all times and locked when no authorized personnel are present.
- Do not leave hazardous substances unattended or unsecured at any time.
- Restrict access to freezers, refrigerators, storage cabinets, and other equipment where hazardous substances are stored.
- Limit laboratory access to approved laboratory personnel who are properly trained with regard to the hazards present in the laboratory and the type of work they will perform.
- Restrict off-hours access to individuals authorized by the PI/LS.
- Escort visitors to and from the laboratory.
- Challenge or question unfamiliar or suspicious individuals that gain access to restricted areas or to the laboratory. Report these incidents to University Police.

- Report any missing inventory to University Police.
- Report all acts of vandalism, theft, or suspicious activities to University Police.

4.3.1 Restricted Access for Visitors

Visitors should not be allowed to enter laboratories unattended and should be escorted to and from the laboratory by George Mason University personnel. *Laboratory Safety Awareness Training* and an *Assumption of Risk* form are required for visitors to enter a laboratory. Assumption of Risk forms must be kept on file by the PI/LS and forwarded to EHS.

4.3.2 Restricted Access for Minors

There may be occurrences where experiential learning and the laboratory environment will intersect. PI/LS may be asked to facilitate this type of experience in his or her laboratory with an individual under the age of 18 who is not enrolled in courses at George Mason University (herein referred to as *a minor*). Due to potential risks associated with the laboratory environment, access to all university laboratories is restricted for minors.

EHS has developed the *Minors in the Laboratory Guid*e (available on the EHS website) to assist PI/LS with the process of reviewing, approving, and assessing risk for minors who would like to participate in laboratory activities. This document also provides guidance for PI/LS on activities that involve minors such as: laboratory tours, required safety training, documentation of risk assumption by the minor's parent or legal guardian, as well as medical clearance requirements and forms.

General guidelines are listed below:

- Individuals under 12 years of age are not permitted in laboratories.
- Individuals between the ages of 12 and 18 may tour laboratories as part of chaperoned tour groups**.**
  - Two chaperones are required per tour group.
  - A tour group will consist of no more than 12 visitors (entering the laboratory at any given time).
- Visitors that tour laboratories must be approved by the PI/LS and must be given basic laboratory safety instruction by the PI/LS (or their designee) before entering laboratories.
- As a general practice, individuals between the ages of 12 and 15 are not permitted to perform experiments in the laboratory.
- EHS will perform a detailed risk assessment, in collaboration with the PI/LS, for proposed work involving minors ages 16 to 18. In these situations, the following will be required:
  - The minor and parent(s)/legal guardian(s) must complete the *Acknowledgement of Laboratory Risk for Minors.*
  - The minor must submit a recommendation from their school's science teacher.
  - The minor must submit the *Laboratory Volunteer Form*.
  - The minor must complete all required safety training and appropriate medical requirements (e.g., vaccinations, exams), as determined by EHS, prior to beginning volunteer work in the laboratory.

- Minors that have been approved to perform laboratory work must be supervised at all times in the laboratory by the PI/LS (or their designee). The PI/LS is responsible for laboratory specific training applicable to the work being performed by the minor.
- Minors may not perform work involving the following:
  - Cutting/dissecting of unfixed human samples or the processing of whole blood.
  - Acutely toxic chemicals or substances listed as Group 1, Group 2A, or Group 2B substances by the International Agency for Research on Cancer.
  - Sources of ionizing radiation.

All documentation must be sent to the Laboratory Safety group within EHS for review and risk assessment in conjunction with the PI/LS. All forms are available on the EHS website (ehs.gmu.edu). For more information or specific assistance regarding minors working in the laboratory, contact the Laboratory Safety group within EHS (labsafe@gmu.edu).

4.3.3 Laboratory Access for Support Services Staff

All support services staff (e.g., housekeeping, facilities management, police, etc.) must receive *Laboratory Safety Awareness Training* prior to entering laboratories or laboratory support rooms. Once trained, support services staff may enter laboratories and laboratory support rooms with the exception of specific rooms designated as restricted areas.

Restricted areas include laboratories that house animals, hazardous waste storage rooms, BSL-3 laboratories, and other areas with unique hazards. These laboratories are labeled with a restricted access symbol (Figure 2). Support services staff are not permitted to enter restricted areas unless requested by PI/LS or EHS. In this situation, the person requesting the service must submit a work order and arrange for access. EHS maintains a current list of restricted areas. For BSL-3 laboratories, staff must be escorted by EHS personnel.

Figure 2. Restricted Access Symbol.

For nonrestricted laboratories and laboratory support rooms, support services staff must notify the unit of nonroutine services (e.g., mopping and waxing floors, light bulb replacement, and equipment inventory) at least 10 university working days in advance of when the work is to occur. PI/LS may request that services be scheduled at a time that does not interfere with ongoing laboratory operations or critical experiments.

Support services staff has been instructed that they need not perform any services which make them uncomfortable or of which they are unsure. Specifically, housekeeping staff has been instructed that they should not clean pools of liquid off laboratory floors.

4.4 Biosecurity

The term *biosecurity* refers to protection of biological materials from loss, theft, diversion, or intentional misuse. The objective of a biosecurity program is to develop and implement practices and procedures that prevent the loss, theft, or misuse of microorganisms, biohazardous materials, and research related information. Biosecurity includes:

- Physical security designed to prevent unauthorized removal of materials.
- Material accountability procedures established to track the inventory, storage, use, transfer, and destruction of biohazardous material.
- Information security policies for handling sensitive information such as security plans, access codes, inventories, and storage locations.
- Material transport policies that include accountability measures for the movement of materials within the university.
- Accident, injury, and incident response plans.
- Reporting and communication procedures.
- Training and, in some cases (e.g., BSL-3 laboratories), practice drills.
- Personnel and visitor identification and screening policies.
- Routine security updates and evaluations.

Biosecurity is effectively achieved through the implementation of access controls and training requirements for BSL-1 and BSL-2 laboratories outlined in this manual and the BMBL.

4.5 Signs and Labels

Signs and labels are used to clearly identify specific laboratory hazards, safety equipment, emergency supplies, critical information, and designated areas within the laboratory. The following signage requirements apply to all laboratories and laboratory support rooms at George Mason University.

4.5.1 Laboratory Entryway Signs

The entrance to all laboratories and laboratory support rooms must be posted with signs that indicate the hazards present in the laboratory, the National Fire Protection Association rating of the laboratory, appropriate PPE to be worn, access restrictions, and contact information to be used in the event of an emergency. All laboratories and laboratory support rooms classified as BSL-2 or BSL-3 are required to have the universal biohazard symbol (Figure 3) posted at all entrances to the laboratory along with the word “biohazard” and the term “BSL-2” or “BSL-3.” In animal laboratories, entryway signs must be designated with the appropriate animal biosafety level. Special provisions for entry such as vaccination requirements must be posted as well. PI/LS are responsible for notifying EHS when entryway signs to their laboratory need to be updated.

Figure 3. Universal Biohazard Symbol

4.5.2 Labeling Requirements

All laboratory equipment (e.g., refrigerators, freezers, centrifuges, and incubators), biohazardous waste containers, and shipping or transport containers in which biohazardous material is used, stored, or disposed of must be labeled with the universal biohazard symbol (Figure 3) and the word "biohazard." Labels should be affixed to the container or as close as possible to the container using string, wire, adhesive, or any other method that prevents their loss or unintentional removal.

Equipment that has been used to store or manipulate biohazardous material must be decontaminated and labeled with a *Decontamination Certificate* before being sent out for repair or disposal. The method of decontamination employed will depend on the design of the equipment and the type of biological materials used. Equipment components that cannot be completely decontaminated must be clearly indicated with a label. A *Decontamination Certificate* is available on the EHS website.

4.6 Safety Records

PI/LS are required to maintain records regarding laboratory safety and compliance. Records should be kept in a central location where they are available to laboratory personnel and inspectors. The *Laboratory Safety Manual* provides a comprehensive list of laboratory records that PI/LS are required to maintain in the laboratory. These records are to be kept in the *Safety and Compliance* bins available in each laboratory and must be available to laboratory personnel, emergency responders, and inspectors. Following is a list of records required for biological laboratories.

4.6.1 Biological Safety Training Records

Training certificates must be kept in the *Safety Records and Resources* binder for all laboratory personnel who have completed training offered by EHS. *Laboratory Training Signature Page*s verifying that laboratory personnel have received specific laboratory training must also be kept in the *Safety Records and Resources* binder.

4.6.2 Biological Material Safety Data Sheets (MSDS)

MSDS have been developed for a number of infectious agents. These MSDS provide laboratory personnel the information necessary to safely manage biohazardous material, identify potential hazards, and effectively contain or clean up a biological spill or release. PI/LS are required to have copies of available MSDS for the infectious agents used or stored in their laboratory. A link to infectious agent MSDS can be found on the EHS website.

4.6.3 Biological Inventory

Each research and instructional laboratory is required to maintain a biological inventory that includes all of the biological materials stored in the laboratory. Biological inventories are maintained through the EHS Assistant database. PI/LS, or their designee, are responsible for updating the database and maintaining current inventory records. To access the EHS Assistant, click on the *Member* Login link on the EHS website (http://ehs.gmu.edu/). Use your MESA login and password. An updated Biological Inventory Report should be kept in the *Safety Records and Resources* binder.

4.6.4 Select Agent and Toxin Log

A *Select Agent and Toxin* log must be used to track inventory, usage, and destruction of each select agent and toxin (including toxins that are below the quantity threshold) in the laboratory.

4.7 Inspections

George Mason University is periodically inspected by federal, state, and local agencies. These regulatory agencies may visit George Mason University at any time, with or without prior notification, to assess safety and compliance at the university. During these visits, inspectors may ask to examine laboratories and laboratory support rooms, question laboratory personnel, and examine laboratory records.

EHS routinely inspects laboratories and laboratory support rooms. Inspections are performed in accordance with government regulations and funding agency stipulations, and are used to address safety issues identified in the laboratory. The IACUC conducts facility inspections twice a year to evaluate the effectiveness of control measures in place to reduce the risk of injury and illness of personnel working with or near laboratory or research animals. EHS and IACUC inspections also serve to prepare laboratories for inspections from outside agencies.

Information about the inspection process is available in the *Laboratory Safety Manual*. A copy of the *Inspection Checklist* is available on the EHS website. In laboratories and laboratory support spaces where biological material is present, inspections will include examination of:

- Signage and labeling.
- Safety records.
- Facility design.
- Emergency equipment (use and condition) and procedures.
- Safety equipment.
- Proper waste handling.
- Storage of biological material.
- Animal Bedding and food is stored off the floor.

4.8 Medical Surveillance Program

George Mason University is required by 29 CFR and Virginia Administrative Code (16VAC 25-90) to ensure that employees exposed to health hazards at work are included in a medical surveillance program. Medical surveillance is a series of medical services provided by a PLHCP for the primary prevention of occupational injuries and illnesses, including a review of

occupational and medical history, physical exams, diagnostic and performance testing, and vaccinations. George Mason University's *Medical Surveillance Plan* complies with applicable regulations and guidelines and establishes minimum medical surveillance requirements to prevent occupational injuries and illnesses for George Mason University employees whose job duties place them at risk to occupational hazards. The full program is available on the EHS website (ehs.gmu.edu).

Among the services offered through EHS are medical screening and health assessments, immunizations, consultation regarding health risks, and exposure incident monitoring. These services are provided at no cost to employees. In addition, an employee has the right to seek medical care pursuant to 29 CFR 1450(g), without the loss of pay at a reasonable time and place, should any of the following occur:

- Personnel experiences signs or symptoms associated with exposure.
- A spill, leak, explosion or other occurrence resulting in the likelihood of an exposure.
- Exposure monitoring reveals an exposure level above the permissible exposure limit.
- Routine handling (e.g., three times a week use of chemicals with high chronic toxicity).

Laboratory personnel who are not George Mason University employees (e.g., unpaid visiting faculty, volunteers, students) may not be covered under the *Medical Surveillance Plan*. These individuals should discuss the nature of their laboratory work with their healthcare provider and should have personal health coverage.

EHS maintains medical surveillance records (i.e., employee name, employee G number, PLHCP written opinion) for 30 years after termination of employment.

4.8.1 Animal Handlers

Participation in the Medical Surveillance Program is required for laboratory personnel, animal caretakers, technicians, veterinarians, facility maintenance engineers, housekeepers, and security personnel who have direct or indirect contact with animals or animal facilities. These personnel are required to complete the *Animal Handler Questionnaire*. The degree of participation in the Medical Surveillance Program for animal handlers will be based on hazard identification and risk assessment via the *Animal Handler Questionnaire* and may include:

- Medical evaluation for respirator use.
- Audiometric testing for employees exposed to occupational noise in excess of the OSHA action level for noise, 85 A-weighting decibels (dBA) with a 5 dBA doubling rate.
- Tetanus booster (required every 10 years).
- Measles, Mumps, Rubella booster, for personnel working with NHP.
- Hepatitis B vaccination (or TWINRIX, a combination of Hepatitis A and B) for personnel working with human blood or other potentially infectious materials.
- PPD test every six months for personnel working with NHP.
- Rabies vaccine or booster or positive titer within two years, as necessary.
- Additional vaccinations may be required for personnel working with specific infectious agents as part of their research.
- Additional diagnostic testing deemed necessary by the PLHCP.

- Postexposure incident evaluation (bites, scratches, sharps sticks, mucous membrane exposure, animal-related injuries, physical or mechanical injuries, allergy symptoms).

Other factors that will be considered to determine an employee's fitness for duty include the amount of animal contact, exposure intensity, exposure frequency, physical and biological hazards presented by animal, hazardous properties of agents used in research, and the susceptibility of the employee. Physicals for all animal handlers will be conducted upon initial assignment and repeated every three years.

4.8.2 Exposure Incidents

In the event of a personal exposure, an individual's primary concern must be to minimize the degree of exposure and the possible effects. The emergency procedures employed depend on the type of hazardous substance to which the individual was exposed and the extent of exposure. Refer to Section 12.3 Personal Exposure for accident procedures. Medical care as a result of work-related exposure may be provided at no cost to the employee and is dependent on the type of exposure.

4.8.3 Immunocompromised Individuals

Individuals with a compromised immune system or other condition that increases their susceptibility to infection must take extra precautions when working in a laboratory where infectious agents and opportunistic pathogens are present. It is recommended that immunocompromised individuals consult EHS to discuss the risks associated with laboratory work and the precautions that may be taken to avoid exposure.

4.8.4 Women of Childbearing Age

Some workplace hazards can affect a woman's reproductive health, her ability to become pregnant, or the health of her unborn child. Exposure to the following agents may be harmful to the embryo and fetus: HBV, Hepatitis E virus, HIV, Human parvovirus B19, Rubella (German measles), Lymphocytic Choriomeningitis virus, *Toxoplasma gondii*, *Listeria monocytogenes,* Varicella-zoster virus (chicken pox) and Cytomegalo-virus. PI/LS actively working with reproductive hazards are required to discuss the risks associated with reproductive hazards to all personnel at time of hire. Individuals who are pregnant or may become pregnant are urged to consult their physician and EHS to discuss the risks associated with certain biological materials and recommended precautions for work in the laboratory. EHS is available to advise personnel regarding pregnancy in the workplace and may, at the request of an individual, serve as a liaison between pregnant personnel and their respective supervisors. When requested or deemed necessary, EHS will review work procedures in the laboratory to minimize the potential for exposure. Individuals may be reassigned to tasks that do not involve exposure to reproductive hazards.

5.0 Personal Protective Equipment (PPE)

PPE must be provided to and worn by all laboratory personnel, students, and visitors, when entering a laboratory including spaces where research animals are present. The extent and type of PPE selected for a particular activity depends on the risks associated with laboratory operations to be performed. At a minimum, a lab coat, gloves, clothing that covers the legs, and closed–toe shoes must be worn when working with biological materials. Shoe covers, forearm protection, eye protection, or a respirator may be required depending on the type of work being conducted.

Specifically for areas where research animals are present, shoe covers or the use of sticky mats is also required. Personnel are also required to wear safety glasses, disposable coveralls, hair cover, and an N95 respirator when changing bedding in animal cages. In addition, personnel who have allergies to animals may also be required to wear an N95 respirator. Coveralls and N95 respirators are recommended for all personnel working with or near laboratory or research animals.

While PPE is an important component of any biological safety program, it is not a replacement engineering controls, administrative controls, good work practices, and safety equipment. PPE is most effective when used in conjunction with these controls. OSHA requires the use of PPE to reduce employee exposure to hazards when engineering and administrative controls are not feasible or effective in reducing these exposures to acceptable levels.

5.1 Personal Clothing

Personal attire must be considered when working in a laboratory, as clothing, accessories, and hair may become entangled in equipment, accidentally spill substances, or pass through flames unintentionally. Proper personal attire includes clothing that provides adequate coverage for the legs and closed toe footwear that provides adequate support and has suitable traction for laboratory activities. Hair should be confined or tied back. The following may not be worn in the laboratory: loose sleeves, dangling jewelry, clothing that leaves the legs exposed, or shoes with heels greater than one inch.

5.2 Eye Protection

Eye protection must be worn when working with substances or equipment that present a hazard to the eye such as when changing bedding in animal cages, handling hazardous chemicals or radioactive materials, and when working with biological materials outside of a biological safety cabinet. Eye protection must meet design requirements set forth by ANSI (Z87.1-1998) and must be appropriate for the activity being performed.

Safety glasses should fit securely and be free of smudges or scratches that may obstruct vision. Safety glasses equipped with side shields provide more complete protection than those without. Safety goggles provide an increased level of protection and should be worn when splashes may occur or glassware may explode/implode under pressure.

Contact lenses should not be worn when working in the laboratory because chemical vapors can permeate the lenses and become trapped on the surface of the eye.

5.3 Face Shields

When working outside of a biosafety cabinet, face protection (e.g., goggles, face shield) should be used for anticipated splashes or sprays of biological materials or chemicals. Face shields are designed to be used in combination with safety goggles to provide additional protection to the face and eyes against splashes and particulate matter. Face shields do not provide adequate protection against large projectiles or liquids, unless they are used in combination with safety goggles. Polycarbonate face shields that offer protection against ultraviolet (UV) radiation should be worn when using instruments that produce UV light.

5.4 Gloves

Gloves should always be worn to prevent direct contact with biological materials and chemicals. Gloves should be comfortable, of sufficient length to prevent exposure of the hand and wrist, and should be appropriate for the type of work to be performed. Gloves should be inspected for visible tears before use, removed aseptically when they become soiled or compromised, and discarded appropriately after use.

Gloves come in a variety of materials that provide different levels of protection. Laboratory personnel should use gloves that provide the highest level of protection against the substances to be used. The *Laboratory Safety Manual* provides additional information about selecting appropriate gloves. Some individuals develop allergies to the materials used to manufacture safety gloves. If this occurs, select a comparable glove made of an alternate material.

5.5 Lab Coats, Gowns, and Coveralls

Lab coats are required for BSL-1 and BSL-2 research laboratories and for instructional laboratories operating at BSL-2. Lab coats are recommended for instructional laboratories operating at BSL-1. Lab coats should cover the entire upper body, extend to the knees, and fit comfortably without hanging too loosely from the arms. For research activities, only single use disposable lab coats or lab coats that are routinely laundered by an approved vendor should be used. Lab coats may not be laundered by laboratory personnel. If disposable gowns are worn, they should be secured in the back, cover the entire upper body, extend to the knees, and fit comfortably without hanging too loosely from the arms. For instructional laboratory activities, disposable lab coats may be used if disposed of and replaced when soiled or compromised.

In animal laboratories, disposable gowns should be worn and secured in the back, cover the entire upper body, extend to the knees, and fit comfortably without hanging too loosely from the arms. It is recommended that gowns be changed between each laboratory or animal room entered. Disposable gowns or coveralls are recommended for all personnel working with or near laboratory or research animals. Personnel are required to wear disposable coveralls when changing bedding in animal cages.

BSL-3 and ABSL-3 laboratories have additional PPE requirements that must be adhered to and require specific training prior to use.

5.6 Hair and Shoe Covers

Personnel working in animal facilities are required to wear disposable a hair cover over the head when conducting cage changes. In addition, wearing shoe covers or using sticky mats is required

at all times in animal spaces. Hair and shoe covers are to be removed prior to exiting the facility. Sticky mats must be replaced every three months or when they are no longer tactile, whichever is more frequent.

5.7 Respiratory Protection

For work with biological materials, respiratory protection should only be considered after feasible engineering controls have been put into place and additional controls are still needed. Personnel are required to wear an N95 respirator when changing bedding in animal cages. In addition, personnel who have allergies to animals may also be required to wear an N95 respirator. N95 respirators are recommended for all personnel working with or near laboratory or research animals.

In general, respiratory protection should fit snugly and form a seal so that air may not leak through the sides of the respirator. George Mason University's *Respiratory Protection Program* is available on the EHS website and provides additional information and guidance on the use, care, and maintenance of respirators. If your work requires you to wear respiratory protection, (e.g., half face, full face, Powered Air Purifying Respirator, particulate mask including N95 or N99), contact EHS prior to beginning work.

5.8 Hearing Protection

George Mason University's Hearing Conservation Program covers any employee exposed to noise levels in excess 85 dBA over an 8-hour period. The program is available on the EHS website and provides additional information on the use and care of hearing protection devices. Hearing protection, provided by ear plugs or ear muffs, should be worn by personnel exposed to the American Conference of Governmental Industrial Hygienists Threshold Limit Value of 85 dBA over an 8-hour period.

In some laboratories, the combination of noises generated by continuously running equipment (e.g., refrigerators, freezers, and incubators) in combination with intermittent use of equipment such as centrifuges, motors, and homogenizers may reach levels that exceed 85 dBA. As a general rule, if an employee must raise his/her voice to speak with someone less than one meter away, then noise levels probably exceed 85 dBA. If you believe noise levels may exceed the action level, contact EHS.

6.0 Procurement of Biological Material

Laboratory personnel who receive infectious materials for research or instructional purposes must be aware of university and government regulations regarding the type and quantity of biological materials they are permitted to receive.

Compliance requirements for acquiring and working with biological material depend on the type of material and its potential hazard. Access to certain material may require the recipient to apply for a permit, license, or registration. Currently, George Mason University laboratories are not authorized to possess agents classified as RG-4. More information on common biological materials requiring a license, permit, or registration is provided below.

6.1 Material Transfer Agreements (MTA)

George Mason University requires a MTA be in place when acquiring or transferring radioactive materials, select agents and toxins, agents on the Commerce Control List, or proprietary materials to or from another institution or investigator. Contact OSP for more information.

6.2 Permit Requirements

Permits may be required for collection or transfer of material within the United States. Field research involving specimen collection may require permits from the United States Fish and Wildlife Service and/or the National Park Service, as well as state or local agencies. The United States Fish and Wildlife Service requires permits for take, import, or export of threatened and endangered species and migratory birds. The National Park Service requires a Scientific Research and Collection Permit for most scientific activities (e.g., fieldwork, specimen collection) pertaining to natural resources or social studies in the National Park System. Contact EHS or these agencies for more information.

Certain materials require import permits prior to entering the United States. Table 2 outlines permit requirements of the CDC, USDA, and the United States Department of Interior (USDI). Contact the appropriate agency for more information. Additionally, permits may be required to collect and export field samples from other countries. PI/LS are responsible for understanding international permitting regulations pertaining to their work.

6.3 Select Agents and Toxins

Select agents and toxins are specific biological materials that have been identified by the HHS and USDA as agents that have potential use in biological terrorism or warfare. PI/LS desiring to acquire, possess, use, or transfer select agents and toxins must notify EHS for assistance in registering with CDC and/or APHIS and be in compliance with pertinent HHS and/or USDA regulations before these materials may be acquired.

6.3.1 Excluded Strains of Select Agents and Toxins

PI/LS must receive IBC approval for acquisition and use of excluded strains (e.g., attenuated strains) and toxins prior to acquiring these materials. This approval must be attained whether the material is acquired through commercial vendors or transferred from another institution or investigator.

6.3.2 Biologically-derived Toxins

PI/LS must receive approval from the IBC before using biologically-derived toxins. Certain toxins are regulated and must be registered with the HHS/USDA above specific possession limits. Table 3 lists these toxins and the amount of toxin that may be stored and used at any one time by PI/LS without registering the toxin with the CDC/APHIS. PI/LS planning to acquire or possess select agents and toxins above specific possession limits must notify EHS for assistance in registration. PI/LS must keep a logbook that lists all toxins and toxin quantities present in the laboratory.

6.3.3 Research Animals

Animals are often used in research or teaching laboratories. PI/LS must obtain IACUC approval prior to procuring all animals for research or instructional purposes. Contact the Office of Research Integrity and Assurance for more information on IACUC approval.

6.4 Receiving Biological Materials

All hazardous materials shipped to George Mason University from a vendor or transferred from another institution must be packaged and transported in accordance with requirements set forth by the DOT or International Air Transport Association (IATA).

Packages should be examined before they are accepted. Any packages that are improperly labeled, contain prohibited materials, or show signs of damage, tampering, or leaking should not be accepted and personnel should contact EHS immediately. Contact EHS in the event that the transporter does not accept the package. Unknown or suspicious packages should be reported to University Police.

Packages containing biological materials should remain in the original packaging and be kept in a secure area until they can be opened using appropriate containment. The dry ice in these packages should be placed in a chemical fume hood to sublime. Never place dry ice into a laboratory sink, as it may cause pipes to rupture.

Table 2. Required Permits for Importation of Biological Material

Agency	Material to be Imported
CDC	Etiologic agents: Infectious agents known to cause disease in man. This includes, but is not limited to, bacteria, viruses, rickettsia, parasites, yeasts and molds. In some instances, agents which are suspected of causing human disease also require a permit.
	Biological materials: Unsterilized specimens of human and animal tissue (including blood), body discharges, fluids, excretions or similar material, when known or suspected of being infected with disease transmissible to man.
	Animals: Any animal known or suspected of being infected with any disease transmissible to man. Importation of turtles of less than four inches in shell length and all NHP require an importation permit issued by the CDC Division of Migration and Quarantine.
	Insects: Any living insect, or other living arthropod, known or suspected of being infected with any disease transmissible to man. Also, if alive, any fleas, flies, lice, mites, mosquitoes, or ticks, even if uninfected. This includes eggs, larvae, pupae, and nymphs as well as adult forms.

Agency	Material to be Imported
	Snails: Any snails capable of transmitting the causative agent of schistosomiasis. No mollusks are to be admitted without a permit from CDC or the USDA. Any shipment of mollusks with a permit from either agency will be cleared immediately.
	Bats: All live bats. Bats may also require a permit from the U.S. Department of Interior, Fish and Wildlife Services.
USDA	Material derived from animals or exposed to animal-source materials, including animal tissues, blood, cells or cell lines of livestock or poultry origin, RNA/DNA extracts, hormones, enzymes, monoclonal antibodies for *in vivo* use in nonhuman species, certain polyclonal antibodies, antisera, bulk shipments of test kit reagents, and microorganisms including bacteria, viruses, protozoa, and fungi. (Note: Exceptions to this requirement are human and NHP tissues, serum, and blood).
	Dairy products (except butter and cheese), and meat products (e.g., meat pies, prepared foods) from countries with livestock diseases exotic to the United States.
	Foreign plant pests injurious to plants grown in the United States.
	Designated noxious weeds, which are of foreign origin and new to or not widely prevalent in the United States.
	Insects, mites, and nematodes introduced for biological control of weeds in the United States.
	Biological control organisms imported, shipped, and released in the United States.
	Insects and mites commonly included in shipments as host material for biological control agents.
	Domestic plant pests regulated by federal or state quarantines.
	Low-risk organisms, including arthropods, and pathogens.
	Nonregulated domestic plant pests shipped into an area in the United States where the pests do not occur.
	Genetically engineered organisms that are plant pests or contain portions (plasmids, DNA fragments, etc.) of plant pests.
USDI	Certain live animals and all live bats.

Table 3. CDC/APHIS Limits on Possession of Toxins

Toxin	Total Amount Allowed per PI/LS (mg)
Abrin	100
Botulinum neurotoxins	0.5
Clostridium perfringens epsilon	100
Conotoxins	100
Diacetoxyscirpenol	1000
Ricin	100
Saxitoxin	100
Shigatoxin	100
Shiga-like ribosome inactivating proteins	100
Staphylococcal enterotoxins	5.0

Toxin	Total Amount Allowed per PI/LS (mg)
Tetrodotoxin	100
T-2 toxin	1000

7.0 Storage of Biological Materials

All biohazardous materials must be kept in areas with restricted access. Additional security precautions must be taken for areas where strains of select agents (including excluded or attenuated strains), toxins, controlled substances, and agents on the Commerce Control List are stored and where animals are housed. All storage areas (e.g., rooms, refrigerators, freezers, and cabinets) must be labeled with the universal biohazard symbol. All containers and/or racks are to be clearly labeled so that contents can be easily identified. Any substance being stored in a freezer should be placed in a labeled container designed for low temperature storage and, in order to prevent unnecessary handling, material should be inventoried by storage location. Under no circumstances should personal items (food and beverages) to be stored in laboratory refrigerators, freezers, or incubators.

8.0 Procedures for Using Biological Materials

The most important element of biological safety is strict adherence to standard microbiological practices and procedures. Individuals working with biohazardous materials must be aware of potential hazards and must be trained and proficient in the practices and techniques required for handling such material safely. The PI/LS is responsible for identifying and adopting biosafety practices and procedures designed to minimize or eliminate exposure to laboratory hazards and for training laboratory personnel.

8.1 Standard Laboratory Practices for BSL-1 Laboratories

BSL-1 is assigned to work involving well-characterized, nonbiohazardous agents not known to cause disease in healthy human adults. A *Project Review Form* must be completed and submitted to EHS for all projects involving BSL-1 materials. All personnel who will be working in or frequenting BSL-1 laboratories must receive appropriate training and should contact EHS for appropriate medical services if they have health considerations that warrant special procedures for work with BSL-1 materials. The following list provides standard laboratory practices to be implemented in BSL-1 laboratories:

- Keep the door to the laboratory closed at all times and locked when the laboratory is not in use.
- Restrict access to the laboratory to appropriately trained individuals when experiments or work with cultures and specimens are in progress.
- Wear appropriate PPE. At a minimum, a lab coat, gloves, and closed toe shoes must be worn when working in the laboratory. Do not wear PPE outside of the laboratory. Lab coats are recommended for instructional laboratories operating at BSL-1. (Refer to Section 5 for additional guidance on PPE.)
- Conduct work either on the open bench top or in a biosafety cabinet. Biosafety cabinets are not required for BSL-1 work. However, in some instances, (e.g., tissue culture) the use of a biosafety cabinet may be needed to prevent contamination of cultures.
- Do not eat, drink, smoke, apply cosmetics, handle contact lenses, or store food and beverages in the laboratory.
- Do not pipette by mouth. Use mechanical pipetting devices.
- Perform procedures carefully to minimize the creation of splashes or aerosols.
- Handle and dispose of sharps safely. Avoid the use of sharps when alternative methods are available.
- Use an appropriate disinfectant to decontaminate work surfaces, equipment, instruments, and glassware daily and immediately after a spill. This includes a wipe down of the biosafety cabinet if one is used.
- Wash hands after completing work and before leaving the laboratory.
- Dispose of all BSL-1 waste according to waste handling procedures.
- Follow additional laboratory procedures outlined in the *Laboratory Safety Manual* for work involving chemicals.

8.2 Standard Laboratory Practices for BSL-2 Laboratories

BSL-2 is assigned to work with infectious agents, potentially infectious agents, and material that cause disease in humans with a varying degree of severity and are a moderate hazard to laboratory personnel and low hazard to the community. IBC approval is required before beginning any BSL-2 projects. All personnel who will be working in or frequenting BSL-2 laboratories must receive appropriate training and should contact EHS for appropriate medical services before beginning work in the laboratory. Personnel may be required to receive vaccinations before beginning work. In addition, a *Supplemental Laboratory Safety Plan* specific to the infectious materials, activities, safety issues, and operations of that laboratory must be developed and implemented. The following list provides standard laboratory practices to be implemented in BSL-2 laboratories:

- Follow all of the standard practices for BSL-1 laboratories.
- Restrict access to those personnel who have been properly trained with regard to hazards present in the laboratory and who meet specific entry requirements (e.g., immunizations). It is recommended that individuals who are at increased risk of acquiring infection or for whom infection may be unusually hazardous do not enter or work in the laboratory.
- Follow the *Supplemental Laboratory Safety Plan* and procedural SOP when handling infectious material.
- Follow Universal Precautions for all work involving material of human origin.
- Make sure that the laboratory entrance is properly posted to indicate the hazards present and specific entry requirements.
- Label equipment, storage areas, and usage areas where BSL-2 materials are used or stored.
- Perform all procedures that generate aerosols or have the potential to splash or splatter in a biosafety cabinet.
- Wear face protection (goggles, mask, face shield, or other splatter guard) for anticipated splashes or sprays when infectious material must be handled outside the biosafety cabinet.
- Follow safety guidelines provided below for the use of pipettes, needles, and syringes with biohazardous materials.
- Use an appropriate disinfectant to decontaminate work surfaces, equipment, biosafety cabinets, instruments, and glassware after each use and immediately after a spill, splash, or other contamination by biohazardous material.
- Decontaminate all equipment before it is sent for repair or maintenance. Equipment must also be decontaminated before removal from the facility when it must be packaged for transport. Packaging and shipment shall be in accordance with applicable local, state, and federal regulations.
- Maintain biological spill supplies within the laboratory.
- Report spills, accidents, narrowly-avoided accidents (i.e., near misses), and disease symptoms that may be related to laboratory acquired infection to the PI/LS and EHS.
- Dispose of all BSL-2 waste according to waste handling procedures.
- Use mechanical means such as tongs, forceps, or a brush and dustpan to remove broken glass. Never handle broken glass directly with hands. Dispose of broken glass contaminated with infectious material as BSL-2 waste.
- Follow additional laboratory procedures outlined in the *Laboratory Safety Manual* for work involving chemicals.

8.2.1 Safety Guidelines for the Use of Pipettes with Biohazardous Material

The following guidelines should be followed when pipetting biohazardous material:

- Never pipette by mouth.
- Substitute plastic pipettes for glass, whenever possible.
- Use only cotton plugged pipettes for transferring infectious or toxic material.
- Always use pipetting aids when pipetting infectious material.
- Pipette biohazardous material in a biosafety cabinet.
- To catch drops or spills, place a towel or other absorbent material dampened with disinfectant on the work surface. Plastic backed bench paper is suitable for this purpose. Treat discarded bench paper or absorbent material as biohazardous waste.
- To reduce the likelihood of splashes, dispense material from a pipette as close as possible to the bottom of the container, allowing the material to run down the inner wall of the container.
- Contaminated pipettes should be disposed of in sharps containers or pipette keepers and treated as biohazardous waste.
- Reusing pipettes is discouraged, however; if it is necessary to reuse certain types of pipettes, disinfect the pipettes by placing them horizontally in a pan or tray containing enough suitable disinfectant, such as 10% bleach, to allow complete immersion of the pipettes. After 20 minute contact time, rinse and autoclave all pipettes.

8.2.2 Safety Guidelines for the Use of Sharps with Biohazardous Material or Research Animals

To lessen the risk of accidental infection, aerosol generation, or spills, the use of sharps should be avoided when alternate methods are available. Sharps are instruments or equipment capable of causing a puncture or cut, including needles, scalpels, razor blades, glass Pasteur pipettes, slides, and broken glassware.

Sharps should be stored in a manner that prevents injury and should never be left unattended in a manner that could result in an accidental injury. Personnel should be familiar with proper storage, use, and disposal of sharps. For animal handlers, injury could occur when restraining an animal during a procedure involving the use of needles.

The following precautions should be observed when working with sharps:

- Always use safe needle devices (e.g., self-sheathing needle, retractable needle), unless not feasible for work being conducted.
- Use disposable needles and safety needle-locking syringes. Replace glass syringes with plastic disposable syringes whenever possible.
- Never reuse needles.
- Dispose of entire unit (syringe and safety needle) into sharps container. Do not bend, shear, recap or otherwise manipulate the needle.
- Be sure that the safety needle is locked securely into the barrel before performing any operations.
- Use the syringe and safety needle in a biosafety cabinet when possible.
- Avoid quick and unnecessary movements while holding the syringe.

- Fill the syringe carefully to minimize air bubbles and frothing of the inoculum.
- Expel excess air, liquid, and bubbles from a syringe vertically into a cotton pledget moistened with an appropriate disinfectant.
- Immediately discard used pledget into a biohazard bag.
- Do not use the syringe to forcefully expel a stream of infectious fluid into an open vial for the purpose of mixing.
- When appropriate, restrain or sedate research animals during procedures to decrease risk of sharps injury.

8.3 ABSL-1 Laboratory Practices

ABSL-1 is suitable for animal work that involves 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. All personnel who will be working in or frequenting animal facilities must take *Animal & Vivarium Safety* training before beginning work in the facility and all projects must be approved by IACUC before the project may begin. Animal projects involving biological agents must also be reviewed by the IBC. The following list outlines standard laboratory practices to be implemented in ABSL-1 laboratories:

- Follow standard practices for BSL-1 laboratories.
- Attend *Animal & Vivarium Safety* training before beginning work in the facility.
- Complete the *Animal Handler Questionnaire* and get a physical upon initial assignment to an animal facility, as well as every three years.
- Keep the door to the laboratory closed at all times and locked when the laboratory is not in use.
- Restrict access to personnel required for program or support purposes. Personnel should not enter the laboratory before they have been advised of the potential hazards and instructed in proper safeguards.
- Wear appropriate PPE. At a minimum, a lab coat, gloves, eye protection, and closed toe shoes must be worn when working in the facility. Personnel are also required to wear safety glasses, disposable coveralls, hair cover, and an N95 respirator when changing bedding in animal cages.
- Do not wear PPE outside of the facility.
- Do not eat, drink, smoke, apply cosmetics, handle contact lenses, or store food and beverages in the facility.
- Do not pipette by mouth. Use mechanical pipetting devices.
- Perform procedures carefully to minimize the creation of splashes or aerosols.
- Handle and dispose of sharps safely. Avoid the use of sharps when alternative methods are available.
- Use an appropriate disinfectant to decontaminate work surfaces, equipment, and instruments, daily.
- Wash hands after handling cultures and animals, after removing gloves, and before leaving the animal facility.
- Report all bite or scratch incidents to EHS.
- Dispose of all animal facility waste according to waste handling procedures.

- Post a biohazard sign on the entrance to the animal room whenever infectious agents are present. The hazard warning sign identifies the infectious agent(s) in use, lists the name and telephone number of the responsible person(s), and indicates the special requirements for entering the animal room.
- Follow additional laboratory procedures outlined in the *Laboratory Safety Manual* for work involving chemicals.

8.4 ABSL-2 Laboratory Practices

ABSL-2 is assigned to animal work involving agents associated with human disease. All personnel who will be working in or frequenting ABSL-2 must take *Animal & Vivarium Safety* training before beginning work in the facility and all projects must be approved by IACUC before the project may begin. Animal projects involving biological agents must also be reviewed by the IBC. In general, persons who may be at increased risk of acquiring infection, or for whom infection might be unusually hazardous, are not allowed in the animal facility unless special procedures can eliminate the additional risk. The following list provides standard laboratory practices to be implemented in ABSL-2 laboratories:

- Follow all of the standard practices for ABSL-1 facilities and BSL-2 laboratories.
- Restrict access to the room to the smallest number of individuals possible.
- Limit the animal population to those used for the experiment.
- Make sure that the laboratory entrance is properly posted.
- Label equipment, storage areas, and usage areas where BSL-2 materials are used or stored.
- Use a biosafety cabinet, or other physical containment device, and/or PPE (e.g., face shields) when conducting procedures with a high potential for creating aerosols. These include necropsy of infected animals, harvesting of tissues or fluids from infected animals or eggs, or intranasal inoculation of animals.
- Follow the *Supplemental Laboratory Safety Plan* and SOP when handling infected animals.
- Follow safety guidelines for the use of pipettes, needles, and syringes with biohazardous materials.
- When inoculating animals:
  - Position the hand that is holding the animal so as to prevent needle stick injuries.
  - Be sure the animal is properly restrained prior to inoculation and be alert for any unexpected movements of the animal.
  - Before and after injection, swab the injection site with an appropriate antiseptic.
- Equipment and work surfaces in the room must be routinely decontaminated with an effective disinfectant after work with the infectious agent, and especially after spills, splashes, or other contamination by infectious materials.
- Decontaminate all equipment before it is removed from the facility.
- Collect, label, transport, and process all infectious material in a manner that contains and prevents transmission of the agent(s).
- Wash hands after handling cultures and animals, after removing gloves, and before leaving the animal facility.
- Limit housing to animals being used for the experiment(s).

- When needed, animals should be housed in primary biosafety containment equipment appropriate for the animal species. Handle filter top cages in properly designed and operating animal biocontainment cabinets recommended for rodents.
- Maintain biological spill supplies within the laboratory.
- Report spills, accidents, narrowly-avoided accidents (i.e., near misses), bites, scratches, and disease symptoms that may be related to laboratory acquired infection to the PI/LS and EHS. Spills and accidents which result in exposure to infectious materials must be immediately reported to the PI/LS and EHS. Medical evaluation, surveillance, and treatment are provided as appropriate and written records are maintained.
- Dispose of all animal facility waste according to waste handling procedures.
- Follow additional laboratory procedures outlined in the *Laboratory Safety Manual* for work involving chemicals.

8.5 Special Provisions for Select Agents and Toxins

In addition to the general safety guidelines mentioned above, special precautions are required when handling select agents and toxins, high consequence livestock pathogens or toxins, and plant pathogens with a high degree of acute toxicity. These procedures are outlined in the *Biomedical Research Laboratory Select Agent Program*.

8.6 Shipping and Transport of Biological Material

Shipping and transport of biological materials (including biological specimens, infectious material, and clinical specimens), as well as other hazardous materials (chemicals, dry ice, radioactive materials), are strictly regulated. The Public Health Service determines the hazard classes of etiologic agents and biohazardous materials. DOT addresses how these materials should be regulated in packaging, shipment, and transport of hazardous materials by ground. IATA publishes regulations for how these materials must be packaged and shipped by air. The following sections outline procedures that must be followed when transporting biological material on or between campuses or shipping material offsite. EHS provides *Shipping Class 6.2 Dangerous Goods and Dry Ice* training as needed.

8.6.1 Intracampus Transport of Biological Material or Animals

Caution must be used to prevent spills or accidental exposure when transporting biological material. The following procedures should be followed when transporting biological material between laboratories or buildings:

For nonbiohazardous material:

- Place material in a nonbreakable, sealable primary container.
- Place the sealed primary container in a sealable secondary container.

For biohazardous material:

- Decontaminate the outside of the primary container before placing it into the secondary container.
- Place a “biohazard” label on the outside of the secondary container.

- Place sufficient absorbent material between the primary container and secondary container to absorb the volume being transported.
- Use a cart to transport materials.

For research animals:

Prior to transport, relocation of research animals must be authorized and approved by the IACUC. Due to public health, animal health, security, and public relations concerns, intracampus transport of laboratory animals must be done only in their primary enclosure or cage, or in escape-proof IACUC-approved transport boxes. In general, when transporting laboratory animals, cages or other transport devices must have solid bottoms or be placed in a secondary container such that animal waste is contained. Transport devices must be appropriately sanitized between uses. Animals may not be transported in passenger elevators or in personal vehicles.

8.6.2 Intercampus Transport of Biological Material

Exempt patient specimens and noninfectious material may be transported in personal vehicles between campuses by laboratory personnel (excluding students). Biohazardous material, other than clinical specimens, may not be transported in personal vehicles. These materials must be shipped according to DOT/IATA regulations.

Transportation of research animals between campuses is prohibited without IACUC approval and authorization. Due to public health, animal health, security, and public relations concerns, intercampus transport of laboratory animals must be done only in their primary enclosure or cage, or in escape-proof IACUC-approved transport boxes. In general, when transporting laboratory animals, cages or other transport devices must have solid bottoms or be placed in a secondary container such that animal waste is contained. Transport devices must be appropriately sanitized between uses. Animals may not be transported in passenger elevators or in personal vehicles.

8.6.3 Shipping Biological Material

International and domestic transport regulations for infectious substances are designed to prevent the release of these materials in transit to protect the public, workers, property, and the environment from the harmful effects that may occur from exposure to these materials. Protection is achieved through rigorous packaging requirements and hazard communication. Packages must be designed to withstand rough handling and other forces experienced in transportation, such as changes in air pressure and temperature, vibration, stacking, and moisture. Hazard communication includes shipping papers, labels, markings on the outside of packaging, and other information necessary to enable transport workers and emergency response personnel to correctly identify the material and respond efficiently in an emergency situation. In addition, shippers and carriers must be trained on these regulations so they can properly prepare shipments and recognize and respond to the risks posed by these materials.

Personnel who plan to ship biological materials must complete *Shipping Class 6.2 Dangerous Goods and Dry Ice Training* offered by EHS. Packaging and shipping must be conducted in accordance with DOT and IATA regulations as outlined in this training. An MTA may be required when transferring materials to another institution or Investigator. More information on

MTAs are available from OSP. In addition, depending on the material to be shipped and the shipping destination, a United States export permit or an import permit from the country that is to receive the package may be required. PI/LS are responsible for understanding international permitting regulations pertaining to their work. EHS is available to assist laboratory personnel in shipping biological materials.

Select agents and toxins include infectious substances that have been identified by HHS and the USDA as having the potential to pose a severe threat to public health and safety. Additional requirements for shipping select agents and toxins are provided in the *Biomedical Research Laboratory Select Agent Program Manual*.

9.0 Decontamination Procedures

Decontamination is a process by which contaminated surfaces, equipment, instruments, or waste is rendered safe to handle. The primary objective of decontamination is to reduce the level of microbial contamination so that the potential for infection transmission is eliminated. The decontamination process may involve washing contaminated items with soap and water, sterilization, or disinfection.

9.1 Sterilization

Sterilization is a process that kills all microorganisms, including a high number of bacterial endospores. The probability of a microorganism surviving the process is one in one million. Sterilization can be accomplished by heat, ethylene oxide, hydrogen peroxide, plasma, ozone, and radiation.

9.1.1 Steam Sterilization

Moist heat sterilization (autoclaving) is used to sterilize laboratory equipment and culture media, and to decontaminate biological waste. Autoclaving uses steam under pressure (approximately 15 pounds per square inch) to achieve a chamber temperature of at least 121°C (250°F). To be effective, air in the autoclave chamber must be replaced by steam for an adequate exposure time. RMW must be sterilized at 121°C at 15 pounds per square inch for 2 hours. Noninfectious waste from BSL-1 laboratories may be sterilized for a shorter period of time (30 minutes) at 121°C at 15 pounds per square inch.

Caution should be used when using steam sterilization. Steam under pressure can be a scalding hazard. Laboratory personnel should not use autoclaves without proper training and should exercise caution when opening an autoclave. Allow fluids to cool prior to removal from the autoclave.

9.1.2 Dry Heat Sterilization

Dry heat sterilization is used for sterilization of anhydrous oils, greases, powders, etc., that cannot be easily permeated by steam. Dry heat is less efficient than moist heat sterilization and requires longer times and higher temperatures (160–170°C [320-338°F]) for periods of 2 to 4 hours.

9.2 Disinfection

Disinfection eliminates nearly all recognized pathogenic microorganisms but not necessarily all forms (bacterial endospores) on inanimate objects. The effectiveness of a disinfection procedure is controlled significantly by a number of factors:

- Nature and number of contaminating microbes.
- Amount of organic matter present.
- Type and condition of items to be disinfected.
- Temperature.

Chemical disinfectants are used in the laboratory to treat a surface or an item before and/or after routine use, or after a spill or other contamination. Disinfection can be achieved with liquid or

gas germicidal agents. Because disinfectants are antimicrobial, they may, by their nature, also have toxic effects for personnel. Therefore, MSDS and other manufacturer's product information should be available and thoroughly reviewed before using these products. Appropriate PPE must be worn when using disinfectants and these compounds must be used in well-ventilated areas.

9.2.1 Liquid Disinfectants

Liquid disinfectants are frequently used for surface decontamination and, at sufficient concentration, to decontaminate liquid waste. All disinfectants are not equally effective in decontaminating biohazardous material. Factors such as temperature, contact time, pH, dispersion rate, penetrability and reactivity of the material at the application site must be considered when selecting the appropriate disinfectant. Hazardous properties of the disinfectant, relative to its efficacy must also be considered. A description of different types of liquid disinfectants is provided below and in Table 4.

Because the local sewer authority sets limitations on drain disposal of chemicals, EHS requires that PI/LS seek written approval from EHS regarding sink disposal of any chemical disinfectants other than 70% alcohol (ethanol or isopropanol) and bleach. Disinfectant solutions not authorized for sewer disposal may be collected as hazardous waste.

9.2.1.1 Alcohols

The most commonly used alcohols, ethanol and isopropanol, are most effective at concentrations of 70%. Both higher and lower concentrations are less effective. Alcohols are active against vegetative bacteria, fungi, and lipid viruses but not against spores. They are only moderately effective against nonlipid viruses. Alcohols are difficult to use as contact disinfectants because they evaporate rapidly and do not penetrate organic material well. When using alcohols, it is best to clean an object and then submerge it in alcohol for the appropriate time. Alcohol is not a registered tuberculocidal or HIV listed disinfectant.

9.2.1.2 Chlorine Compounds

The most commonly used and generally effective disinfectant is sodium hypochlorite (household bleach). Sodium hypochlorite is an appropriate disinfectant for a wide range of bacterial and viral agents. Household bleach contains 5% sodium hypochlorite and should be diluted before use. In waste collection flasks, bleach should be added to 10% of the final collection volume to achieve a final bleach concentration of 10%.

Sodium hypochlorite is a strong oxidizing agent and therefore can be corrosive to metal. Additionally, the presence of high concentrations of protein can inactivate the action of chlorine products. A sealed bottle of bleach will lose about 25% of the chlorine in a year. Do not use an unopened bottle of bleach that is more than six months old. An opened bottle will lose 25% of the chlorine over 30 days. Do not use an opened bottle more than 30 days old. A freshly prepared stock solution is only effective for 24 hours.

9.2.1.3 Glutaraldehyde

Glutaraldehyde is usually supplied as a 20% solution and requires activation by the addition of an alkaline agent prior to use. The activated product may be kept for about two weeks and should be discarded when turbid. Glutaraldehyde is active against all microorganisms, but is toxic,

irritating, and mutagenic and should be used only when necessary. Please follow the manufacturer's guidance when using glutaraldehyde-based products because there are many different formulations that have been designed for specific uses.

9.2.1.4 Hydrogen Peroxide

Hydrogen peroxide is usually available as a 30% solution. It may be diluted 1:5 for use as a disinfectant. It is active against a wide array of microorganisms. However, it is an oxidizing agent and should not be used on aluminum, copper, zinc, or brass. Hydrogen peroxide is unstable at high temperatures and in light.

9.2.1.5 Iodine and Ionophors

Iodine and iodophors, compounds in which the iodine is combined with a solubilizing or carrier agent, are general, all-purpose disinfectants with an action similar to that of chlorine products. The appropriate concentration for iodine-containing products is 75 parts per million available iodine for disinfecting work surfaces. Concentrations may be much higher for other purposes. Like chlorine compounds, the effectiveness of iodine compounds may be diminished in the presence of protein/organic material. Iodophor compounds that are used for antisepsis (germicide applied to tissue or skin) are not appropriate for use as hard-surface disinfectants and vice versa. Please read the product material for appropriate dilutions and applications.

9.2.1.6 Phenol and Phenolic Compounds

Phenolic compounds are active at 0.2 to 3% concentrations against all forms of vegetative microorganisms but not against spores. They have only limited effectiveness against nonlipid viruses. There are many common disinfectants based on phenol and they should be used according to the manufacturer's recommendations.

9.2.1.7 Quaternary Ammonium Compounds

Compounds in this class are active at concentrations of 0.1 - 2%. They are active against vegetative bacteria, lipid viruses, but not against bacterial spores, nonlipid viruses, or tubercle bacilli. These compounds should be used only when a low-level disinfectant is required.

9.2.2 Gas and Vapor Disinfectants

Certain gases, when used in closed systems and under controlled conditions of temperature and humidity, are an effective means of disinfection. Gas disinfectants are primarily used to decontaminate biosafety cabinets, bulky or stationary equipment not suited to liquid disinfectants, instruments or optics that might be damaged by other decontamination methods, and rooms, buildings, and associated air-handling systems. Due to their hazardous nature, the use of gas disinfectants is tightly controlled. If gas disinfectants are going to be used in the laboratory, contact EHS for special monitoring requirements.

9.2.2.1 Ethylene Oxide

Ethylene oxide gas is most effective at a concentration of 400 to 500 mg/liter at 50-60°C (122-140°F) with 30 to 40% humidity and a contact time of 2 to 24 hours depending on temperature. Ethylene oxide is a suspected carcinogen with explosive properties.

9.2.2.2 Vapor Phase Hydrogen Peroxide

Vapor phase hydrogen peroxide is effective at a 30% concentration (less than 10 mg/liter) to disinfect surfaces. Contact time is a factor of temperature. The higher the temperature, the less contact time is needed. The end products of disinfection are nontoxic (water and oxygen) which makes vapor phase hydrogen peroxide safer to use than other gas disinfectants. However, hydrogen peroxide vapors are corrosive to some materials and degrade natural rubber and nylon.

9.2.2.3 Ozone

Ozone is effective at a concentration of 2 to 5 mg/liter at 25°C (77°F). Ozone possesses oxidizing properties that inhibit bacterial growth and react with amino acids and nucleic acids.

9.2.2.4 Formaldehyde Gas (from heating paraformaldehyde)

Gaseous formaldehyde is effective at a concentration of 0.3 gm/cu ft at a temperature of 20-22°C and humidity of 60 to 85% and a contact time of 6 to 8 hours. Formaldehyde is a toxic irritant and a human carcinogen. Disinfection with formaldehyde gas requires aeration and time for the formaldehyde to off-gas (usually 8 hours).

9.3 Ultraviolet Radiation

The UV-C band of UV radiation (250-270 nm, 265 optimum) is effective in destroying most microorganisms in air, water, and on surfaces when direct exposure to UV radiation is possible. The effectiveness of UV disinfection is reduced when dirt, dust, or shadows interrupt or interfere with the path of the UV rays. EHS does not recommend UV sterilization for biosafety cabinets.

9.4 Decontamination of Spore-forming Microorganisms

Bacterial spores, by nature, are resistant to extreme physical, chemical, and thermal conditions, and are second only to prions in their resistance to different types of decontamination. For this reason, particular care and diligence should be used to decontaminate equipment and apparatus used for work with spore-forming agents. Adequate sterilization requires direct exposure to 121°C (250°F) for at least 90 minutes. For chemical decontamination methods, refer to Table 4.

Table 4. Chemical Disinfectant Selection and Use

	Quaternary Ammonium Compounds	Phenolic Com-pounds	Chlorine Com-pounds	Iodophor Com-pounds	Alcohol (ethyl or isopropyl)	Glutaral-dehyde
Concentration of active ingredient	0.1-2%	0.2-3%	0.01-5%	0.47%	70-85%	2%
Contact time (minutes)	10-30	10-30	10-30	10-30	10-30	10-600
Vegetative bacteria	+	+	+	+	+	+
Bacterial spores			+/-			+
Lipo viruses	+	+	+	+	+	+
Hydrophilic viruses		+/-	+	+/-	+/-	+
Tubercle bacilli		+	+	+		+
HIV	+	+	+	+	+	+
HBV		+/-	+	+/-	+/-	+
Contaminated liquid discard			+			

	Quaternary Ammonium Compounds	Phenolic Com-pounds	Chlorine Com-pounds	Iodophor Com-pounds	Alcohol (ethyl or isopropyl)	Glutaral-dehyde
Contaminated glassware	+	+	+		+	+
Contaminated instruments		+				+
Equipment total decontamination						

Key: + denotes very positive response
+/- denotes a less positive response
Blank denotes a negative response or not applicable

9.5 Inactivation of Biologically-derived Toxins

Although biologically-derived toxins are often considered hazardous chemicals, they will be addressed in this manual because of their frequent use in biomedical research. Equipment and apparatus contaminated with toxins and waste generated from procedures involving toxins must undergo decontamination to inactivate the toxins.

Table 5 lists effective methods for inactivating or denaturing toxins. In general, high molecular weight, proteinacious bacterial toxins are inactivated by steam sterilization (autoclaving 1 hour at 121°C), where as low molecular weight toxins (e.g., mycotoxins, marine and reptile venoms) are more effectively inactivated by treatment with sodium hypochlorite (NaOCl) or a mixture of sodium hypochlorite and sodium hydroxide.

Table 5. Inactivation of Biologically-derived Toxins

Toxin	Autoclave at 121°C for 1hr	2.5% NaOCl for 1hr	2.5% NaOCl, 0.25N NaOH for 1 hr	2.5% NaOCl, 0.25N NaOH for 4 hr
Abrin	Yes	NO	NO	NO
Botulinum neurotoxins	Yes	Yes	Yes	Yes
Clostridium perfringenes epsilon toxin	Yes	Yes	Yes	Yes
Diacetoxyscirpenol (DAS), T-2	NO	NO	NO	Yes
Palytoxin	NO	Yes	Yes	Yes
Ricin	Yes	Yes	Yes	Yes
Saxitoxin	NO	Yes	Yes	Yes
Shigatoxin and Shiga-like toxins	Yes	Yes	Yes	Yes
Staphylococcal enterotoxins	Yes	Yes	Yes	Yes
Tetrodotoxin	NO	NO	Yes	Yes

9.6 Decontamination Procedures for Prions

Prions are characterized by resistance to conventional inactivation procedures including irradiation, boiling, dry heat, and chemicals (formalin, betapropiolactone, alcohols). While prion infectivity in purified samples is diminished by prolonged digestion with proteases, results from boiling in sodium dodecyl sulfate and urea are variable. Likewise, denaturing organic solvents such as phenol or chaotropic reagents such as guanidine isothiocyanate have also resulted in

greatly-reduced but not complete inactivation of prions. Formalin-fixed and paraffin-embedded tissues, especially of the brain, remain infectious. The safest and most unambiguous method for ensuring that there is no risk of residual infectivity on contaminated instruments and other material is to discard and destroy them by incineration. Current methods for inactivation of prions are based on the use of sodium hypochlorite, sodium hydroxide, and the moist heat of autoclaving in combination. Contact EHS for more information on deactivating prions.

9.7 Decontamination of Equipment

Equipment must be decontaminated before being removed from the laboratory (e.g., repair, laboratory relocation, surplus, or disposal). A *Decontamination Certificate* certifying that the equipment was properly decontaminated must accompany the equipment. *Decontamination Certificates* are available on the EHS website. If you feel you cannot properly decontaminate equipment or materials, contact EHS for further assistance.

10.0 Waste Management

Biological waste must be managed in accordance with state, local, and federal regulations. To facilitate proper handling of waste, EHS provides all containers and supplies, and is available to provide laboratory-specific training regarding laboratory waste disposal.

10.1 BSL-1 Waste

It is industry standard to disinfect solid and liquid BSL-1 (nonbiohazardous) waste. However, if the BSL-1 waste contains any hazardous chemicals (e.g., heavy metals, organic solvents) it must be handled as hazardous waste as outlined in the *Laboratory Safety Manual*. Similarly, if the BSL-1 waste contains radioactive materials it should be handled as outlined in the *Radiation Safety Manual*.

Containers with autoclavable liners are provided by EHS for disposal of solid BSL-1 waste. Bags with the biohazard symbol or bags that are red or orange in color must not be used for BSL-1 waste.

10.2 Regulated Medical Waste (RMW)

BSL-2 and BSL-3 waste is biohazardous and is treated as RMW in accordance with regulations set forth by the Virginia DEQ (9VAC20-120). RMW, as defined in 9VAC20-120-150, includes:

- **Cultures and stocks of biological materials:** Discarded cultures, stocks, specimens, vaccines, and associated items likely to contain agents that are likely to be pathogenic to healthy humans; discarded etiologic agents; wastes from the production of biological materials; and antibiotics likely to have been contaminated by agents that are likely to be pathogenic to healthy humans.
- **Human Materials:** Wastes consisting of human blood or human body fluids or items contaminated with human blood or human body fluids; all human anatomical wastes and all wastes that are human tissues, organs, or body parts.
- **Sharps:** Sharps likely to be contaminated with agents that are pathogenic to healthy humans; all needles, syringes with attached needles, suture needles, and scalpels, including all sharps generated through veterinary practice.
- **Animal carcasses, body parts, bedding, and related wastes:** Animal carcasses, body parts, bedding, and all other wastes likely to have been contaminated as a result of animals being intentionally infected with agents likely to be pathogenic to healthy humans.
- **Contaminated soil, water or other debris**: Any residue or contaminated soil, water, or other debris resulting from the cleanup of a spill of any RMW.
- **Contaminated solid waste:** Any solid waste contaminated by or mixed with RMW (including animal bedding from ABSL-2 facilities).

According to state regulations, solid RMW can be managed either by steam sterilization or incineration. RMW, with the exception of sharps containers, clinical waste, and animal carcasses, is autoclaved prior to disposal. Clinical waste and sharps are placed in biohazard burn boxes for incineration (see below). The Virginia DEQ stipulates that facilities appoint a Waste Management Facility Operator who oversees waste operations. George Mason University has a Waste Management Facility Operator at both the Fairfax and Prince William campuses.

Red containers and autoclavable red biohazard bags that display the biohazard symbol are provided by EHS for disposal of BSL-2 waste. Containers should be double lined with two red, autoclavable biohazard bags. Sharps containers are provided for disposal of needles and syringes, scalpels, suture needles, lancets, glass Pasteur pipettes, glass slides or cover slips, pipette tips, and broken glass.

Procedures for handling BSL-3 and ABSL-3 waste are outlined in the *Biomedical Research Laboratory Biosafety Plan*.

10.3 Ethidium Bromide and Acrylamide Waste

Waste products resulting from electrophoresis procedures are common to biological and biochemical laboratories. Ethidium bromide is a potent mutagen. Solid ethidium bromide waste (e.g., contaminated paper towels, bench paper, and empty storage containers) and agarose or polyacrylamide gels containing ethidium bromide must be disposed of in a burn box. Solutions containing ethidium bromide must be handled and labeled as hazardous waste as outlined in the *Laboratory Safety Manual*.

Acrylamide is a severe neurotoxin and potential carcinogen and mutagen. Solid acrylamide waste including contaminated solids (paper towels, bench paper, empty storage containers) and polyacrylamide gels should be discarded in a burn box provided by EHS. Aqueous solutions containing acrylamide should be handled and labeled as hazardous waste as outlined in the *Laboratory Safety Manual*.

10.4 Waste Containing Biologically-derived Toxins

Waste contaminated with toxins may be autoclaved or chemically deactivated via sodium hypochlorite or a mixture of sodium hypochlorite and sodium hydroxide prior to disposal. Table 4 provides effective decontamination methods for a variety of toxins.

10.5 Animal Waste

Animal carcasses should be disposed of in burn boxes to be incinerated offsite. If the animal carcasses are contaminated with RG-2 organisms, they should be treated as RMW and placed in a container with the biohazard symbol. If the animal carcasses are not contaminated, they should be placed in an unmarked burn box.

Bedding waste collected in bedding dump stations may be disposed of as municipal waste if the animals were not contaminated. If the animals were contaminated, the bedding waste should be treated as RMW. The contaminated bedding waste should be collected, autoclaved, and disposed of accordingly.

10.6 Autoclaving Waste

All laboratory personnel whose position requires them to use an autoclave for the purposes of treating waste are required to attend *Biological Safety for BSL-2 Laboratories* provided by EHS.

The following types of waste can be autoclaved:

- BSL-1 waste (with the exception of animal carcasses and waste containing radioactivity or hazardous chemicals).
- RMW from BSL-2, BSL-3, ABSL-2, and ABSL-3 laboratories on the Prince William campus (with the exception of clinical waste, biological/radioactive waste, biological/chemical waste, and animal carcasses).

The procedure for autoclaving waste follows:

- Autoclave waste within 24 hours.
- Take waste to autoclave room for sterilization. Never leave waste in the autoclave room unattended (if the autoclave is not available take waste back to the laboratory). Use a cart to safely transport waste and keep the waste off of the floor.
- Fill in the autoclave logbook with date, time, name, type of waste, and approximate weight of RMW treated.
- Autoclave waste according to regulations provided in *Biological Safety for BSL-2 Laboratories*. *Biological Safety for BSL-2 Laboratories* covers the regulations associated with treating RMW. Loading, unloading, programming, and activation of the autoclave should be covered in *Laboratory Specific Training* conducted by the PI/LS.
- Do not autoclave liquid waste that contains hazardous chemicals (e.g., heavy metals, organic solvents, and corrosives including bleach) or radioactive materials.
- **For solid BSL-1 waste:**
  - After autoclaving, place the waste in an opaque regular trash bag and discard in domestic trash.
- **For solid RMW generated in BSL-2 laboratories:**
  - Place autoclaved RMW in an orange bag that is labeled with a Treatment Certification Label (available in the Autoclave Log in each autoclave room) that includes a certification that the waste is no longer hazardous, the date the waste was autoclaved, generator name (George Mason University), full address, and contact number.
  - Discard orange bags in the regular trash dumpster within one day.
  - Decontaminate the cart and red waste container after each use.
- **For RMW generated in ABSL-2 laboratories:**
  - Place autoclaved waste in a lined biohazard burn box.
  - Close the burn box when it is 75% full. Do not place more than 40 lbs of waste in a burn box or overfill the burn box so that the box liner cannot be easily tied or taped shut.
  - Label box with a waste sticker indicating the laboratory the waste came from and a general description of the contents.
  - Call EHS for pickup.
  - Decontaminate the cart and red waste container after each use.
- **For liquid waste:**
  - Use pH paper to verify that pH is between 5.5 and 10. Adjust pH if necessary.
  - Dispose of autoclaved BSL-1 liquid waste down the drain with copious amounts of water.
- **For RMW and liquid waste generated in BSL-3 and ABSL-3 laboratories:**
  - Follow procedures for handling BSL-3 and ABSL-3 waste outlined in the *Biomedical Research Laboratory Biosafety Plan.*

10.7 Packaging Waste for Incineration

EHS supplies biohazard burn boxes and nonbiohazard burn boxes for waste that is to be incinerated.

10.7.1 Biohazard Burn Boxes

The following types of waste may be placed in biohazard burn boxes:

- RMW generated in clinical laboratories.
- Sharps containers.
- Contaminated animal carcasses.

Package waste in biohazard burn boxes as follows:

- Obtain burn box and red biohazard burn box liners from EHS. Do not use burn boxes obtained from other sources unless they are the same type of box.
- Set up the burn box and tape the bottom and the side seams with packing tape to prevent leakage.
- Line the burn box with a red biohazard bag.
- Place closed waste bags and containers into the burn box until the box is 75% full. Do not place waste directly into the burn box. Do not overfill the box or exceed a maximum weight of 40 lbs.
- Tie or tape the box liner. Use packing tape to seal the top of the box.
- Contact EHS for pickup.

10.7.2 Nonbiohazard Burn Boxes

The following types of waste may be placed in nonbiohazard burn boxes:

- ABSL-1 animal carcasses.
- Specimens (e.g., fetal pigs, sheep hearts, eyes) used in instructional courses.
- Solid ethidium bromide waste.
- Solid acrylamide waste.

Package waste in nonbiohazard burn boxes as follows:

- Obtain burn box and burn box liners from EHS. Do not use burn boxes obtained from other sources unless they are the same type of box.
- Set up the burn box and tape the bottom with packing tape.
- Line the burn box with a plastic liner.
- Place closed waste bags and containers into the burn box until the box is 75% full. Do not place waste directly into the burn box. Do not overfill the box or exceed a maximum weight of 40 lbs.
- Tie or tape the box liner. Use packing tape to seal the top of the box.
- Contact EHS for pickup.

10.8 Disinfecting Liquid Waste Using 10% Bleach

- Prior to the collection of the liquid waste, add 10% bleach to the primary container (e.g., vacuum flask) to equal 10% of the maximum collection volume. Maximum collection volume should be no more than two-thirds full.
- After adding bleach to the maximum volume (two-thirds of the container), allow a minimum contact time of 20 minutes.
- Use pH paper to verify that pH is between 5.5 and 10. Adjust pH if necessary.
- Empty the flask into the laboratory sink with copious amounts of water.

Because the local sewer authority sets limitations on drain disposal of chemicals, EHS requires that PI/LS seek written approval from EHS regarding the use of any chemical disinfectants other than 10% bleach. Disinfectant solutions not authorized for sewer disposal may be collected as hazardous waste.

10.9 Biological/Radioactive Waste

Biological waste containing radioactive materials should be placed in a waste container labeled with the universal symbol for radiation and the name of the radioisotope the waste contains. EHS provides waste containers for radioactive waste. If the waste is biohazardous, the container must also be labeled with the universal biohazard symbol and the word "biohazard". Biohazardous liquid waste should be placed in containers filled to 10% of the final volume with bleach or other appropriate disinfectant. Dry or liquid biological/radioactive waste must never be autoclaved. Records for biological/radioactive waste disposal should be maintained as described in the *Radiation Safety Manual*. EHS should be contacted to pick up waste as needed.

10.10 Chemical Inactivation of Toxins

Follow these guidelines and Table 4 when using sodium hypochlorite or a mixture of sodium hypochlorite and sodium hydroxide to inactivating toxins:

- Work in a chemical fume hood lined with absorbent-backed paper and wear appropriate PPE.
- Use secondary containment.
- Put the toxin into solution and add an equal volume of deactivating chemical(s). Do not replace the cap on primary container.
- Place a "WARNING / DO NOT USE" sign on the chemical fume hood.
- Use appropriate exposure time.
- Dispose of and label inactivated toxin waste as hazardous waste as outlined in the *Laboratory Safety Manual.*
- Update appropriate electronic biological inventory.

11.0Laboratory Closeout or Renovation

Whether leaving and moving to a different institution, or relocating or renovating a laboratory within George Mason University, PI/LS are responsible for following proper closeout procedures. These procedures address material packaging and transfer, waste disposal, and laboratory and equipment decontamination.

11.1 Laboratory Closeout Procedures When Leaving George Mason University

When leaving George Mason University, it is important that PI/LS return the laboratory to a condition that will be immediately usable to the university. Material and equipment must either be transferred to a new institution or be properly decontaminated for reuse. In addition, wastes must be handled appropriately before final departure. PI/LS are responsible for the following when leaving George Mason University:

- Notify EHS 30 days prior to your confirmed move date, but no later than two weeks (14 days) in advance.
- Provide EHS with the following information within 14 days prior to leaving:
  - An itemized list of hazardous substances and equipment to be relocated.
  - The method of relocation (e.g., commercial mover, commercial shipping company, etc.).
  - An itemized list of equipment and instruments that will remain at the university.
  - An itemized list of materials and equipment that will be transferred to another George Mason University PI/LS and the name(s) of the PI/LS who will assume responsibility for these items.
- Prepare laboratory waste for disposal within the Satellite Accumulation Area and schedule waste pickup with EHS.
- Decontaminate and label equipment or instruments that will remain at the university with a *Decontamination Certificate* prior to departure.
- Provide EHS with the *Safety Records and Resources* binder including the *Laboratory Training Signature Page* and other research-related documentation (e.g., material/reagent transfer or purchase agreements, equipment decontamination certifications, etc.).
- Contact Office of Technology Transfer to prepare an MTA as necessary.
- Contact EHS to request personal exposure records.
- Remove all material, equipment, personal items, and reference materials from the laboratory that do not belong to George Mason University.

11.2 Laboratory Closeout Procedures When Switching Laboratories While at George Mason University

When moving to a new space or switching laboratories within George Mason University, it is important that PI/LS return their previous laboratory to a condition that will be immediately usable to the university. Material and equipment must either be properly disposed of or decontaminated and relocated to the new space. In addition, waste must be handled appropriately. PI/LS are responsible for the following when leaving George Mason University:

- Notify EHS 30 days prior to your confirmed move date, but no later than two weeks (14 days) in advance.
- Provide EHS with the following information within 14 days prior to your move date:

  - An itemized list of equipment and hazardous substances to be relocated.
  - Method of relocation (e.g., commercial mover coordinated by the Office of Space Management, department personnel, etc.) for both equipment and hazardous substances. Note that the transfer of hazardous substances between campuses is highly regulated by the DOT and must be conducted by a licensed contractor.
  - An itemized list of material and equipment that will remain in the laboratory and therefore will be transferred to another George Mason University PI/LS, and the name(s) of the PI/LS who will assume responsibility for these items.
  - An itemized list of materials and substances that may require temporary storage (less than 30 days).
- Prepare laboratory waste for disposal within the Satellite Accumulation Area and schedule waste pickup with EHS.
- Decontaminate and label equipment or instruments that will remain at the university with a *Decontamination Certificate* prior to departure.
- Relocate the *Safety Records and Resources* binder, MSDS binders, and safety manuals to wall bins located in the new space as part of the move process. EHS will replace these items as necessary in the space being vacated. If desired, contact EHS for new binders for binder contents (i.e., MSDS, training records, etc.) that will move with the lab to the new space.
- Remove all material, equipment, personal items, and reference materials from the laboratory.

11.3 Laboratory Renovation Procedures

EHS must approve renovations of existing laboratory spaces and alteration of a work space to or from a laboratory. PI/LS must observe the following procedures for laboratory renovation projects:

- Notify EHS as soon as possible but at least one month in advance.
- Consult with EHS regarding laboratory design.
- Provide EHS an itemized list of hazardous substances that will require temporary storage during the laboratory renovation project.
- Provide EHS an itemized list of hazardous substances and equipment that are proposed to be housed in the laboratory during renovation.
- Prepare laboratory waste for disposal and schedule waste pickup with EHS prior to commencement of renovation activities.
- Decontaminate equipment and surfaces prior to the commencement of renovation activities.

12.0 Spills and Accident Procedures

Laboratory personnel should be prepared to respond to a spill or accidental exposure involving the hazards and materials used in the laboratory. The *Supplemental Laboratory Safety Plan* completed by PI/LS should outline laboratory specific spill and accident response procedures.

EHS provides general spill procedure guidelines for biological spills below. In addition, the *Laboratory Safety Manual* provides procedures for handling chemical spills and other laboratory accidents. Contact EHS for spills involving a combination of biohazardous materials, hazardous chemicals, and/or radioactive materials. In this situation, the area should be cordoned off and signage posted.

Laboratory personnel are not required to respond to a spill. An individual who is uncomfortable responding to a spill should contact EHS. If a spill poses imminent danger to health and safety and cannot be isolated or contained, evacuate the area and contact University Police by dialing 911 and provide the following information:

- Name and telephone number of the caller.
- Location of the emergency (building name, room number, and building specific address, if known).
- Nature of the emergency (e.g., agent or material involved, fire, injuries).
- Special considerations (e.g., inhalation hazards present, potential for explosion, people trapped in rooms or buildings, number of people injured and type of injuries, electrical hazards, property damage, and access routes to the emergency).

12.1 Biological Spill Supplies

A spill kit is an essential safety item for biological laboratories. A basic biological spill kit should include:

- Disinfectant (that is most effective and appropriate for killing or inactivating the specific organisms stored and used in the particular laboratory).
- Spray bottle.
- Absorbent material (i.e., such as pads, sheets, spill socks, and paper towels).
- Red biohazard autoclave bags for the collection of contaminated items.
- Autoclave tape.
- Tongs.
- Sharps container.
- Boundary marking tape to cordon off the contaminated area until it is properly cleaned and disinfected.
- Warning sign.
- Spill supply inventory.

12.2 Spill Response

When a biological spill occurs, it is important to understand the potential routes of exposure for the material involved and to employ proper response procedures. The procedures below are

designed for spills involving biohazardous material. These procedures may be adapted for spills involving RG-1 agents or nonbiohazardous material, as these materials maybe harmful in large quantities or to individuals with a compromised immune system.

12.2.1 Laboratory Spills

1) If the biological material involved poses an inhalation hazard, stop breathing in order to avoid inhaling airborne material, and quickly leave the room.
2) Signal to others to leave, close door, and post a warning sign. No one should enter the laboratory for 30 minutes.
3) Go to a support space or adjacent laboratory. Avoid the hallway and publicly-accessed areas.
4) Remove contaminated PPE and clothing, turning exposed areas inward, and place in a biohazard bag.
5) If a personal exposure has occurred, follow procedures outlined in Section 12.3 and contact EHS or University Police to handle spill response.
6) Call 911 for medical assistance, when needed.
7) If the nature of the spill requires the use of a HEPA-filtered respirator, do not attempt to handle the spill. EHS will assume responsibility for the situation. If the microorganism does not pose an inhalation threat and you are qualified and comfortable cleaning up the spill, proceed to the next step.
8) Assemble spill supplies and use appropriate PPE including lab coat, gloves, and eye or face protection.
9) Cover the area of the spill with absorbent pads or towels, and carefully pour disinfectant around the spill. Because the volume of the spill will dilute the disinfectant, a concentrated disinfectant should be used. Allow at least a 20 minute contact time.
10) Pick up any visible sharp objects with tongs and discard in a sharps container.
11) Wipe surrounding areas (where the spill may have splashed) with disinfectant.
12) Disinfect contaminated laboratory equipment as needed.
13) Treat contaminated spill supplies and PPE as biohazardous waste.
14) Wash hands with antiseptic soap and warm water.
15) Students who have been exposed should report to Student Health Services. In the event that Student Health Services is closed, seek medical attention at the closest medical facility. Faculty/Staff should report to the nearest medical facility as listed on the *Supplemental Laboratory Safety Plan* associated with the lab where potential exposure occurred.
16) Notify the PI/LS and EHS of the incident.
17) Submit a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

12.2.2 Centrifuge Spills

1) If a centrifuge malfunctions while in operation or a tube breaks, turn the centrifuge off immediately and unplug it (if you can do so easily).
2) If you notice a spill has occurred after opening the centrifuge lid, stop breathing in order to avoid inhaling airborne material and close the centrifuge to allow aerosols to settle.

3) Leave the laboratory and signal for others to leave the laboratory.
4) Go to a support space or adjacent laboratory. Avoid the hallway and publicly-accessed areas.
5) Remove contaminated PPE and clothing, turning exposed areas inward, and place in a biohazard bag.
6) If a personal exposure has occurred, follow procedures outlined in Section 12.3 and contact EHS or University Police to handle spill response.
7) Call 911 for medical assistance, when needed.
8) If the nature of the spill requires the use of a HEPA filtered respirator, do not attempt to handle the spill. EHS will assume responsibility for the situation. If the microorganism does not pose an inhalation threat and you are qualified and comfortable cleaning up the spill, proceed to the next step.
9) Assemble spill supplies and use appropriate PPE including lab coat, gloves, and eye or face protection.
10) Remove rotor and place it in the biosafety cabinet. Open rotor, remove tubes using tongs or forceps. Disinfect the rotor with an appropriate chemical disinfectant and contact time. Dry the rotor thoroughly after disinfection.
11) Cover the bottom of the centrifuge with disinfectant-soaked towels. Concentrated disinfectant should be used. Allow at least a 20-minute contact time.
12) Wipe the inside of the centrifuge and the lid with an appropriate disinfectant. Dry the inside of the centrifuge thoroughly.
13) Treat contaminated spill supplies and PPE as biohazardous waste.
14) Wash hands with antiseptic soap and warm water.
15) Students who have been exposed should report to Student Health Services. In the event that Student Health Services is closed, seek medical attention at the closest medical facility. Faculty/Staff should report to the nearest medical facility as listed on the *Supplemental Laboratory Safety Plan* associated with the lab where potential exposure occurred.
16) Notify the PI/LS and EHS of the incident.
17) Submit a *First Report of Accident Form* to the Workers' Compensation department of Human Resources and Payroll.

12.3 Personal Exposure

In the event of a personal exposure, an individual's primary concern must be to minimize the degree of exposure and the possible effects. The emergency procedures employed depend on the type of biohazardous material to which the individual was exposed and the extent of exposure. Each laboratory where biological materials are used or stored should have a *Supplemental Laboratory Safety Plan* that outlines specific procedures to be followed in the event of an exposure to biohazards present in their laboratory. Immediate emergency response procedures for inhalation or skin exposure incidents are provided below.

In general, laboratory personnel who have experienced an exposure should immediately: a) decontaminate themselves; b) Call 911 for medical assistance or report to the closest medical facility, when needed; c) notify the PI/LS and EHS; and d) submit a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

Medical care as a result of work-related exposure may be provided at no cost to the employee and is dependent on the type of exposure. Facilities closest to the Fairfax and Prince William campuses are listed below.

Prince William Hospital	**INOVA Fairfax Hospital**	**INOVA Emergency Care Center**
8700 Sudley Road	3300 Gallows Road	4315 Chain Bridge Road
Manassas, VA	Falls Church, VA	Fairfax, VA
(703) 369-8000	(703) 776-4002	(703) 877-8200

12.3.1 Inhalation Exposure

Follow the steps below when there is a potential for inhalation exposure:

1) Stop breathing in order to avoid inhaling airborne material, and quickly leave the room.
2) Signal to others to leave, close the door, and post a warning sign. No one should enter the laboratory for at least 30 minutes.
3) Go to a support space or adjacent laboratory. Avoid the hallway and publicly-accessed areas.
4) Remove contaminated PPE and clothing, turning exposed areas inward, and place in a biohazard bag.
5) Wash all exposed skin and hands with antiseptic soap and warm water for 15 minutes. Wash gently so as not to break the skin.
6) Call 911 for medical assistance or report to the closest medical facility, when needed.
7) Immediately notify PI/LS and EHS. Notify EHS who must clear the laboratory for re-entry. If EHS is not available or it is after normal business hours, contact University Police.
8) Complete and submit a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

12.3.2 Skin or Mucous Membrane Exposure

Skin or mucous membrane exposure can occur through splashes to the eye, face, exposed skin, or clothing; by touching mucous membranes with contaminated hands; or from a needlestick, puncture with a contaminated sharp object, an animal scratch or bite, or through wounds, abrasions, and eczema. In the event of a skin or mucous membrane exposure:

1) Remove contaminated PPE and clothing, turning exposed areas inward, and place in a biohazard bag.
2) For mucous membrane exposure, flush the affected area with the eyewash for at least 15 minutes.
3) For skin exposure, wash the affected skin with antiseptic soap and warm water for at least 15 minutes. Wash gently so as not to break the skin.
4) Call 911 for medical assistance or report to the closest medical facility, when needed. Immediately notify PI/LS and EHS. Notify EHS who must clear the laboratory for re-entry. If EHS is not available or it is after normal business hours, contact University Police.

5) Immediately notify PI/LS and EHS. Notify EHS who must clear the laboratory for re-entry. If EHS is not available or it is after normal business hours, contact University Police.
6) Complete and submit a *First Report of Accident Form* to the Workers' Compensation Department within Human Resources and Payroll.

Appendix A
Definitions

Acrylamide: (C_3H_5NO) A white odorless crystalline solid used to make electrophoresis gels; a severe neurotoxin and possible human carcinogen.

Administrative controls: Work procedures, such as written safety policies, rules, supervision, and training, with the goal of reducing the duration, frequency, and severity of exposure to hazardous materials or situations.

Aerosol: Solid particles or liquid droplets, ranging in diameter from 0.01 to a few microns, suspended in a gaseous medium (e.g., air).

Affiliate: A person who is not a George Mason University employee, faculty, staff, or student but is participating in laboratory activities in facilities owned or under the control of George Mason University. This includes volunteers, visiting faculty, and visiting research associates.

Animal biosafety level (ABSL): Biosafety levels established by the Centers for Disease Control and the National Institutes of Health in *Biosafety in Microbiological and Biomedical Laboratories (BMBL)* for work involving biological agents in animal studies (see biosafety level).

Biohazardous material: All infectious agents, vectors known to carry and transmit infectious agents, infected or potentially-infected animals, infectious material, recombinant DNA, and biologically-derived toxins that present either a risk or a potential risk to the health of humans, animals, or plants either directly through infection or indirectly through damage to the environment.

Biological inventory: List of all biological materials present, used, or stored in the laboratory.

Biological material: As used in this manual, a general term referring to all prokaryotic and eukaryotic organisms (and their components), viruses, subviral agents, recombinant DNA, and biologically-derived toxins used in research and instructional laboratories.

Biological safety: (see *Biosafety*).

Biologically-derived toxin: All molecules produced by animals, plants, microorganisms or other agents that have an LD_{50} value of <50 mg/kg when administered orally to rates.

Biosafety: A concept that promotes safe laboratory practices, procedures, and proper use of containment equipment and facilities by laboratory personnel in the research and instructional laboratory environment. The purpose of a biological safety program is to prevent laboratory-acquired infections.

Biosafety cabinet: A devise enclosed (except for necessary exhaust purposes) on three sides and top and bottom, designed to draw air inward by means of mechanical ventilation, operated with insertion of only the hands and arms of the user, and in which virulent pathogens are used.

Biosafety level (BSL): Classification system established by the CDC and NIH in *BMBL* for work involving biological materials; four levels (BSL1-4) provide combinations of laboratory practices and techniques, safety equipment, and laboratory facilities appropriate to minimize the risk of exposure to infectious agents. The NIH *Guidelines for Research Involving Recombinant DNA Molecules* also makes use of this classification system in its requirements for safety practices regarding laboratory activities involving organisms that contain recombinant DNA.

Biosecurity: Protection of microbial agents from loss, theft, diversion, or intentional misuse. The objective of a biosecurity program is to develop and implement practices and procedures that prevent the loss, theft, or misuse of microorganisms, biological materials, and research related information.

Bloodborne pathogens (as defined by OSHA): Pathogenic microorganisms that are present in human blood and can cause disease in humans. These pathogens include, but are not limited to, HBV and HIV.

Carcinogen (as defined by OSHA in 29 CFR 1910.1450): Any substance which meets one of the following criteria: (i) It is regulated by OSHA as a carcinogen; or (ii) it is listed under the category, "known to be carcinogens," in the Annual Report on Carcinogens published by the National Toxicology Program (NTP) (latest edition); or (iii) it is listed under Group 1 ("carcinogenic to humans") by the International Agency for Research on Cancer Monographs (IARC) (latest editions); or (iv) it is listed in either Group 2A or 2B by IARC or under the category "reasonably anticipated to be carcinogens" by NTP, and causes statistically significant tumor incidence in experimental animals in accordance with any of the following criteria: (A) After inhalation exposure of 6-7 hours per day, 5 days per week, for a significant portion of a lifetime to dosages of less than 10 mg/m(3); (B) After repeated skin application of less than 300 (mg/kg of body weight) per week; or (C) After oral dosages of less than 50 mg/kg of body weight per day.

Chemical fume hood (as defined by OSHA in 29 CFR 1910.1450): Device located in a laboratory, enclosed on five sides with a movable sash or fixed partial enclosed on the remaining side; constructed and maintained to draw air from the laboratory and to prevent or minimize the escape of air contaminants into the laboratory; and allows chemical manipulations to be conducted in the enclosure without insertion of any portion of the individual's body other than hands and arms.

Chemical waste: Solid or liquid laboratory waste containing chemicals that must be disposed of through George Mason University's chemical waste management program.

Code of Federal Regulations (CFR): The codification of the general and permanent rules and regulations published in the Federal Register by the executive departments and agencies of the Federal Government.

Contact time: Length of time that a chemical disinfectant must be in contact with a surface or instrument in order to decontaminate that instrument or surface.

Containment (as defined in the BMBL): Safe methods for managing infectious materials in the laboratory environment where they are being handled or maintained. The purpose of containment is to reduce or eliminate exposure of laboratory workers, other persons, and the outside environment to potentially hazardous agents.

Corrosive: Having a pH less than 2 or greater than 12.5 or the ability to destroy human tissue upon contact.

Decontamination: Process by which contaminated surfaces, equipment, instruments, or waste are rendered nonhazardous. Physical and chemical means of decontamination include the use of heat, liquid decontaminants, and gasses.

Disinfection: Use of antimicrobial substances to destroy or suppress the growth of microorganisms such as bacteria, viruses, or fungi.

Employee**:** A person who works for the university full-time or part-time and is paid through the university's payroll system or receives compensation in any form.

Engineering controls: Controls that eliminate or reduce exposure to laboratory hazards through the use or substitution of engineered machinery or equipment. Examples include self-capping syringe needles, ventilations systems such as a chemical fume hood, sound-dampening materials to reduce noise levels, safety interlocks, and radiation shielding.

Ethidium bromide ($C_{21}H_{20}BrN_3$): An intercalating agent capable of fluorescing red-orange when exposed to ultraviolet light; commonly used as a nucleic acid stain in gel electrophoresis; potent mutagen.

Etiologic agent: A biological agent or its toxin known to be the causative agent of human disease.

Exempt recombinant DNA: Recombinant DNA molecules exempt from requirements set forth by the NIH *Guidelines for Research Involving Recombinant DNA Molecules.*

Exposure incident: A blood/body fluid exposure incident occurs when blood or other potentially infectious material enters the body via one of the following routes: inhalation, a percutaneous injury (e.g., a needlestick or cut with a sharp object), contact with mucous membranes (eyes, nose, mouth) contact with nonintact skin (especially when the exposed skin is chapped, abraded, or afflicted with dermatitis, or the contact is prolonged or involving an extensive area).

Faculty: An employee who is appointed as a member of the instructional, research, or administrative faculty, including visiting faculty and post-doctoral fellows.

Hazardous chemical (as defined in 29 CFR 1910.1450): A chemical for which there is statistically significant evidence based on at least one study conducted in accordance with established scientific principles that acute or chronic health effects may occur in exposed employees. The term "health hazard" includes chemicals which are carcinogens, toxic or highly toxic agents, reproductive toxins, irritants, corrosives, sensitizers, hepatotoxins, nephrotoxins, neurotoxins, agents which act on the hematopoietic systems, and agents which damage the lungs, skin, eyes, or mucous membranes.

Hazardous substance: Any material that may present a danger to human health and welfare or the environment. This includes hazardous chemicals biohazardous materials, and sources of ionizing radiation.

Hazardous waste: A waste with properties that make it dangerous or potentially harmful to human health or the environment and exhibits at least one of four characteristics: ignitability, corrosivity, reactivity, or toxicity.

Heavy metal: A metal whose specific gravity is approximately 5.0 or higher (e.g., arsenic, barium, cadmium, chromium, lead, mercury, selenium, silver).

High Efficiency Particulate Air (HEPA) filtration: Filtration of air through filters that have an efficiency of 99.97% for particles with a diameter greater or equal to of 0.3 microns.

Immunocompromised: A state in which the immune system's ability to fight infectious disease is compromised or entirely absent. Most cases of immunodeficiency are either congenital or acquired. An immunocompromised person is very vulnerable to opportunistic infections.

Infectious agents: All human, animal, and plant pathogens (bacteria, parasites, fungi, viruses, prions).

Infectious material: Infectious agents and all biological material that contains or has the potential to contain infectious agents. Examples of infectious material include all human or NHP materials (e.g., blood and other body fluids, organs, tissues, cultured cells), infected animals and material from infected animals, and environmental samples likely to contain infectious agents.

Instructional laboratory: Facility located on George Mason University property that meets the requirements for a laboratory set forth in 29 CFR 1919.1450 and where academic laboratory courses are conducted.

Laboratory exposure: Occupational exposure to an infectious material or other hazardous material that takes place in the laboratory.

Laboratory personnel: Faculty (professional, administrative, post-doctoral, and research), staff (classified, wage, and student wage), affiliates (visiting faculty, volunteers, visiting research associates), and students (graduate students, undergraduate students, laboratory assistants, etc.) working in laboratories and laboratory support areas. This term does not refer to students enrolled in instructional laboratory courses.

Laboratory support room: Space auxiliary to a laboratory that is used by laboratory personnel to prepare reagents or store materials for their laboratory.

Lethal dose 50 (LD_{50}): Quantity of material than when ingested, injected, or applied to the skin as a single dose will cause death to 50% of test animals who are exposed to it. The test conditions should be specified; the value is expressed in g/kg or mg/kg of body weight.

Material Safety Data Sheet (MSDS): A standard formatted information sheet prepared by a material manufacturer, describing the potential hazards, physical properties, and procedures for safe use of a material.

MSDS Library: Binder present in each laboratory that contains MSDS for each chemical and biohazardous material present in the laboratory.

Mutagen: Agent giving rise to an increased occurrence of mutation in populations of cells and/or organisms.

Nonbiohazardous material: Material that is not normally infectious, including nonpathogenic microorganisms, viruses, and subviral agents; plants and nonprimate animals (except those listed as biohazardous material), biological material not likely to contain infectious agents, recombinant DNA molecules exempt from NIH Guidelines, environmental samples not likely to contain infectious agents, and biologically-derived nontoxic molecules.

Nonexempt recombinant DNA: Recombinant DNA molecules subject to requirements set forth by the NIH *Guidelines for Research Involving Recombinant DNA Molecules.*

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

Pathogenic: Capable of causing disease.

Personal Protective Equipment (PPE): Clothing and other work accessories designed to create a barrier against workplace hazards. Examples include safety goggles, blast shields, hard hats, hearing protectors, gloves, respirators, aprons, and work boots.

Pledget: Small flat absorbent pad made from cotton or wool.

Polyacrylamide: An acrylate polymer formed from acrylamide subunits that are readily cross-linked. Polyacrylamide is not toxic, but unpolymerized acrylamide can be present in the polymerized acrylamide. Therefore, it is recommended to handle it with caution. It is highly water-absorbent, forming a soft gel used in such applications as polyacrylamide gel electrophoresis. It is also used as a thickener and suspending agent.

Primary containment: Use of appropriate safety equipment, microbiological techniques, and PPE to protect personnel and the immediate laboratory environment from an exposure.

Prion: *(*proteinaceous infectious particle) — Infectious protein structure that propagates through conversion of normal host proteins of the same type. Though the exact mechanisms of their actions and reproduction are unknown, it is now commonly accepted that prions are responsible for a number of previously known but little-understood diseases generally classified under transmissible spongiform encephalopathy diseases, including scrapie (a disease of sheep), kuru (found in members of the formerly cannibalistic Foré tribe in Papua New Guinea), Creutzfeldt-Jakob disease, Chronic Wasting Disease, Fatal Familial Insomnia, Gerstmann-Sträussler-Scheinker syndrome, and bovine spongiform encephalopathy (BSE or mad cow disease). Prions are highly resistant to common decontamination techniques.

Radioactive waste: Liquid and solid laboratory waste that contains radioactive material.

Recombinant DNA (as defined in the NIH document: Guidelines for Research Involving Recombinant DNA): Molecules that (i) are constructed outside living cells by joining natural or synthetic DNA segments to DNA molecules that can replicate in a living cell, or (ii) result from the replication of those described in (i) above.

Regulated medical waste (RMW) (as defined in 9VAC20-120-150): Includes (1) discarded cultures, stocks, specimens, vaccines, and associated items likely to contain agents that are likely to be pathogenic to healthy humans; discarded etiologic agents; wastes from the production of biological materials; and antibiotics likely to have been contaminated by agents that are likely to be pathogenic to healthy humans; (2) wastes consisting of human blood or human body fluids or items contaminated with human blood or human body fluids; all human anatomical wastes and all wastes that are human tissues, organs, or body parts; (3) sharps likely to be contaminated with agents that are pathogenic to healthy humans; all needles, syringes with attached needles, suture needles, and scalpels, including all sharps generated through veterinary practice; (4) animal carcasses, body parts, bedding, and all other wastes likely to have been contaminated as a result of animals being intentionally infected with agents likely to be pathogenic to healthy humans; (5) any residue or contaminated soil, water, or other debris resulting from the cleanup of a spill of any RMW; (6) any solid waste contaminated by or mixed with RMW.

Reproductive hazard: A material (chemical, agent, or toxin) that has the potential to affect reproductive capabilities or to cause damage to the unborn embryo/fetus.

Research laboratory: Facility located on George Mason University property that meets the requirements for a laboratory set forth in 29 CFR 1910.1450 and where scientific research is conducted.

Restricted area: Area that contains unique hazards (e.g., animal rooms, hazardous waste storage) and therefore requires more stringent access restrictions than other laboratories or laboratory support rooms.

Risk Group (RG): Categories for biological agents (including unknown samples and environmental samples) that classify agents based on their relative degree of pathogenicity in healthy, human adults, mode of transmission and host range, availability of preventative

measures and the availability of effective treatment; RG take into account the risk posed to laboratory personnel and the community.

Secondary containment: Use of facility design and operational practices, such as restricted access, ventilation, directional airflow and air treatment systems to protect the protection of the environment external to the laboratory from exposure.

Staff: A part-time or full-time employee who is not a member of the faculty or a student.

Select Agent: Biological agent or toxin that could pose a severe threat to public health and safety; to animal or plant health; or to animal or plant products and are therefore covered under the Select Agent Rule (7 CFR Part 331, 9 CFR Part 121, and 42 CFR Part 73).

Sterilization: Use of mechanical or chemical means to inactivate all forms of microbial life.

Student: A person who is officially enrolled in a course or program of study offered by the university.

Subviral agent: Infectious particle, such as a prion, that is capable of infecting and causing disease in a living organism.

Visitor: A person that is not an employee, faculty, staff, or student but is participating in laboratory activities in facilities owned or under the control of George Mason University.

Appendix B
Supplemental Laboratory Safety Plan

Supplemental Laboratory Safety Plan
George Mason University

Under 29 CFR 1910.1450, George Mason University is required to provide a *Chemical Hygiene Plan* that establishes minimum safety standards for working with chemicals in the laboratory and outlines procedures that minimize both the risk of chemical exposure to laboratory personnel and the risk of chemical releases into the environment. Additionally, under 29 CFR 1910.1030, the university is required to establish an *Exposure Control Plan* designed to minimize or eliminate the risk of exposure to infectious materials which is available on the Environmental Health and Safety (EHS) webpage, ehs.gmu.edu. The *Laboratory Safety Manual* serves as the Chemical Hygiene Plan for George Mason University, and the *Biological Safety Manual* outlines safety practices to minimize the risk of exposure to infectious or potentially infectious materials in laboratories.

The *Supplemental Laboratory Safety Plan* is a supplement to the *Laboratory Safety Manual* and *Biological Safety Manual* that provides laboratory-specific information for responding to health and safety issues and laboratory emergencies. The plan, which must be completed for all laboratories, must list the types of hazards present in the laboratory and outline laboratory-specific engineering and administrative controls, personal protective equipment (PPE), operational procedures (e.g., decontamination, waste handling), and procedures for spill or exposure response. The *Supplemental Laboratory Safety Plan* must be kept in the *Safety Records and Resources* binder located within the laboratory where it is readily available to laboratory personnel and must be routinely reviewed and updated to reflect current laboratory activities. A copy should also be on file with EHS. The template for completing this plan is available on the EHS website (ehs.gmu.edu). Laboratory personnel must be trained on the contents of this plan and must verify receipt of training by signing the *Laboratory Training Signature* page, also located within the *Safety Records and Resources* binder and on the EHS website (ehs.gmu.edu).

General Information

Date modified:

Principal Investigator or Laboratory Supervisor: _____

Unit or Department: _____

Office Location: _____ **Office Phone: _____**

Lab Location: _____ **Lab Phone: _____**

Email Address: _____

Emergency Response Procedures

1. Emergency Contact Information

University Police:	911 from a university phone or (703) 993-2810
Environmental Health and Safety:	703-993-8448
PI/LS after hours contact #:	_____
Other: _____	_____
Other: _____	_____
Other: _____	_____

2. Local Medical Care Facilities

Fairfax Campus:

1) Inova Emergency Care Center
4315 Chain Bridge Road, Fairfax, VA 22030
703-877-8200

2) Inova Fairfax Hospital
3300 Gallows Road, Falls Church, VA 22042
703-776-4002

Prince William Campus:

Prince William Hospital Emergency Room
8700 Sudley Road, Manassas, VA 20110
(703)-369-8000

3. Emergency Equipment available in or near the laboratory

Eye wash location: _____	Flushed every two weeks by: _____
Emergency shower location: _____	Flushed every two weeks by: _____
Fire extinguisher location: _____	Fire extinguisher type: _____
Spill supplies location: _____	First aid kit location: _____
Other: _____	Other: _____

4. Emergency Notification

- Contact University Police by dialing 911 from any university phone or (703) 993-2810.
- Provide the following information:
  - Name and telephone number of the caller.
  - Location of the emergency (building name, room number, and building specific address, if known).
  - Nature of the emergency (e.g., chemical spill and chemical(s) involved, fire, injuries).

Special considerations (e.g., the potential for explosion, acutely hazardous gases present, people trapped in rooms or buildings, number of people injured and type of injuries, electrical hazards, property damage and access routes to the emergency).

5. Evacuation Procedure (Follow these steps, if safe to do so.)

1. Notify other laboratory personnel.
2. If conditions permit, cap and secure open vials, bottles, and other materials and turn off laboratory equipment.
3. Leave the laboratory and close the door.
4. Activate the fire alarm to evacuate the building.
5. If it is safe to do so, assist anyone who may be in danger. Otherwise notify emergency response personnel once you have evacuated the building.
6. Exit the building according to the Building Evacuation Plan in a calm manner using the closest available emergency exit. Never use elevators.
7. Congregate at the pre-designated assembly point for the building.

6. Laboratory Fire (Personnel are not required to fight fires and should evacuate the building immediately in the event of a fire.)

1. Notify other laboratory personnel.
2. If conditions permit, cap and secure open vials, bottles, and other materials and turn off laboratory equipment.
3. Leave the laboratory and close the door.
4. Activate the fire alarm to evacuate the building.
5. If it is safe to do so, assist anyone who may be in danger. Otherwise notify emergency response personnel once

you have evacuated the building.

6. Notify University Police or emergency response personnel that you have specific information regarding the fire.
7. Fight a fire with a fire extinguisher **ONLY IF**:
    a. You have been trained in the proper use of a fire extinguisher and are confident in your abilities to cope with the hazards of the fire.
    b. The fire is a small, incipient fire (no larger than a waste basket).
    c. Terminate fire fighting efforts when it becomes obvious that there is a danger from smoke, heat, or flames.

7. Gas Leaks

Situations involving uncontrollable leaking gas from a cylinder should be considered extremely hazardous and warrant immediate evacuation of the building. If the gas leak is minimal, innocuous, and safely within reach, the cylinder valve should be closed. Otherwise leave the area, call University Police by dialing 911 from a campus phone or 703-993-2810, and activate the fire alarm to evacuate the building.

8. Equipment Failures

Equipment failures can result from power failure, defects, or malfunctions. If a piece of equipment fails while in use, take steps to contain or control possible exposures to the substances being used. It is inappropriate to continue use of hazardous substances and equipment during a power failure or equipment malfunction. In the event of a power failure, all personnel must secure the materials they are working with, turn off equipment, and leave the laboratory until power is restored.

9. Ventilation Failure

If laboratory building ventilation fails, all operations concerning chemicals within that laboratory or building must be discontinued. Laboratory operations may resume in the laboratory or building once ventilation has been restored and it is confirmed that all ventilation systems are operating correctly. Chemical Fume hoods that have failed can not be used until they are repaired and re-tested.

10. Other: List other probable emergencies for your laboratory and appropriate emergency response for laboratory personnel.

Exposure Response

11. Exposure Response–Skin or Mucous Membrane

In the event of a personal exposure, an individual's primary concern must be to minimize the degree of exposure and the possible effects. Skin or mucous membrane exposure can occur through splashes to the eye, face, exposed skin, or clothing; by touching mucous membranes with contaminated hands; or from a needlestick, puncture with a contaminated sharp object, an animal scratch or bite, or through wounds, abrasions, and eczema. **A general exposure response is provided below. This response may not be adequate for all materials present in the laboratory. Please provide additional exposure response procedures, as necessary, for chemicals and biological agents that require a specific exposure response.**

Chemical or Biological Agent:	**Exposure Response:**
General	1. Remove contaminated PPE and clothing, turning exposed areas inward and place in a bag. Dispose as laboratory waste. 2. Notify other laboratory personnel of the incident and of any surface or equipment decontamination that needs to be done. 3. For mucous membrane exposure, flush the affected area with the eyewash for at least 15 minutes. 4. For skin exposure, wash affected skin with soap and cold water for at least 15 minutes. Cold water has the effect of closing the skins pores thereby slowing the rate of absorption into the body. Wash gently so as not to break the skin. For skin exposures not limited to the hands and forearms, the emergency shower should be used. Apply first aid as needed. 5. Call 911 for emergency medical assistance or seek medical attention at the closest medical facility listed above. 6. Report all possible exposure incidents to PI/LS and EHS. 7. Complete and submit a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

12. Exposure Response-Inhalation

Inhalation exposure can occur when working with volatile chemicals in a poorly ventilated area or as the result of inhaling airborne substances aerosolized by laboratory procedures such as centrifugation or vortexing. **A general exposure response is provided below. This response may not be adequate for all materials present in the laboratory. Please provide additional exposure response procedures, as necessary, for chemicals and biological agents that require a specific exposure response.**

Chemical or Biological Agent:	**Exposure Response:**
General	1. Stop breathing in order to avoid inhaling airborne substances and quickly leave the room. 2. Signal to others to leave, close the door, and post a warning sign. 3. Leave the area immediately and seek fresh air. 4. Remove contaminated PPE and clothing, turning exposed areas inward and place in a polyethylene bag. 5. Review the Material Safety Data Sheets (MSDS) for the chemical involved to evaluate exposure data. 6. Call 911 for emergency medical assistance or seek medical attention at the closest medical facility listed above. 7. Report all possible exposure incidents to PI/LS and EHS. 8. Notify EHS who must clear the laboratory for re-entry. If EHS is not available or it is after normal business hours, contact University Police. 9. Complete and submit a *First Report of Accident Form* to the Workers' Compensation

department within Human Resources and Payroll.

13. Exposure Response–Ingestion

Accidental ingestion may occur as a result of splashes to the face, touching the face with contaminated hands, eating, drinking, or applying cosmetics in the laboratory, or through the out-dated and unacceptable practice of mouth pipetting. **A general exposure response is provided below. This response may not be adequate for all materials present in the laboratory. Please provide additional exposure response procedures, as necessary, for chemicals and biological agents that require a specific exposure response.**

Chemical or Biological Agent:	**Exposure Response:**
General	In the event of accidental ingestion of a chemical, seek medical attention (dial 911 or the Poison Control Center at 800-962-1253). Do not induce vomiting unless directed to do so by a health care provider. Report all possible exposure incidents to PI/LS and EHS and submit a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

Spill Response

14. Spill Response

Laboratory personnel are not required to respond to a spill. If you are uncomfortable in responding to a spill, if a spill poses imminent danger to health and safety or cannot be isolated, contained or controlled, move to a safe area and contact University Police. Do not attempt to clean the spill.

Spill Supplies Available in the Laboratory

☐ Chemical Spill Kit containing absorbent material (pads, sheets, spill socks, and paper towels), nitrile gloves, polyethylene bags, boundary marking tape, warning sign, spill supply inventory, and 5-gallon pail with screw top lid.

☐ Biological Spill Kit containing disinfectant (that is most effective and appropriate for killing or inactivating the specific organisms stored and used in the particular laboratory), spray bottle, absorbent material (e.g., sheets, spill socks, and paper towels), red biohazard autoclave bags for the collection of contaminated items, autoclave tape, tongs, sharps container, boundary marking tape to cordon off the contaminated area until it is properly cleaned and disinfected, warning sign, and spill supply inventory.

☐ Other Absorbent:

☐ Acid Neutralizer

☐ Caustic Neutralizer

☐ Other:

15. Spill Response-Chemical Spills

A general spill response is provided below. This response may not be adequate for all chemicals present in the laboratory. Please provide additional spill response procedures, as necessary, for chemicals that require a specific spill response.

Chemical or Biological Agent:	**Spill Response:**
General	1. Contact University Police for any spill that: • poses an inhalation hazard. • cannot be isolated, contained, or controlled quickly. • poses imminent danger to health and safety. • poses imminent danger to property or the environment. • you are uncomfortable responding to on your own. 2. Signal to others to leave, close the door, and post a warning sign. 3. Go to a support space or adjacent laboratory. Avoid the hallway and publicly accessed areas. 4. Remove contaminated PPE and clothing, turning exposed areas inward and place in a polyethylene bag. 5. If a personal exposure has occurred or you experience symptoms of exposure, follow exposure procedures in this plan and contact University Police. 6. Call 911 for emergency medical assistance or seek medical attention at the closes medical facility listed above. 7. If you can safely proceed in cleaning the spill, notify other laboratory personnel and consult the MSDS regarding the physical, chemical, and toxicological properties and hazards of the chemical to determine the appropriate response. 8. Do not attempt to clean a spill alone. Employ the assistance of a co-worker to facilitate cleanup activities. 9. Assemble spill supplies and use appropriate PPE including lab coat, gloves, and eye or face protection. 10. Take steps to limit the impact of the spill by preventing spilled substances from reaching drains and by isolating equipment and materials that may escalate the danger of the situation. 11. Contain the spill with absorbent materials. 12. Pick up any visible sharp objects with tongs and discard into a sharps container. 13. Clean the spill by working from the outer edges of the spill towards the center. 14. Clean surrounding areas (where the spill may have splashed). 15. Clean contaminated laboratory equipment as needed.

16. Place the waste generated from cleaning the spill and contaminated PPE in a polyethylene bag. Place the bag into a sturdy pail such as the one provided with the spill kit. Label the container with a Hazardous Waste label and place the waste in the satellite accumulation area. Sharps containers labeled with a biohazard symbol must be disposed of as biohazardous waste.
17. Wash hands with soap and warm water.
18. Report all possible exposure incidents to PI/LS and EHS and follow the exposure response outlined above.
19. Submit completed a *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

16. Spill Response–Biological Materials

When a biological spill occurs, it is important to understand the potential routes of exposure for the material involved and to employ proper response procedures. **A general spill response is provided below. For each infectious material in the laboratory, indicate the appropriate disinfectant, concentration and contact time required to clean the spill.**

1. If the biological material involved poses an inhalation hazard, stop breathing in order to avoid inhaling airborne material and quickly leave the room.
2. Signal to others to leave, close door, and post a warning sign. No one should enter the laboratory for 30 minutes.
3. Go to a support space or adjacent laboratory. Avoid the hallway and publicly accessed areas.
4. Remove contaminated PPE and clothing, turning exposed areas inward and place in a biohazard bag.
5. If a personal exposure has occurred, follow procedures outlined above and contact EHS or University Police to handle spill response.
6. Call 911 for medical assistance, when needed.
7. If the nature of the spill requires the use of a HEPA filtered respirator, do not attempt to handle the spill. EHS will assume responsibility for the situation. If the microorganism does not pose an inhalation threat and you are qualified and comfortable cleaning up the spill, proceed to the next step.
8. Assemble spill supplies and use appropriate PPE including lab coat, gloves, and eye or face protection.
9. Cover the area of the spill with disinfectant-soaked towels, and carefully pour disinfectant around the spill. Because the volume of the spill will dilute the disinfectant, a concentrated disinfectant should be used. Allow at least a 20-minute contact time.
10. Pick up any visible sharp objects with tongs and discard in a sharps container.
11. Wipe surrounding areas (where the spill may have splashed) with disinfectant.
12. Disinfect contaminated laboratory equipment as needed.
13. Treat contaminated spill supplies and PPE as biohazardous waste.
14. Wash hands with antiseptic soap and warm water.
15. Report all possible exposure incidents to PI/LS and EHS.
16. Notify EHS of the incident.
17. Submit completed *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

Infectious Material	Disinfectant	Concentration	Contact Time (min)

17. Spill Response-Centrifuge Spills

1. If a centrifuge malfunctions while in operation or a tube breaks, turn the centrifuge off immediately and unplug it (if you can do so easily).
2. If you notice a spill has occurred after opening the centrifuge lid, stop breathing in order to avoid inhaling airborne material and close the centrifuge to allow aerosols to settle.
3. Leave the laboratory and signal for others to leave the laboratory.
4. Go to a support space or adjacent laboratory. Avoid the hallway and publicly accessed areas.
5. Remove contaminated PPE and clothing, turning exposed areas inward and place in a biohazard bag.
6. If a personal exposure has occurred, follow procedures outlined above and contact University Police to handle spill response.

7. Call 911 for emergency medical assistance or seek medical attention at the closest medical facility listed above.
8. If the nature of the spill requires the use of a HEPA filtered respirator, do not attempt to handle the spill. EHS will assume responsibility for the situation. If the microorganism does not pose an inhalation threat and you are qualified and comfortable cleaning up the spill, proceed to the next step.
9. Assemble spill supplies and use appropriate PPE including lab coat, gloves, and eye or face protection.
10. Remove rotor and place it in the biosafety cabinet. Open rotor, remove tubes using tongs or forceps. Disinfect the rotor with an appropriate chemical disinfectant and contact time. Dry the rotor thoroughly after disinfection.
11. Cover the bottom of the centrifuge with disinfectant-soaked towels. Concentrated disinfectant should be used. Allow at least a 20-minute contact time.
12. Wipe the inside of the centrifuge and the lid with an appropriate disinfectant. Dry the inside of the centrifuge thoroughly.
13. Treat contaminated spill supplies and PPE as biohazardous waste.
14. Wash hands with antiseptic soap and warm water.
15. Report all possible exposure incidents to PI/LS and EHS.
16. Submit completed *First Report of Accident Form* to the Workers' Compensation department within Human Resources and Payroll.

Administrative Controls, Engineering Controls, and PPE

18. Administrative Controls
List any laboratory specific administrative controls in addition to those listed in the *Laboratory Safety Manual* and *Biological Safety Manual*

19. Safety and Compliance Bins Contain:

- ☒ Laboratory Safety Manual
- ☐ Biological Safety Manual
- ☐ Radiation Safety Manual
- ☐ Chemical Inventory
- ☐ Biological Inventory
- ☒ Laboratory Training Signature Page
- ☐ MSDS Library (For libraries too large to fit in the wall bins, list the location):

20. Facility Requirements
List any laboratory specific facility requirements in addition to those outlined in the *Laboratory Safety Manual* and *Biological Safety Manual* (example: hands-free sink).

Safety Equipment Available:

- ☐ Biosafety cabinet ☐ Not ducted ☐ Ducted
- ☐ Chemical fume hood
- ☐ Glove box
- ☐ Sealed lids for centrifuge rotors
- ☐ Safe needle devices
- ☐ Other:

21. Location of Designated Areas

Chemical Storage:
Satellite Accumulation Area:
Radiation Usage Areas:
Other:

22. Personal Protective Equipment (PPE)
Check each type of PPE available for use in the laboratory. Equipment should be inspected, cleaned, or replaced as needed.

- ☐ Disposable lab coat
- ☐ Laundered lab coat
- ☐ Chemical resistant apron
- ☐ Disposable shoe covers
- ☐ Disposable sleeves
- ☐ Hair covering
- ☐ Glove liners
- ☐ Utility/autoclave gloves
- ☐ Animal-handling gloves
- ☐ Disposable gloves
  - ☐ Powder-free
  - ☐ Latex-free
  - ☐ Chemical-resistant
- ☐ Safety goggles
- ☐ Safety glasses
- ☐ Face shield
- ☐ Hearing protection
- ☐ Respiratory Protection
- ☐ Other:
- ☐ Other:

Infectious Material

23. Infectious material includes infectious agents (bacteria, parasites, fungi, viruses, prions) and all biological material that contains or has the potential to contain infectious agents. Examples include human blood and blood components, human tissues and body fluids, cultured cells from human and non-human primates, infected animals and animal tissues, non-human primates and any tissues from non-human primates, tissues from sheep, and environmental samples likely to contain infectious agents. Check all materials present in the laboratory.

- ☐ Human blood or blood components
- ☐ Other human bodily fluids (list):
- ☐ Unfixed human tissues or organs
- ☐ Fixed human or animal brain/neural specimens
- ☐ Experimental animal blood, organs, or tissue
- ☐ Infectious materials listed on the Biological Inventory (primary and continuous cell lines, bacteria (including chlamydial and rickettsial agents), viruses, fungi, parasites, subviral agents, etc.)

24. Exposure Determination

The following job classifications are at risk for exposure to infectious material in this laboratory:

- ☐ Faculty (Professional, administrative, research)
- ☐ Staff (classified, wage, student wage)
- ☐ Visiting Faculty
- ☐ Volunteers
- ☐ Visiting Research Associates
- ☐ Other:
- ☐ Post doctoral Fellows
- ☐ Graduate Students
- ☐ Undergraduate students
- ☐ Students working for credit
- ☐ High School Students
- ☐ Other:

25. The following activities place individuals at risk for exposure to infectious material**:**

- Handling or manipulating samples containing infectious material or potentially infectious material
- Using equipment potentially contaminated with infectious material
- Performing maintenance on equipment, instruments, or machinery potentially contaminated with infectious material
- Responding to spills involving infectious material
- Handling waste potentially contaminated with infectious material
- Packaging infectious material for shipping or transport

26. Certain tasks and procedures increase the risk of parenteral exposure, inhalation exposure, or contact with mucous membranes. Check each of the following tasks or procedures performed by laboratory personnel:

- ☐ Use of sharps (needles, scalpels, blades, glass thermometers, pipettes, slides and coverslips)
- ☐ Injections or perfusions
- ☐ Use of french press, sonicator, homogenizer, or safety blender
- ☐ High speed centrifugation
- ☐ Dissection (human and non-human primate tissues and organs, any intentionally infected tissue or organ)
- ☐ Slicing tissue using a microtome or cryostat
- ☐ Pipetting, mixing, vortexing, or homogenization
- ☐ Handling infected animals and working in animal rooms containing infected animals

27. List other tasks, procedures, and activities that increase the exposure risk for laboratory personnel.

28. Laboratory Procedures

The *Biological Safety Manual* outlines general laboratory practices for work involving infectious materials, including a discussion of Universal Precautions to be followed when working with materials of human origin. List any additional laboratory specific practices and procedures in place for this laboratory (example: no sharps or glass permitted).

29. Decontamination Procedures

The *Biological Safety Manual* outlines general practices for decontamination and disinfection of infectious material. List the types of disinfectants used in the laboratory as well as the optimal concentration and contact time in number 16 above.

30. Waste handling procedures
The *Biological Safety Manual* outlines procedures for handling infectious waste. Additionally, EHS provides waste supplies (sharps containers, bags, burn boxes, etc.) upon request. Please provide laboratory specific waste handling information in number 16 above.:

Particularly Hazardous Substances

31. Particularly Hazardous Substances: List select carcinogens, acutely toxic chemicals, and reproductive toxins used in the laboratory and provide information on the storage and usage location, the type of containment devices used (e.g., chemical fume hood, glove box), the method used for decontamination, and specific waste handling procedures (e.g., location of waste receptacles). Provide information for each Particularly Hazardous Substance located in the laboratory.					
Chemical Name	Designated Areas		Containment Devises used	Decontamination Procedures	Specific Waste Handling Procedures (disposal of liquid waste, paper trash, PPE, and other contaminated materials)
	Storage	Usage			

Animal Handling

32. Animal Handling: Select details related to animal handling, including species, type of study, test substance, procedures, and locations of designated areas

Animal Species

Include approximate number housed.

- ☐ Guinea pig
- ☐ Mouse
- ☐ Non-Human Primates (NHP)
- ☐ Rabbit
- ☐ Rat
- ☐ Other

If multi-generational, number of generations in study

Type of Study

- ☐ Behavioral
- ☐ Sensitization
- ☐ Toxicological
- ☐ Other

Test Substance

Name of Test Substance:

Purity:

Concentration:

Method of Administration

- ☐ Oral feed
- ☐ Oral gavage
- ☐ Injection
- ☐ Dermal absorption
- ☐ Aerosolization

Procedures

Check all that apply.

- ☐ Breeding
- ☐ Tail bleeds
- ☐ Oral gavage
- ☐ Cannula
- ☐ Surgery, type
- ☐ Anesthetization, method
- ☐ Euthanization, method

Location of Designated Areas

Bedding

Storage location:

Type of bedding:

Feed

Storage location:

Type of feed:

Administrative Controls, Engineering Controls, and PPE Related to Animal Handling

Administrative Controls
List any facility specific administrative controls in addition to those listed in the *Laboratory Safety Manual* and *Biological Safety Manual.*

☒Vermin control program
☐Other:

Facility Requirements
Specific facility requirements in addition to those outlined in the *Laboratory Safety Manual* and *Biological Safety Manual* (example: hands-free sink).

☒Doors are self-closing and locking.
☒Doors open inward.
☒Walls, floor, and ceilings are water resistant and designed to facilitate cleaning and housekeeping.
☒Penetrations in walls, floor, and ceilings are sealed, to include openings around ducts, doors, and door frames, to facilitate pest control and proper cleaning.
☒Ventilation is provided in accordance with the *Guide for Care and Use of Laboratory Animals*.
☒Heat and humidity is adjustable to accommodate a range of animal species.

Safety Equipment
Check each type of safety equipment available for use in the facility.

☐Biosafety cabinet ☐Not ducted ☐Ducted
☐Chemical fume hood
☐Glove box
☐Downdraft table
☐Safe needle devices
☐Cage Wash
☐Bedding station
☐Other:

Required Personal Protective Equipment (PPE)

☒Disposable or laundered lab coat
☒Disposable shoe covers
☒N95 respirator when completing bedding or cage changes
☒Disposable coveralls or laundered scrubs when completing bedding or cage changes
☒Disposable gloves (latex-free)
☒Safety glasses
☒Hair covering when completing bedding or cage changes

Additional Personal Protective Equipment (PPE)
Check each type of PPE available for use in the facility. Equipment should be inspected, cleaned, or replaced as needed.

☐Disposable sleeves
☐Utility/autoclave gloves
☐Animal-handling gloves
☐Safety goggles
☐Face shield
☐Hearing protection
☐N95 respirator
☐Other: