

ODU Biosafety Manual

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**Office of Research
&
Environmental Health and Safety**

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Purpose, Scope, and Responsibilities

PURPOSE

Old Dominion University has developed this Biosafety Manual to provide guidance to all faculty, staff, and students on the safe handling of biological materials and the proper safety practices to minimize the risk of exposure to potentially infectious materials. The practices and procedures listed in this manual will, when followed properly, reduce the likelihood of an occupational exposure to infectious agents, biological toxins, and/or recombinant/synthetic nucleic acid molecules. This manual is applicable to all faculty, staff, students, volunteers, and visitors at ODU who handle any of these types of materials. The successful implementation of this manual depends largely on those involved to be committed to maintaining a safe working environment and to be knowledgeable about laboratory safety.

The recommendations and requirements listed in this manual are derived from the Centers for Disease Control and Prevention (CDC) / National Institutes of Health (NIH) publication, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, 6th edition, the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines)*, and the Occupational Safety and Health Administration (OSHA) *Occupational Exposure to Bloodborne Pathogens* standard (29 CFR 1910.1030).

In the event of a regulatory or statutory change, then this manual should be construed to conform to that change. A Principal Investigator or any other concerned person should bring any change to the attention of the Institutional Biosafety Committee (IBC) and/or the Office of Research.

For further information, both the ODU Environmental Health and Safety Office and the Biosafety Officer are available to answer questions and to provide additional advice and training when necessary.

SCOPE

Instructional and research laboratories contain hazards that must be properly managed to minimize the risks they pose. These hazards include hazardous substances such as chemical, biological, and radioactive materials, as well as physical hazards associated with equipment and instruments within the laboratory. This manual only addresses those hazards associated with biological materials. All research and teaching conducted by faculty, staff, students, volunteers, and visitors at ODU involving any of the materials listed below must be approved by the ODU Institutional Biosafety Committee (IBC) prior to initiation:

- a. Microorganisms - bacteria, viruses, fungi, parasites
- b. Prions and other infectious agents
- c. Recombinant and synthetic nucleic acid molecules
- d. Animals infected with human pathogens (including human and non-human primate cells/tissues, recombinant or synthetic nucleic acid molecules) and non-human pathogens, as well as animals as sources of zoonotic diseases (e.g., Q fever associated with sheep, goats, and cattle).
- e. Plants work involving infectious agents and/or recombinant or synthetic nucleic acid molecules
- f. Unfixed human or animal source materials (tissues, cells, fluids)

- g. Bloodborne pathogens or other potentially infectious material (human blood, fluids, tissues, blood products, or cell lines; including non-human primate derived materials and established cell lines).
- h. Any **exempt** select agent or toxin below the **permissible toxin amount**.
Note: At the present time, ODU is not authorized to possess, use, or transfer select agents or toxins, as defined by the select agent regulations, 7 CFR 331 (agriculture), 9 CFR 121 (animals and animal products), and 42 CFR 73 (public health).
- i. Dual use research of concern (DURC) and Pathogens with Enhanced Pandemic Potential (PEPP)

Further information can be obtained on the [ODU Biohazards webpage](#).

RESPONSIBILITIES

Implementation of the Biosafety Program is the responsibility of each member of the Old Dominion University community. Principal Investigators bear primary responsibility for ensuring that their research complies with all applicable federal and state regulatory standards and ODU policies. All Principal Investigators shall be familiar with the regulations, policies, and guidelines applicable to their research. Failure to comply with established regulations and procedures is cause for disciplinary action. The following roles and responsibilities constitute an administrative framework in which safety is an essential and integral part of research.

Vice President for Research

The Vice President for Research (VPR) serves as the administrator responsible for oversight of the overall Biosafety Program. The VPR is the institutional officer responsible for ensuring institutional compliance with regulations and statutes. Specific responsibilities of the VPR are as follows:

- a. Appoint the members of the IBC, with the assistance of the Deans, and in accordance with this manual.
- b. Ensuring constructive communication among administrators, Deans, Principal Investigators, and others as a means to achieving safe teaching and research laboratories across campus.
- c. Conducting periodic review of federal and state regulations and guidelines along with the IBC, and amending this manual as needed.
- d. For all research subject to federal guidelines, the VPR shall promptly report to the NIH, or other sponsoring federal entity, within 30 days of:
 - i. Any research related accidents or illnesses
 - ii. Any significant programs or violations of the *NIH Guidelines* (e.g., failure to adhere to the containment and biosafety practices articulated in the *NIH Guidelines*)

Note: Certain types of accidents must be reported on a more expedited basis (e.g., spills or accidents involving recombinant DNA within a BSL-2 laboratory resulting in an overt exposure).

- e. Prepare and submit reports required by applicable guidelines.

- f. At a minimum, the VPR shall maintain research records for at least three years after completion of the research activity.
- g. When required, nominate two members who are not affiliated with the University (apart from their membership on the IBC) and who represent the interest of the surrounding community.

College Deans

The Dean of the College is responsible for ensuring that research conducted in his/her college complies with federal and state laws and guidelines. However, these responsibilities in no way diminish the primary responsibilities of the Principal Investigators. Specific responsibilities of the Deans are as follows:

- a. Ensure that their Principal Investigators are properly trained and equipped so as to be capable of compliance with this manual. The Dean shall remain reasonably familiar with the substance of the research in the College that is subject to this Procedure.
- b. Each year prior to July 1, shall nominate no fewer than four faculty members to the Vice President of Research to become members of the IBC. The Dean's nominees shall include faculty members who are qualified in plant and animal containment research.

Institutional Biosafety Committee (IBC)

The IBC is an advisory committee consisting of at least seven (7) members appointed by the Vice President for Research and is responsible for the oversight of biosafety at ODU. The members represent a collection of faculty, staff, and community members with a diversity of expertise and knowledge related to working with biohazardous materials.

This committee reviews all biosafety related research and teaching proposals to ensure compliance with federal, state, and university regulations and requirements. The committee assess each proposal to identify potential risks and ensure proper work practices and procedures are implemented to mitigate the risks. Specific responsibilities of the IBC are as follows:

- a. Develop policies and procedures which provide guidance for activities involving biological material to ensure they are handled and disposed of safely and properly.
- b. Ensure that all research and teaching projects involving the acquisition, use, storage, or disposal of biological materials, including biologically-derived toxins, are in compliance with all federal and state regulations and guidelines. Refer to Section III for more information regarding these regulations and guidelines
- c. Ensure all projects involving the acquisition, use, storage, or disposal of recombinant or synthetic nucleic acids (rsNA) research are in compliance with the *NIH Guidelines*.

Biosafety Officer

The Biosafety Officer (BSO) is responsible for facilitating and managing the Biosafety Program. Specific responsibilities of the BSO are as follows:

- a. Review and dissemination of research/teaching applications involving the use of biological materials to the Institutional Biosafety Committee.
- b. Conducting periodic inspections to ensure laboratory standards are rigorously followed.

- c. Report to the IBC and the University's administration any significant problems, including any violations of the *NIH Guidelines*, and any significant research-related accidents or illnesses of which the BSO becomes aware.
- d. Assisting laboratories in developing laboratory specific emergency plans for handling spills and personnel contamination, and investigating laboratory accidents involving rsNA research.
- e. Providing advice on laboratory security, as it relates to the protection and safeguarding of biological materials.
- f. Providing technical advice to Principal Investigators and the IBC on research safety procedures for the purpose of assuring that the use of human etiologic agents conforms to the University policy and applicable governmental regulations.
- g. In support of Principal Investigators, determine the necessity for health surveillance of research personnel, and require, if appropriate, a health surveillance program for the project.

Principal Investigator

The Principal Investigator (PI) is the faculty member in charge of a research project or class. PIs bear the primary responsibility for compliance with all applicable laws, regulations, and guidelines. A subordinate role on a research project does not excuse non-compliance. For a comprehensive list of the investigator responsibilities for conducting rsNA research, refer to the [Investigator Responsibilities](#) in *The NIH Guidelines*. In addition, the specific responsibilities of the PI are as follows:

- a. To determine whether experiments are covered by any statute, regulation, or guideline, and to comply with the applicable requirements before, during, and after the experiment.
- b. To initiate and/or modify research or teaching activities which require BSO review and/or IBC approval until that research or the proposed modification thereof has been approved and has met all other requirements of all applicable references.
- c. Develop, review, and approve laboratory-specific and/or protocol-specific procedures, consulting with the BSO when necessary.
- d. Ensure that all personnel under his/her supervision are adequately trained in good microbiological techniques and have received required training for the procedures and materials involved in their work.
- e. To provide personal protective equipment to all personnel based on the experimental procedures used in the lab.
- f. To diligently supervise the laboratory staff to ensure that all required safety practices and techniques are employed.
- g. Provide training and information to all employees under his/her supervision regarding laboratory-specific and protocol-specific hazards and document such training (e.g., familiarize staff with signs and symptoms associated with the agent they will be working with, and any other pertinent information such as incubation period, before allowing personnel to work with the agent).
- h. Ensure that all at-risk personnel have been informed of risk assessments and/or provisions for any recommended precautionary medical practices, such as

vaccinations and any special health or handling requirements regarding potentially biohazardous materials or toxins used or stored in the laboratory or work area. Inform personnel of the reasons and provisions for any precautionary medical practices advised or requested.

- i. To report any significant incidents (including failure of safety controls), deviations from the BMBL or *NIH Guidelines*, or any significant research-related accidents and/or illnesses to the BSO and Environmental Health and Safety immediately.
- j. Act upon requests and/or directives from Environmental Health and Safety and/or BSO in order to correct any unsafe laboratory conditions or behaviors.
- k. Ensure that appropriate containment devices and other engineering controls are in place and operate correctly, are current with certifications (where applicable) and are used according to established procedures.
- l. Conduct regular laboratory safety inspections and participate in audits and evaluations.
- m. To make a copy of all applicable guidelines (BMBL, *NIH Guidelines*, OSHA Bloodborne Pathogen standard, etc.) available for laboratory personnel.
- n. To maintain written documentation for all training activities, including instruction in laboratory safety procedures for all research staff personnel.
- o. To refrain from activities which require biosafety level 3 or 4 containment.
- p. To ensure that any research proposal budget includes funding for all safety, administrative, transportation, and waste disposal costs associated with the activity.

These responsibilities are standard and should not be construed as exhaustive. Any applicable guidance could impose particular procedures and safety practices beyond those listed here. In the event of any questions, Principal Investigators shall contact the BSO and/or the Office of Research.

Employees / Laboratory Workers

All employees performing work with biological materials must accept a shared responsibility for operating in a safe manner. Ultimately, each individual is responsible for his/her own safety. Worker responsibilities include:

- a. Ensure that all work is conducted in accordance with established policies and guidelines described in this document and/or specific laboratory protocols and procedures.
- b. Be familiar with the hazards of the materials or substances used.
- c. Read and understand all guidelines, rules, and regulations pertaining to the work being conducted.
- d. Promptly report any job relating injuries, exposures, and/or illnesses to the PI and/or BSO and seek medical treatment immediately.
- e. Immediately report all hazardous conditions to the PI and/or BSO.
- f. Wear and properly maintain all personal protective equipment necessary to perform each task.
- g. Ensure proper use of all engineering controls (e.g., biosafety cabinet, centrifuge safety cups/rotors).

- h. Complete appropriate training as required by federal, state, and university requirements. Consult with supervisor to receive appropriate instruction and training specific to the laboratory, such as handling and disposal of biohazardous materials and incident management and response procedures.

Associated Committees

The following committees consult and coordinate with the IBC when any proposed research involves the use of potentially biohazardous materials or activities.

- a. Institutional Review Board (IRB)

This committee reviews and oversees all research involving human subjects to ensure that the subject's rights and welfare are adequately protected. This committee consults and coordinates with the IBC when any proposed research involves the use of potentially biohazardous materials or activities.

- b. Institutional Animal Care and Use Committee (IACUC)

This committee reviews and oversees all research involving animal subjects and ensures the ethical and humane use of these animals by providing quality animal care in a research environment that promotes good science. This committee consults and coordinates with the IBC when any proposed research involves the use of potentially biohazardous materials or activities.

I. Institutional Biosafety Committee Research Registration Requirements

Protocol Submission Process

1. IRBNet

Principal Investigators conducting research with any of the materials listed above, must submit an IBC Registration Form to the IBC. All submissions, including new protocols, amendments, annual renewals, and close-out reports, must be submitted electronically via IRBNet (www.irbnet.org). All submissions must be electronically signed and submitted by the Principal Investigator. It is highly recommended that all IRBNet users complete the online IRBNet training, provided on the [ODU IRBNet webpage](#) (link and login information to the training modules can be found towards the bottom of the webpage). For additional assistance, contact the Office of Research (757-683-3460).

2. Training Requirements

All applicable training must be completed before the IBC will approve any protocol. Training in part is conducted online through the Collaborative Institutional Training Initiative (CITI, www.citiprogram.org), and includes, but is not limited to, Bloodborne Pathogen training, NIH Guidelines training, Basic Biosafety, Select Agent and Toxin training. In addition, all personnel working within any laboratory where hazardous materials are used and/or stored must complete the Environmental Health and Safety (EH&S) Laboratory Safety Training. Additional EH&S training courses can be found on their [Safety Training website](#).

3. IBC Registration Form

This registration form is found on IRBNet under the 'Forms and Templates' section (it is advisable to always download a new form directly from IRBNet as the form is updated periodically). The protocol registration form allows for the PI to describe their proposed research and the necessary work practices, procedures, and engineering controls that have been implemented in order protect all personnel from exposure to potentially hazardous materials. The IBC must review and approve all registration forms before any proposed research may commence.

Once submitted to IRBNet, the IBC Research Compliance Coordinator (RCC) will review the application to ensure all applicable sections have been completed. The RCC will notify the PI of any incomplete application(s) which will require modification and resubmission prior to IBC review.

4. Teaching Laboratory Registration Form

The IBC developed a registration form strictly for teaching laboratories. This form is a shortened version of the full IBC Registration Form and should be used for all laboratory

classes involving the use of biological materials. This form will be reviewed by the IBC electronically through IRBNet and does not require review at a convened meeting. The committee may decide to refer any teaching laboratory registration form for review during the next convened meeting

II. Biosafety Regulations and Guidelines

There are several federal, state, and local agencies that either regulate or provide guidelines covering the use of biological agents. A summary of these regulations and guidelines is provided below.

1. United States Department of Health and Human Services – Centers for Disease Control (CDC) and the National Institutes of Health (NIH): [Biosafety in Microbiological and Biomedical Laboratories \(BMBL\)](#), 6th edition. This publication consists of guidelines for microbiological safe work practices, safety equipment, and facilities that constitute the four biosafety levels. The BMBL is considered the standard for biosafety and is the basis for this manual.
2. NIH: [Guidelines for Research Involving Recombinant and Synthetic Nucleic Acid Molecules \(NIH Guidelines\)](#). This document provides guidance and must be adhered to at all times for any work involving the constructing and/or handling of recombinant or synthetic nucleic acid (rsNA) molecules, and organisms containing rsNA. Institutions that received any NIH funding for rsNA research are required to comply with the guidelines established by this document. The NIH Guidelines require that each institution establish an IBC with the authority to approve proposed rsNA research using the guidelines as a minimum standard.
3. Occupational Safety and Health Administration (OSHA): [Bloodborne Pathogens Standard](#) (29 CFR 1910.1030). This regulation covers occupational exposures to human blood and other potentially infectious material (OPIM), including human tissue and cells. OSHA specifies a combination of engineering [controls](#), work practices, and training to reduce the risk of exposure and subsequent infections to bloodborne pathogens. Personnel potentially exposed to human blood or OPIM must be offered immunization against the Hepatitis B virus (HBV) and receive annual training. Personnel who work with HIV or HBV in a research laboratory must receive additional training and demonstrate proficiency in working with human pathogens.
4. CDC and United States Department of Agriculture (USDA), Animal and Plant Health Inspection Service (APHIS): [Federal Select Agent Program](#). These regulations cover the possession, use, and transfer of biological agents and toxins that affect humans, animals, and plants and which have been determined to be potential bioterrorism agents. These agents have been categorized as select agents and toxins, and entities and personnel who wish to work with these agents or toxins must be registered with the CDC or APHIS before acquiring or having access to them. These regulations mandate strict requirements for biosafety, emergency planning, and security of select agents and toxins.

Note: ODU is not currently authorized to possess, use, or transfer select agents or toxins.

5. Dual Use Research of Concern (DURC) is defined in the [United States Government \(USG\) Policy for Institutional DURC Oversight](#) (released September 24, 2015) as research that can be reasonably anticipated to provide knowledge, information, products, or technologies that could be directly misapplied to pose a significant threat with broad potential consequences to public health and safety, agricultural crops and other plants, animals, the environment, materiel, or national security. This policy, which establishes review procedures and oversight requirements for potential DURC research at institutions receiving Federal funds for life science research, becomes effective on September 24, 2015. Alongside the [March 2012 DURC Policy](#), these two USG policies provide oversight of DURC when the research is funded by the USG or when the research is taking place at institutions receiving funding from the USG. These two policies work to engage the research community and Federal departments and agencies funding such research. For more information, please go to the [NIH Office of Biotechnology Activities DURC website](#).

Note: At the present time, Old Dominion University is not authorized with the Federal Select Agent Program to possess, use, or transfer any of the agents or toxins listed in the USG Policies, and therefore, research as described in these policies cannot currently be conducted at this institution. The Application for the Use of Biological Materials contains a set of specific DURC identifying questions, which is part of the consistent effort to ensure that all local, state, university, and federal regulations and guidelines are always abided by.

6. Virginia Department of Environmental Quality (DEQ): [Virginia Hazardous Waste Management Regulations](#) (9VAC20-60). These regulations closely follow federal standards established under the Resource Conservation and Recovery Act (RCRA) and list the requirements for the storage, treatment, and disposal of hazardous waste, as well as requiring permits for conduct of such activities.
7. Virginia Department of Environmental Quality (DEQ): [Virginia Regulated Medical Waste Management Regulations](#) (9VAC20-121). The purpose of this regulation is to establish standards and procedures pertaining to regulated medical waste management to protect public health and safety, and to enhance the environment and natural resources. This includes specific requirements on the use of autoclaves to treat and dispose of biohazardous waste.
8. Import and export regulations. Both the CDC and APHIS have strict requirements for importing and exporting certain products and organisms into and out of the country. [APHIS](#) issues permits for the import, transit and release of regulated animals, animal products, veterinary biologics, plants, plant products, pests, organisms, soil, and genetically engineered organisms through their Import and Export Guidelines and Regulations. The CDC regulates the importation of infectious biological agents, infectious substances, and vectors of human disease into the United States through the

[Import Permit Program \(IPP\)](#).

9. Packing and Transportation Requirements of Biological Substances. Specimen shipments in the United States and internationally are regulated under either the [Hazardous Materials Regulations](#) (U.S.) or Dangerous Goods Regulations (international). As such, these specimens require packaging that meets Department of Transportation (DOT) (domestic shipments in the United States) Transportation of Hazardous Materials Regulations (HMR) and International Air Transport Association (IATA) (international air shipments worldwide) Dangerous Goods Regulations (DGR). To arrange shipment of hazardous materials, you must contact ODU EH&S (757-683-4495) for guidance and assistance.

III. Biosafety Practices and Principles

Biological safety, or biosafety, is the application of knowledge, techniques, and equipment to prevent personnel and environmental exposure to potentially infectious agents or other biohazards. Biosafety defines the containment conditions under which these materials can be safely manipulated and the necessary work practices to mitigate the risk of exposure. The objective of containment is to confine biohazards in order to prevent exposure to laboratory workers and the outside environment, which can be accomplished through the concept of primary and secondary barriers.

Primary Barriers

Primary containment protects the laboratory workers and immediate laboratory environment from exposure to biological agents. It is achieved through good microbiological work practices and the use of safety equipment and personal protective equipment (PPE). Primary barriers include biological safety cabinets (BSCs), safety centrifuge buckets, gloves, goggles, and respirators.

Secondary barriers

Secondary containment protects the environment outside the laboratory and is provided by facility design and operational procedures. Examples of secondary barriers include controlled room access, negative airflow ventilation systems, handwashing facilities, established decontamination procedures, and appropriate bio waste disposal protocols.

Laboratory Practices and Techniques

The use of good microbiological work practices is the most important element of containment. Laboratory personnel must always be aware of the hazards within the laboratory and must be trained in order to safely handle and dispose of these materials. While each individual is responsible for their own safety and of those around them, the Principal Investigator is ultimately the sole individual responsible for ensuring that all personnel working in their laboratory are adequately trained.

Each individual laboratory must supplement this manual with laboratory specific policies, procedures, and training that will minimize the specific risks present in the laboratory.

Safety Equipment

Safety equipment includes biological safety cabinets (BSC), centrifuge safety cups and rotors, and other engineering controls that are designed to minimize exposure to biological agents. BSCs are the most important safety equipment for the protection of personnel, the laboratory environment, and depending on the type of BSC, product protection. Safety equipment is most effective at minimizing exposure when workers are trained on the proper use of such equipment, provided the equipment is regularly inspected and maintained.

BioSafety Cabinets (BSCs)

Most types of BSCs are designed to provide personnel, environmental, and product

protection, but only when utilized correctly. There are three designs of BSCs, designated as Class I, II, and III, which have been developed to meet various research and clinical needs. BSCs use high efficiency particulate air (HEPA) filters in their exhaust and/or supply systems and are designed for use with infectious or toxic materials. HEPA filters capture a minimum of 99.9% of particles 0.3 μm in size and become more efficient at particle sizes smaller and larger than this.

BSCs should not be confused with other laminar flow devices or “clean benches”. Vertical flow clean benches are useful in some settings, such as in compounding pharmacies, but direct air towards the operator and do not offer personnel protection. Horizontal flow clean benches (Figure 1) discharge HEPA filtered air from the back of the cabinet, across the work surface, and towards the user. Therefore, these pieces of equipment should never be used for working with hazardous materials where personnel protection is required. Both the vertical and horizontal clean benches protect the product, but do not offer protection to the user or the environment (laboratory).

As with any piece of laboratory equipment, it is critical to train all users of the BSC on its correct use and maintenance to ensure that personnel, product, and environmental protection is maintained. The correct location, installation, and certification of the biological safety cabinet is critical to containing infectious aerosols. BSCs should never be installed underneath supply registers or exhaust vents, near laboratory doors, or in high-traffic areas, as these can cause disruptions in the protective airflow patterns. All BSCs must be inspected and certified **annually** by trained service personnel according to the National Sanitation Foundation (NSF) Standard 49, Annex F. Inspection and re-certification is required more whenever the cabinet is relocated and after major repairs, filter changes, etc.

1. Class I Biosafety Cabinets

The Class I BSC provides personnel and environmental protection but does not offer product protection. The air movement in a class I BSC is similar to a chemical fume hood but has a HEPA filter in the exhaust system to protect the environment (Figure 2).

Figure 1: Horizontal Flow Clean Bench - (A) front opening; (B) supply grille; (C) supply HEPA filter; (D) supply plenum; (E) blower

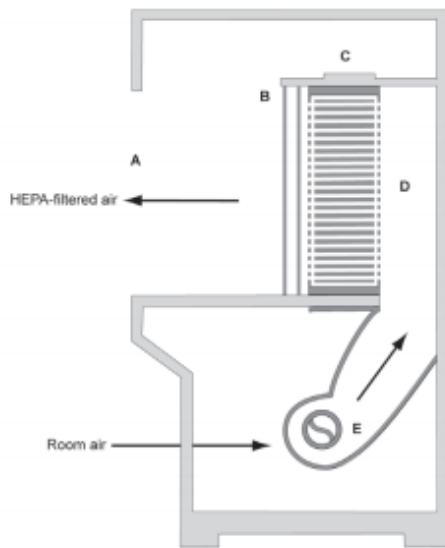
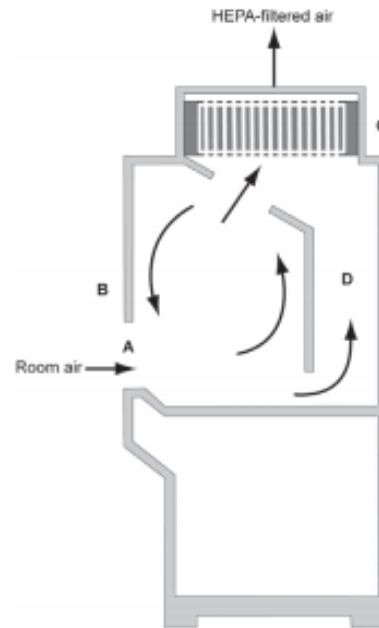


Figure 2: Class I BSC - (A) front opening; (B) sash; (C) exhaust HEPA filter; (D) exhaust plenum



Source: Biosafety in Microbiological and Biomedical Laboratories, 6th Edition

2. Class II Biosafety Cabinets

The Class II BSCs provide personnel, product, and environmental protection. These cabinets operate by drawing air into the front grille of the cabinet, which provides personnel protection. Then, the air is drawn beneath the work surface and then forced to towards the top of the BSC where it is then pulled downwards through a supply air HEPA filter which provides product protection. Some of the air does not recirculate within the cabinet and is exhausted through a HEPA filter and is either recirculated back into the laboratory environment or discharged from the building via a canopy or “thimble” connected to the building exhaust. Class II BSCs are separated into Type A, B, and C cabinets based upon construction, airflow, and exhaust systems.

a. *Class II, Type A1 BSC (Figure 3)*

These cabinets maintain an average inflow velocity of 75 fpm (feet per minute) at the face opening, recirculate approximately 70% of the air through the supply HEPA filter, and discharge approximately 30% through the exhaust HEPA filter. **These cabinets are not to be used for work involving volatile toxic chemicals.**

b. *Class II, Type A2 (Figure 4)*

This type of BSC has an average inflow velocity of 100 fpm and recirculates and discharges air within the cabinet at a rate of 70% and 30%, respectively, similar to the Type A1 BSC. Minute quantities of volatile toxic chemicals or radionuclides can be used in this type of cabinet **only** if it exhausts to the outside via a canopy connection with exhaust alarm.

Figure 3: Class II Type A1 BSC - (A) front opening; (B) sash; (C) exhaust HEPA filter; (D) supply HEPA filter; (E) common plenum; (F) blower

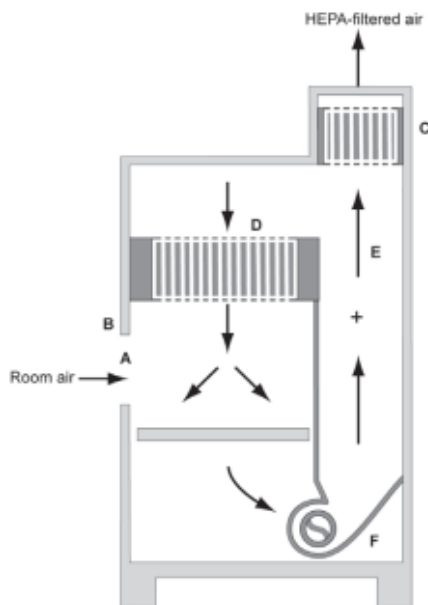
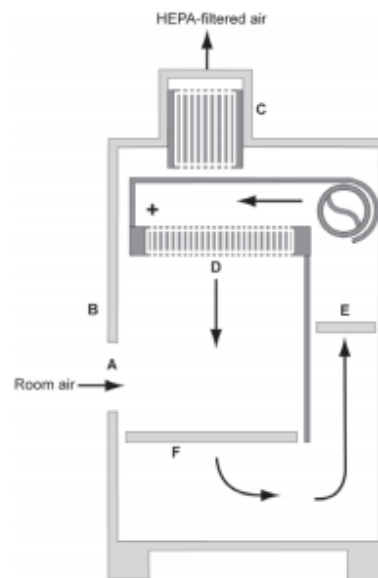


Figure 4: Class II Type A2 BSC- (A) front opening; (B) sash; (C) exhaust HEPA filter; (D) supply HEPA filter; (E) positive pressure common plenum; (F) negative pressure plenum



Source: Biosafety in Microbiological and Biomedical Laboratories, 6th Edition

c. *Class II, Type B1 (Figure 5)*

The Type B1 cabinets maintain an average inflow velocity of 100 fpm at the face opening, recirculate approximately 30% of air through the supply HEPA filter, and discharge approximately 70% through an exhaust HEPA filter. Down-flow air that is pulled through the rear grille of the cabinet is exhausted directly through a HEPA filter and to the outside via hard ducting without recirculation within the cabinet or laboratory. This allows for manipulations of minute quantities volatile chemicals and radionuclides as long as the work is performed in the direct exhaust (rear) portion of the BSC.

d. *Class II, Type B2 (Figure 6)*

Type B2 cabinets maintain an average inflow velocity of 100 fpm at the face opening and are total exhaust cabinets, meaning no air is recirculated within the BSC or laboratory. All air that enters the cabinet is exhausted and passes through a HEPA filter prior to discharge to the outside. These cabinets are expensive to operate, and their purchase and use should be justified by the research. Type B2 cabinets do not provide additional biosafety protection over other Class II BSCs.

Figure 5: Class II Type B1 BSC - (A) front opening; (B) sash; (C) exhaust HEPA filter; (D) supply HEPA filter; (E) negative pressure dedicated exhaust plenum; (F) blower; (G) additional HEPA filter for supply air

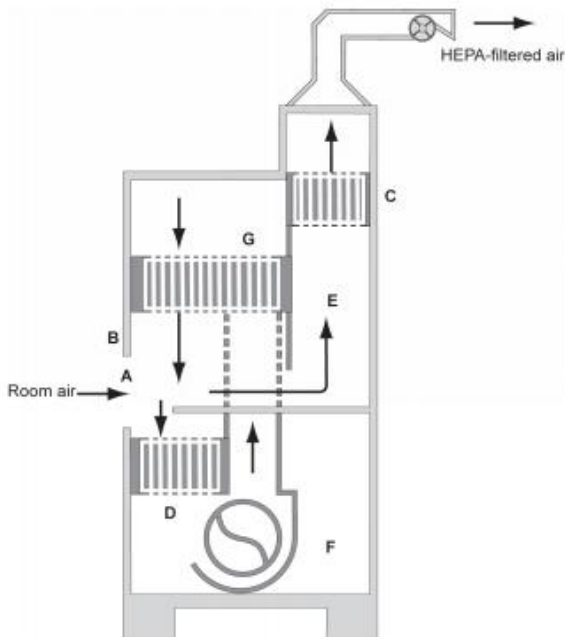
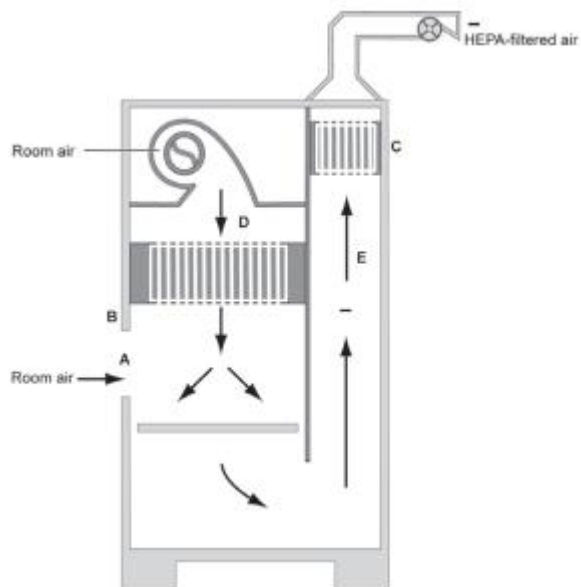


Figure 6: Class II Type B2 BSC - (A) front opening; (B) sash; (C) exhaust HEPA filter; (D) supply HEPA filter; (E) negative pressure exhaust plenum



Source: Biosafety in Microbiological and Biomedical Laboratories, 6th Edition

e. *Class II, Type C1 (Figure 7)*

This BSC is similar to a Type B1 BSC in that it has a special region of the work area intended for work with toxic volatile chemicals that are exhausted from the building. However, it also has an internal exhaust blower that allows the BSC to be either room recirculated if no volatile toxic chemicals or vapors are present or canopy-connected with an exhaust alarm if volatile toxic chemicals are used. The downflow air in the center of the work area is typically directly exhausted and not recirculated.

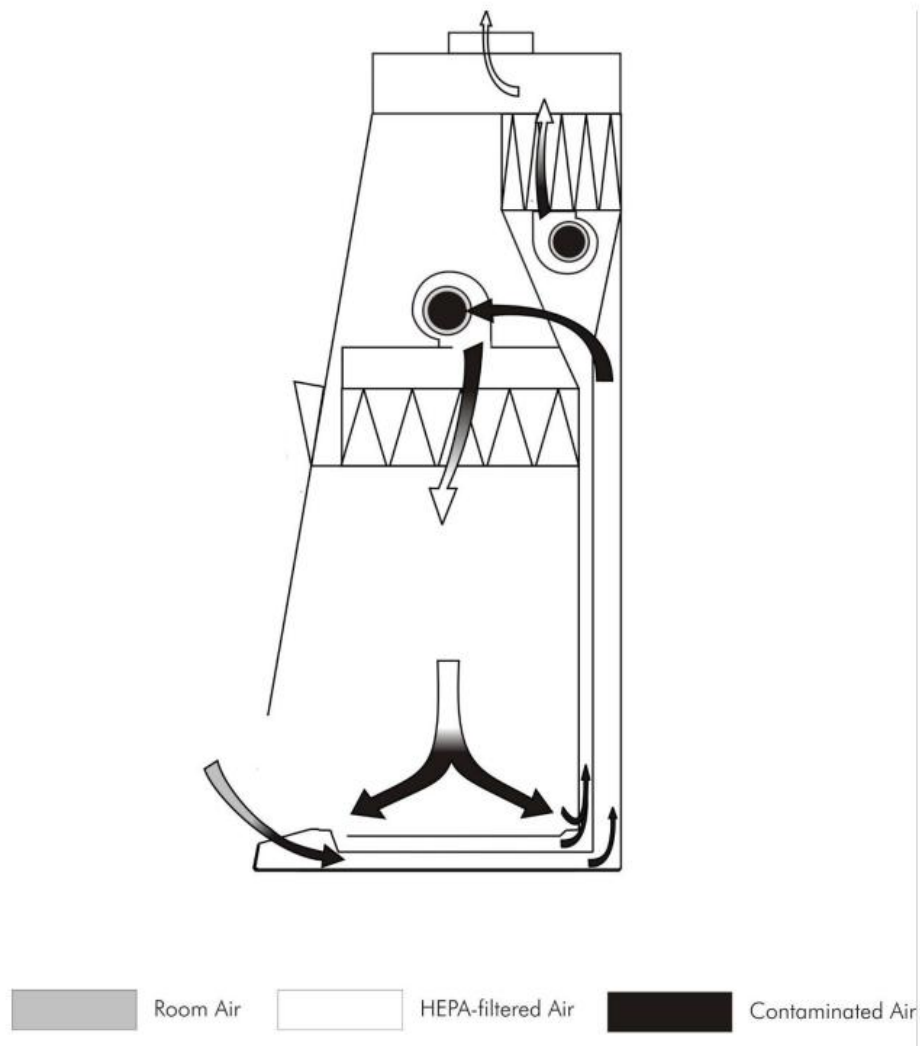


Figure 7: Airflow patterns for Class II Type C1 BSCs.

Source: NSF/ANSI 49 - 2016, Annex E

3. Class III Biosafety Cabinets (Figure 8)

The Class III BSC was designed for work with highly infectious microbiological agents and for the conduct of hazardous operations and provides maximum protection. They are a glove-box design (gas-tight containment) where both the supply and exhaust air is HEPA filtered. All exhaust air is discharge to the outdoors through double HEPA filters. Movement of materials in and out of the Class III cabinet requires passage through a dunk tank (filled with a chemical disinfectant) or double-door pass-through box (e.g., an autoclave) that can be decontaminated between uses. These cabinets are most appropriate for work involving Risk Group 4 agents.

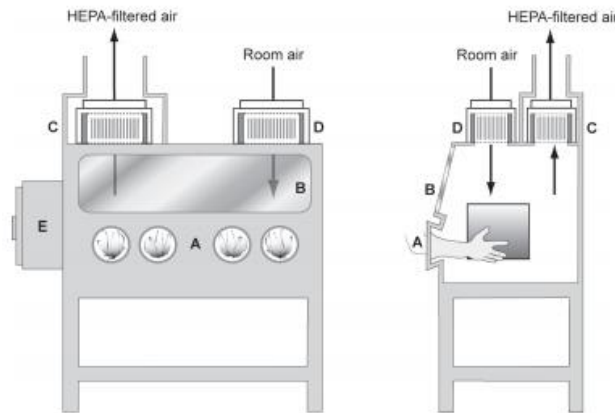


Figure 8: Class III BSC - (A) glove ports with O-ring for attaching arm-length gloves to cabinet; (B) sash; (C) exhaust HEPA filter; (D) supply HEPA filter; (E) double-ended autoclave or pass-through box

Source: Biosafety in Microbiological and Biomedical Laboratories, 6th Edition

4. Guidelines for Safe Use of Biosafety Cabinets

The installation and use of a BSC is an indication that safe work practices are required in order to prevent contamination and infection. BSCs are engineered to provide excellent containment of microorganisms when used properly. They are not substitutes to good work practices and only serve to complement a safe worker in the laboratory. The following recommendations are geared towards Class II Type A2 BSCs, as they are the most commonly used BSC in laboratories and are considered best practices that should be adhered to all times while working inside a BSC.

a. *Preparing for Work*

- Don applicable PPE (gloves, lab coat, safety glasses/goggles, etc.)
- If the BSC is off, turn it on and wait 5 minutes before using. Ensure there are no obstructions at either the front and/or rear air grilles.

- Turn off UV lamp (if equipped). UV light can burn eyes and skin. Personnel should be familiar with the safe and effective use of any UV lamps located inside the BSC.

Note: The use of UV lamps is discouraged for disinfection purposes as numerous factors affect the germicidal activity of UV light, which require regular cleaning, maintenance, and monitoring to ensure germicidal activity.

- Magnehelic gauge or digital airflow display must be checked before each use and should be recorded. If the numbers recorded deviate significantly, the cabinet should not be used until the cause of the deviation has been identified and fixed. Causes of deviations could be issues with the motor blower fan, loaded HEPA filter, or a puncture or tear in the HEPA filter.
- Ensure the front sash is at the correct height and that the chair/stool is at a height so that the user's face is above the front opening.
- To prevent contamination, it is recommended to disinfect all interior surfaces of the BSC before use. In addition, all materials placed inside the BSC should be decontaminated prior to placement within the cabinet.
- Only place materials that are necessary to conduct the desired work. The BSC should never be overloaded with items and must never be used for storage. Ensure that absorbent materials (e.g., paper towels) and appropriate disinfectant are placed in the cabinet.
- If working with sharps, place a sharps container within the BSC for safe disposal
- Place a biohazard bag within the BSC for all non-sharp waste produced during work in the BSC.

b. Working in the BSC

- Never, at any time, block the front or rear air intake grilles.
- Limit work to one person per BSC. BSCs are designed for only a single operator at a time, more than one operator may produce significant disturbances in the airflow to cause a breach in containment. If it is necessary to have two people working inside the BSC at a time, they must coordinate their work in a manner which minimizes airflow disturbances (e.g., slow movements, not overloading the cabinet with supplies, minimizing the need to remove arms from the cabinet while working, etc.). If two people are working inside a cabinet and one of them needs to remove their arms from the inside of the BSC, the other worker should stop working and all cultures, tubes, etc. should be closed prior to removing arms from the cabinet.
- All work should be conducted at least four inches in from the front intake grille.
- Avoid rapidly moving your arms in and out of the cabinet, limit individuals from walking behind the cabinet while working, and limit laboratory doors being

opened, all of which may disrupt the protective air curtain protecting the user.

- Open flames (e.g., Bunsen burners, lighters, etc.) must never be used inside the BSC as they create airflow turbulence that may compromise personnel protection and sample sterility. Additionally, since BSCs recirculate air, the heat build-up may damage the HEPA filters. Instead, use electric devices such as loop sterilizers or incinerators, or disposable loops.
- If centrifuges, blenders, or other pieces of equipment are placed inside the BSC which may create air turbulence, they should be placed in towards the back of the cabinet and no work should be conducted during their operation.
- Work slowly, avoid crossing arms and swift arm movement inside the cabinet.
- When entering or exiting the cabinet, always move arms straight in or out of the BSC.
- If a vacuum system is in place, dual aspirator flasks in series and an in-line HEPA filter between the vacuum trap and source of the valve in the cabinet is required. The flasks must be placed within a secondary container (Figure 8).



Figure 8: To collect liquid biohazardous waste, aspirate liquid into a flask containing appropriate disinfectant (A) connected to second overflow collection flask (B) and separated from vacuum system (D) by an in-line filter (C). All biohazardous waste must be properly neutralized before disposal. Source: Biosafety in Microbiological and Biomedical Laboratories, 6th Edition

- Always work slowly and methodically, in a clean to dirty direction (e.g., have all media and sterile equipment on one side of the BSC, and waste items on the other side, see Figure 9).

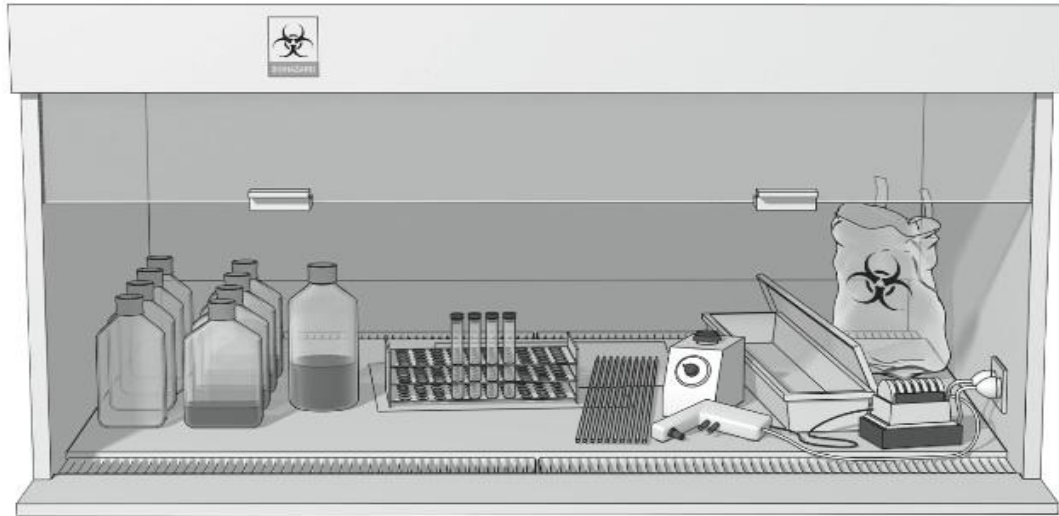


Figure 9: A typical layout for working “clean to dirty” within a Class II BSC.
Source: Biosafety in Microbiological and Biomedical Laboratories, 5th Edition

- Clean all spills immediately
- Replace gloves whenever they are torn, punctured, or contaminated.
- Discard all sharps waste into the sharps container within the BSC. If the container becomes 3/4 full, decontaminate the outside surfaces, and remove from the BSC. Replace with a new sharps container.
- Discard all non-sharps waste in the biohazard bag within the BSC. If the bag becomes full, decontaminate with appropriate disinfectant, seal the bag, and move to a laboratory biohazard waste container. Replace with a new biohazard bag.

c. *Upon Completion of Work*

- Disinfect outside surfaces of all items coming out of the BSC.
- Decontaminate the biohazard bag, seal it, and remove it from the BSC and place in appropriate laboratory biohazard waste container.
- Remove gloves and replace with a new pair for disinfecting the cabinet.
- Disinfect all interior surfaces of the BSC, including the inside of the sash, side walls, and back wall. If using bleach for disinfection, rinse with water to prevent pitting of stainless steel.

Note: Never place your head inside the BSC at any time.

- Remove gloves and wash hands
- Turn the cabinet off and close the front sash.

Note: BSCs are designed for 24-hour per day operation and do not need to be turned off after use. However, many investigators prefer to turn them off to reduce the noise in the laboratory and for energy conservation purposes, especially if the cabinet is infrequently used. If the BSC is left running, do not close the front sash.

5. BSC Certification Requirements

Biosafety cabinets are tested by the manufacturer in accordance with National Sanitation Foundation (NSF) Standard 49 criteria. All BSCs must be inspected and certified after installation, before use, and on an annual basis. The BSC must also be certified whenever it is moved and after any repairs are completed.

The following companies can be used for BSC certification:

- [Environmental Safety Professionals](#) (ESP) 800-688-7167 or (919) 217-2650
- [KeyStone](#) 757-333-4988

Annual certifications are required in order to ensure that the cabinet still performs as it did when it obtained NSF certification at the factory. NSF has established a certification program for certifiers to ensure a minimum level of competency and professionalism. It is recommended that the certifiers are trained service personnel according to National Sanitation Foundation (NSF) Standard 49, Annex F. A BSC that does not pass certification, or that is overdue for its certification, must not be used.

Personal Protective Equipment (PPE)

PPE includes safety eyewear, laboratory coats, gloves, respiratory protection, and other equipment designed to supplement the containment provided by laboratory practices and engineering controls. PPE is considered the last line of defense after engineering controls, work practices, and administrative controls. Lab coats should be discarded or laundered periodically (e.g., weekly) or whenever they become contaminated. Cloth lab coats must never be brought home for laundering. Autoclaving these lab coats is an acceptable practice for decontaminating them for re-use. An alternative option is to establish a contract with a laundering facility who is trained to handle potentially contaminated PPE for cleaning and decontamination purposes. Disposable gloves are to be discarded after each use and must never be re-used.

Facility Design

Facility design features include physical separation of laboratories from public access, sophisticated ventilation system to prevent airborne biological agents from migrating outside of the laboratory, location of autoclaves, security access controls, and other features developed into the design of the laboratory. These features protect both the personnel within the laboratory by reducing the potential for laboratory exposures, as well as the outside environment.

IV. Biosafety Levels and Risk Groups

Biosafety levels

Combinations of laboratory practices, containment equipment, and special laboratory design can be made to achieve different levels of physical containment. There are currently four

biosafety levels which define the level of containment necessary to protect personnel and the environment. Biosafety levels 1 (BSL-1) and 2 (BSL-2) are the least restrictive and are available at ODU. Biosafety levels 3 (BSL-3) and 4 (BSL-4) require special containment laboratories or facilities, and are not available at ODU. This manual will mainly focus on the biosafety levels available at this institution (BSL-1 & BSL-2), however, a summary of all four biosafety levels can be found in Table 1 v .

The recommended biosafety level for an organism or toxin represents the conditions under which the agent can be safely handled. The laboratory Principal Investigator is responsible for assessing risks and for appropriately applying the recommended biosafety levels.

In addition to the four biosafety levels described above, there are also four biosafety levels for work with infectious agents in vertebrate animals, which are described in Section VIII.

Risk groups

The principal hazardous characteristics of an agent are:

1. Its capability to infect and cause disease in a susceptible human or animal host
2. Its virulence as measured by the severity of disease
3. The availability of preventive measures and effective treatments for the disease

The *NIH Guidelines* established a comparable classification and assigned the agents into four risk groups on the basis of hazard. The descriptions of the NIH risk group classifications are summarized below in Table 2. While risk groups correlate with biosafety levels, they do not equate. For example, there are some risk group 2 organisms that require BSL-3 containment, and vice versa. A risk assessment is required to determine the degree of correlation between an agent's risk group classification and required biosafety level.

Table 1: Biosafety Levels				
Biosafety Level	Agents	Work Practices	Primary Barriers and Safety Equipment	Facilities (secondary barriers)
1	Not known to consistently cause disease in healthy adults	Standard microbiological practices	- No primary barriers required. - PPE: laboratory coats and gloves; eye and face protection, as needed	Laboratory bench and sink required
2	- Agents associated with human disease - Routes of transmission include percutaneous injury, ingestion, mucous membrane exposure.	BSL-1 practice plus: - Limited access - Biohazard warning signs “Sharps” precautions - Biosafety manual defining any needed waste decontamination or medical surveillance policies	- BSCs or other physical containment devices used for all manipulations of agents that cause splashes or aerosols of infectious materials - PPE: Laboratory coats, gloves, face and eye protection, as needed	BSL-1 plus: Autoclave available
3*	Indigenous or exotic agents that may cause serious or potentially lethal disease through the inhalation route of exposure	BSL-2 practice plus: - Controlled access - Decontamination of all waste - Decontamination of laboratory clothing before laundering	- BSCs or other physical containment devices used for all open manipulations of agents - PPE: Protective laboratory clothing, gloves, face, eye and respiratory protection, as needed	BSL-2 plus: - Physical separation from access corridors - Self-closing, double-door access - Exhausted air not recirculated - Negative airflow into laboratory - Entry through airlock or anteroom - Hand washing sink near laboratory exit
4*	- Dangerous/exotic agents which pose high individual risk of aerosol-transmitted laboratory infectious that are frequently fatal, for which there are no vaccines or treatments. - Agents with a close or identical antigenic relationship to an agent requiring BSL-4 until data are available to redesignate the level	BSL-3 practices plus: - Clothing change before entering - Show upon exit - All material decontaminated before removal from facility	All procedures conducted in Class III BSCs or Class I or II BSCs in combination with full-body, air-supplied, positive pressure suit	BSL-3 plus: - Separate building or isolated zone - Dedicated supply and exhaust, vacuum, and decontamination systems - Other requirements outlined in the BMBL.

	• Related agents with unknown risk of transmission			
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*Work requiring BSL-3 or BSL-4 containment cannot be conducted at ODU

Table 2: Basis for the Classification of Biohazardous Agents by Risk Group	
Risk Group Classification	Risk
1	Agents that are not associated with disease in healthy adult humans
2	Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available
3	Agents that are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available
4*	Agents that are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available

*Work with RG4 organisms cannot be conducted at ODU. Very limited work with RG3 agents may be approved at the discretion of the Institutional Biosafety Committee.

For more information regarding the biosafety levels or risk groups, contact the ODU Biosafety Officer.

V. Routes of Infection

Awareness of the routes of transmission for the natural human disease is helpful in identifying probable routes of transmission in the laboratory and the potential for any risk to the public health. It is important to remember the nature and severity of disease caused by a laboratory infection and the probable laboratory route of transmission of the infectious agent may differ from the route of transmission and severity associated with the naturally-acquired disease. The predominant probable routes of transmission in the laboratory are:

Skin and mucous membrane contact

Low energy procedures such as decanting of liquids, pipetting, removal of screw caps, vortexing, streaking agar plates, and inoculation of animals can result in the generation of infectious droplets, as well as result in direct contact with infectious material. Eye contact is also a route of exposure and safety eyewear must be worn as needed to prevent sprays or splashes to the eyes.

Percutaneous inoculation

Use of syringes, needles, Pasteur pipettes, and other sharp devices present a high risk of exposure through inoculation. Inoculation can also occur as a result of cuts and scratches from contaminated items and animal bites/scratches.

Ingestion

Mouth pipetting must never be performed in the laboratory as it presents the highest risk for ingestion of infectious material. Splashing of material into the mouth, and indirect oral exposure through touching the mouth with contaminated hands, and eating and drinking in the lab can also result in ingestion of infectious material.

Inhalation

Procedures that impart energy to a microbial suspension will produce aerosols. Aerosols are a serious hazard because they are ubiquitous in laboratory procedures, are usually undetected, and are extremely pervasive, placing the laboratory worker conducting the procedure and others in the laboratory at risk of infection. Many procedures have the potential for generation of respirable aerosols including: sonication, homogenization, centrifugation, shaking incubators, “blowing out” of pipettes, heating inoculating loops, dropping glassware, and changing bedding in dirty animal cages.

VI. Biological Risk Assessments

A risk assessment is a careful examination of what organisms, toxins, or procedures could potentially cause harm to those working with them and within the laboratory, the facility, or the external environment. The identification of the risks allow for necessary precautions to be implemented in order to prevent potential exposures and infections within the laboratory.

There is no standard approach for conducting a biological risk assessment, but the five-step approach provided below will help guide Principal Investigators in the process.

The 5Ps of a risk assessment

1. *Identify agent hazards (Pathogen)*

The principal hazardous characteristics of the agent to be evaluated are its capability to infect and cause disease in a susceptible human host, severity of disease, and the availability of preventive measures and effective treatments. The BMBL provides agent summary statements for some agents associated with laboratory acquired infection. Additionally, the [Pathogen Safety Data Sheets](#) provided by the Public Health Agency of Canada are valuable resources that are available to assist during the risk assessment process.

2. *Identify laboratory procedure hazards (Procedure)*

The primary procedural hazards within a laboratory are agent concentration, suspension volume, equipment and procedures that generate small particle aerosols and larger airborne particles, and the use of sharps. Procedures involving animals and the complexity of a laboratory procedure also present hazards. Each laboratory should conduct risk assessments to identify the specific hazards associated with the procedures to be performed.

3. *Determine the biosafety level and select additional precautions (Place)*

The selection of the appropriate biosafety level and any additional precautions require a comprehensive understanding of proper laboratory work practices, safety equipment, and facility safeguards. The Institutional Biosafety Committee will evaluate each application requesting to utilize biohazardous materials and will verify that the Principal Investigator has identified the correct biosafety level and precautions.

4. *Evaluate proficiencies of staff and integrity of safety equipment (People and PPE)*

The personnel within the laboratory are the most important aspect to consider when conducting a risk assessment. The laboratory staff must have acquired the technical proficiency in the use of microbiological practices and safety equipment required for the safe handling of the agent. All individuals within the laboratory must be willing to accept responsibility for protecting one's self and others within the laboratory.

It is also important to recognize that individuals in the laboratory may differ in their susceptibility to disease. Pre-existing diseases, medications, compromised immunity, and pregnancy or breast-feeding that may increase exposure to infants to certain agents, are some of the conditions that may increase the risk of an individual for acquiring a laboratory acquired infection (LAI).

Each laboratory must be adequately equipped with necessary safety equipment and must ensure it is operating properly. PPE must be available in appropriate materials and sizes for all individuals in the laboratory, and must protect them from the hazards they will be exposed to.

5. *Review all risk assessments at least annually and update whenever changes occur in the laboratory, such as:*
 - a. A move or renovation
 - b. A new employee begins working in the lab
 - c. A new agent is introduced
 - d. A new or different piece of equipment is acquired
 - e. New techniques or procedures are introduced

VII. Animal Biosafety

Animal biosafety levels

Laboratory animal facilities are a special type of laboratory. As a general principle, the biosafety level (facilities, practices, and operational requirements) that is recommended for working with infectious agents in vivo and in vitro is comparable. Within the microbiological laboratory, hazardous conditions are caused by personnel or by the equipment being used. In the animal laboratory, the activities of the animals themselves can present new hazards, such as the generation of aerosols, biting, scratching, and the fact that they may be infected with a zoonotic disease.

Laboratory facilities must provide appropriate containment for laboratory animals exposed to or harboring infectious agents. Ideally, facilities for laboratory animals used for studies of infectious or noninfectious disease should be physically separate from other activities.

There are four escalating containment levels consisting of a combination of work practices, safety equipment, and facilities for experiments on animals infected with agents that produce, or may produce, human infection. These four combinations are designated Animal Biosafety Levels (ABSL) 1 - 4. A summary of the requirements for each animal biosafety level is found in Table 3.

All vertebrate animal related activities must be reviewed and approved by the ODU IACUC prior to any work commencing. In addition to IACUC approval, all animal work involving infectious agents or acute toxins shall be reviewed by the ODU IBC. Contact the Office of Research for more information.

Primary Hazards

There are three primary hazards associated with conducting laboratory animal science:

- a. Physical hazards such as bites, scratches, and kicks are ubiquitously associated with laboratory animal contact. Personnel working with larger animals might sustain crushing injuries if the animals kick, fall, or simply shift their body weight. These types of injuries are largely avoidable through proper training in animal-handling techniques. Following any breaks in the skin caused by equipment within the vivarium or by a laboratory animal, proper first aid is critical to avoiding infection. Immediately wash the site with soap and water for 15 minutes. If there was a splash or splatter of material into the eyes, flush the eyes for at least 15 minutes at the nearest eye wash. Report all incidents to EH&S and your immediate supervisor.
- b. Allergic reactions to animals are among the most common conditions that adversely affect the health of workers involved in animal research. Allergic hazards can be associated with breathing or contacting allergens found in animal dander or urine. This type of hazard can be significantly reduced or even eliminated by wearing proper personal protective equipment when handling animals. Those who experience allergic reactions should seek medical consultation either through ODU's

occupational health provider or through their preferred medical professional.

- c. Exposure to zoonotic diseases from the animals being handled which are capable of causing infection in humans

Overview of Zoonotic Diseases

The transmission of zoonotic disease in animal laboratories is uncommon, despite the fact that humans are susceptible to numerous infectious diseases that affect animals. This is due in part to the knowledge of the animal's health status prior to arrival within the vivarium as well as improved veterinary care which has helped to ensure the availability of healthy research animal populations.

Although transmission of zoonotic diseases may be uncommon, it is still critical for all individuals working with or around animals to be aware of the potential hazards.

Infections of animals may sometimes produce severe disease in humans even when the animals themselves show little, if any, sign of illness. It is therefore critical to always be aware of possible consequences when working with any species of animal and to take appropriate precautions in order to minimize the risk of infection. In the event of an illness, it is important to always inform the attending physician of the occupational exposure to laboratory animals.

Table 3: Animal Biosafety Levels				
Animal Biosafety Level	Agents	Work Practices	Primary Barriers and Safety Equipment	Facilities (secondary barriers)
1	Not known to consistently cause disease in healthy adults	Standard animal care and management practices, including appropriate medical surveillance programs	As required for normal care of each species PPE: laboratory coats and gloves; eye, face protection, as needed.	Standard animal facility - No recirculation of exhaust air - Directional airflow recommended - Hand washing sink is available
2	- Agents associated with human disease - Routes of transmission include percutaneous injury, ingestion, mucous membrane exposure.	ABSL-1 practice plus: - Limited access - Biohazard warning signs “Sharps” precautions - Biosafety manual - Decontamination of all infectious wastes and animal cages prior to washing	ABSL-1 equipment plus primary barriers: - Containment equipment appropriate for animal species that cause splashes or aerosols of infectious materials - PPE: Laboratory coats, gloves, face and eye protection, as needed	ABSL-1 facility plus: - Autoclave available - Mechanical cage washer recommended
3*	Indigenous or exotic agents with potential for aerosol transmission; disease may have serious health effects	ABSL-2 practice plus: - Controlled access - Decontamination of clothing before laundering - Cages decontaminated before bedding removed - Disinfectant foot bath as needed	ABSL-2 equipment plus: - Containment equipment for housing animals and cage dumping activities - Class I, II, III BSCs available for manipulative procedures (inoculation, necropsy) that may create infectious aerosols - PPE: appropriate respiratory protection	ABSL-2 facility plus: - Physical separation from access corridors - Self-closing, double-door access - Sealed penetrations - Autoclave available in facility - Negative airflow into animal and procedure rooms
4*	Dangerous/exotic agents that pose high risk of life threatening disease; aerosol transmission, or related agents with unknown risk of transmission	ABSL-3 practices plus: - Entrance through change room where personal clothing is removed and laboratory clothing donned - Shower upon exiting	ABSL-3 equipment plus: Maximum containment equipment (i.e., Class III BSC or partial containment equipment in combination with full body, air-supplied positive-pressure suit) used for all procedures and activities	ABSL-3 facility plus: - Separate building or isolated zone - Dedicated supply and exhaust, vacuum, and decontamination systems - Other requirements outlined in the BMBL

*Work requiring ABSL-3 or ABSL-4 containment cannot be conducted at ODU

VIII. Laboratory Biosafety Practices

standard laboratory practices

Prudent practices and solid laboratory techniques are of primary importance when it comes to laboratory safety. Both are based on sound technical knowledge, experience, common sense, and a positive attitude towards safety. Unlike administrative controls, which are behaviors dictated by regulation or laboratory policy, personal protective behaviors define an innate part of each individual worker's approach to the laboratory environment. These behaviors are the most important line of defense against preventing accidents in the laboratory.

The prudent biosafety practices outlined in Table 4 are to be adhered to all times, by all individuals, in all laboratories. Strict adherence to these basic principles will greatly reduce the likelihood of laboratory acquired infections.

Table 4: Prudent Biosafety Practices	
Biosafety Practice	Routes of Exposure Blocked
Never mouth pipette	Inhalation, ingestion, skin and mucous membrane contact
Always manipulate infectious fluids carefully to avoid spills and the production of aerosols and droplets	Inhalation, ingestion, skin and mucous membrane contact
Always use a biosafety cabinet for aerosol generating procedures	Inhalation, ingestion, skin and mucous membrane contact
Always adhere to safe sharps practices - minimize the use of needles, syringes, and other sharps where possible and dispose of sharps properly	Percutaneous, inhalation
Always wear personal protective equipment - lab coat, gloves, safety eye wear, and other PPE as necessary, such as respiratory protection (e.g., N-95)	Inhalation, ingestion, skin and mucous membrane contact
Always wash hands upon completion of work involving biological material, and whenever gloves are removed	Ingestion, skin and mucous membrane contact
Always decontaminate work surfaces before and after use, and immediately upon any spills	Ingestion, skin and mucous membrane contact
Never eat (to include chewing gum), drink, store food, smoke, handle contact lenses, or apply cosmetics (including lip balm) in the laboratory	Ingestion, skin and mucous membrane contact

Laboratory practice and technique

Individuals working with biological materials must constantly be aware of the potential hazards involved and be appropriately trained on the laboratory practices and procedures. The PI is responsible for ensuring all personnel within the laboratory receive proper training.

While the responsibility to train remains with the PI, a common approach is for the PI to delegate the laboratory manager, supervisor, or other experienced individual to conduct this training.

Every laboratory on campus is expected develop laboratory and procedure specific standard operating procedures (SOP). The intent of these SOPs are to identify the hazards associated with the laboratory, and to outline the work practices designed to mitigate the risks. All laboratory personnel should verify that they have read these documents, which should occur on an annual basis or whenever updates to the documents occur (such as when a new procedure or biological material is added to the laboratory).

Laboratory housekeeping and personal hygiene

It is critical to always maintain a neat, clean area in the laboratory in order to minimize injuries and exposures. Laboratory staff must rely on each other to maximize efficiency and safety at all times. If laboratory areas are shared, all materials and equipment should be properly labeled. Waste is to be discarded properly and the work surfaces disinfected upon completion of work. Abide by the following housekeeping guidelines at all times:

- Ensure work areas are free of significant sources of contamination and hazards.
- Laboratory personnel are responsible for cleaning and maintaining laboratory benches, equipment, and other areas that require specialized technical knowledge.
- Access to exits, sinks, eyewashes, emergency showers, and fire extinguishers must not be blocked.
- Electrical safety is of primary concern within the laboratory, especially as it relates to the use of extension cords. Extension cords are to only be utilized for temporary use, which is defined as a maximum of 30 days. Equipment should be properly grounded and overloaded electrical circuits and the creation of electrical hazards in wet areas must be avoided.
- Unnecessary items on floors, under benches, or in other areas of the laboratory should be removed.
- All compressed gas cylinders must always be properly secured to a fixed object.
- Always wash hands upon the removal of gloves, after completion of work with biological materials, and before leaving the laboratory. Each laboratory must have a dedicated sink for hand washing with soap and paper towels available. Hands must be washed for a minimum of 15 seconds.
- Never eat, drink, smoke, handle contact lenses, or apply cosmetics within the laboratory, or before washing hands after conducting any work with biological materials.
- Flush eye washes on a regular basis, and at least once a month. This prevents accumulation of sediment and bacteria.

Universal Precautions

Universal precautions is an approach to infection control to treat all human blood, tissues,

and certain body fluids as if they are known to be infectious. All blood or other potentially infectious material (OPIM) described in the University's *Bloodborne Pathogen Exposure Control Plan* shall be handled as though they contain a bloodborne pathogen. Under circumstances in which differentiation between body fluid types is difficult or impossible, all body fluids shall be considered potentially infectious materials. Universal precautions as described above and in the Bloodborne Pathogen Exposure Control Plan shall always be used in such circumstances. This concept creates a heightened awareness of potential risk and adds another level of caution to activities involving these types of materials.

Biological hazard information

Laboratory workers must be knowledgeable of the hazards associated with the biological agents present in the laboratory. The hazards associated with each agent should be available and easily accessible to all laboratory personnel. The following sources contain useful information for a wide variety of biological agents and toxins.

Microbial Agents

- a. The BMBL 6th edition provides descriptions of biosafety levels and recommended biosafety practices for specific biological agents.
- b. The Public Health Agency of Canada publishes [Pathogen Safety Data Sheets](#) (PSDS) which describe the hazardous properties of certain biological agents and recommend safe work practices for handling such agents.
- c. The American Biological Safety Association International (ABSA) maintains a [Risk Group Database](#).

Toxins

Purified biological toxins are chemical hazards, although many such toxins produce adverse effects at doses significantly below that of “traditional” laboratory chemicals. Laboratory use of purified toxins falls under the ODU Chemical Hygiene Plan, and Safety Data Sheets (SDSs) must be maintained and available.

- a. SDSs for the specific toxin should be received from the vendor upon receipt of the toxin.
- b. Toxicology textbooks, such as *Casarett's and Doull's Toxicology*, are also good sources of hazard information for some toxins.
- c. The BMBL also contains information on biological toxins and associated safe work practices.

Written Standard Operating Procedures

In combination with the 6th edition of the BMBL and *NIH Guidelines*, this manual provides a general SOP for working with biological material. However, since this manual only touches on relatively general topics, each individual laboratory must develop SOPs specific to the biosafety concerns and procedures for that particular laboratory. A laboratory specific SOP should address the safe manipulation of the pathogens present in the laboratory, specific

exposure control methods, and specific decontamination and waste handling requirements, amongst other relevant topics. There is no standard format required for these SOPs and laboratories are encouraged to use any format which effectively conveys the necessary biosafety information (including tables, pictures, and illustrations). The laboratory SOPs should complement this biosafety manual and should not duplicate the same general information.

Security and Inventory of Biological Agents

Each PI is responsible for ensuring that their laboratory implements sufficient security measures and procedures in order to protect against loss, theft, release, or unauthorized access to their laboratory and biological materials. When working with [permissible select toxin amounts](#), which are concentrations below the levels which classify them as select toxins and require federal oversight, it is critical to maintain an accurate inventory log. This log provides evidence that at no time has the amount of toxin within the laboratory been over the permissible amount. This inventory log should track the toxin from date received all the way through destruction. For more information on this topic, please contact the Office of Research.

Prevention of Aerosols and Droplets

Aerosols are liquid and solid particles suspended in the air. An aerosol with a diameter of 5 microns or less can remain airborne for a long period of time, spread wide distances, and is easily inhaled. Larger particles tend to settle more rapidly and contaminate skin, bench tops, keyboards, laboratory equipment, ventilation systems, and other surfaces within and surrounding the laboratory.

High energy procedures such as centrifuging, mixing, and pipetting can produce aerosols with diameters of 5 microns or less that stay airborne for extended periods of time, while lower energy procedures such as opening containers and streaking plates may produce larger droplets that settle quickly. Whenever aerosol producing procedures are conducted within the laboratory, a biological safety cabinet (BSC) must be utilized. A properly operating BSC will contain the aerosols and droplets that are generated during work with infectious agents.

Examples of Aerosol Producing Procedures

- a. Shaking or vortexing tubes, stirring
- b. Centrifugation steps such as filling centrifuge tubes, removing plugs or caps from tubes after centrifugation, removing supernatant, resuspending sedimented pellets, breakage of tubes during centrifugation, and centrifugation itself
- c. “Blowing out” pipettes, pipetting rapidly
- d. Cell sorters
- e. Sonication
- f. Homogenizing, blending, grinding

- g. Pouring liquids
- h. Removing gloves
- i. Necropsies
- j. Cage cleaning/changing animal bedding
- k. Opening lyophilized cultures, opening snap top tubes, breakage of culture containers
- l. Flaming loops or slides

Note: Open flames (e.g., Bunsen burners) must never be used within a biosafety cabinet as they will disrupt airflow and could potentially damage the HEPA filter and cause a fire. Contact the BSO for alternative solutions to open flames.

- m. Pulling needles out of septums, filling a syringe

Safe Work Practices to Minimize the Creation of and Exposure to Aerosols

a. Proper Utilization of Pipettes

Pipettes are used for volumetric measurements and the transfer of liquids that may contain infectious, toxic, corrosive, or radioactive agents/materials. Pipetting may lead to exposures when liquid from a pipette is dropped onto the work surface, forcefully/rapidly expelling the liquid directly into a tube, or when the last drop of an inoculum is blown out.

- Never mouth pipette. Always use a mechanical pipetting device.
- Always use filter-barrier or cotton-plugged pipette tips.
- Never forcibly expel liquid out of a pipette.
- Use “to deliver” pipettes rather than “to contain” pipettes, which require “blowout”.
- Always discharge the liquid from a pipette down the sidewall of a container, never directly into the liquid.
- Contaminated, broken, or intact Pasteur pipettes must always be discarded in a sharps container.
- Contaminated pipettes should be placed horizontally in a pan containing an appropriate disinfectant.

b. Types of Centrifuges

Centrifuges are considered high-risk equipment that can produce large amounts of aerosols due to air movement and leakiness of tubes. Centrifuges can be either bench top or floor units, they can have rotors or safety cups and buckets, and they can be low-, high-, or ultra-speed.

- Mini-centrifuges

These types are typically low or high-speed units that are only capable of holding a smaller number of tubes (e.g. less than 12). The rotors in mini-centrifuges are typically not removable and should therefore be used within a biosafety cabinet when used with infectious materials.

- Benchtop centrifuges

These units are the most common type found in laboratories. They are high-speed, can vary in size, can be refrigerated, and have either rotors or safety cups and buckets. These can be placed towards the back of a BSC for use inside the cabinet, but must be decontaminated before removed from the BSC for any reason.

If not used within a BSC, the rotor or safety cups must be removed, taken to the BSC, and the samples containing infectious material must be loaded/unloaded only within the BSC. The rotors or safety cups must be surface disinfected before removed from the BSC and they must have lids with proper seals that have been approved for use with infectious material. These seals must be thoroughly inspected for cracks and wear before and after each centrifugation.

- Floor unit centrifuges

These types of centrifuges are usually high-speed units for large volumes or ultra-speed centrifuges. For use with infectious materials, they must have rotors or safety cups with proper seals, as described above, and should ideally contain a HEPA filtered exhaust. Consider adding the HEPA filtered exhaust as an option when buying new centrifuges or for even retrofitting existing centrifuges.

Samples must be loaded and unloaded in the rotor or safety cup within the BSC. Always ensure proper balancing of tubes prior to centrifugation.

- Ultra-centrifuges

These units present an added safety issue due to the speed at which they operate.

Proper balancing of tubes is crucial in ultra-centrifuges as any difference in weight between tubes might cause severe damage to the rotor. Broken rotors could possibly penetrate through the centrifuge wall into the outside environment (laboratory).

If any suspicious noises are noticed during the operation of an ultra-centrifuge, evacuate the area and if possible, turn off the centrifuge. Notify the laboratory supervisor or EH&S of the situation. Do not attempt to open the centrifuge for at least 30 minutes in order to minimize potential exposure to any aerosols that may have been created. If a spill is noticed within the centrifuge after opening the lid, *slowly* close the lid and evacuate the laboratory. See Section XIII for more detailed information regarding spill response procedures within centrifuges.

c. *Proper Utilization of Centrifuge Equipment*

- All laboratory personnel must be trained on the proper use and specification of all centrifuges within the laboratory.
- High-speed and ultra-centrifuges must have an individual log book kept adjacent to the centrifuge where information such as rotor type, rotor run duration, speed, o-ring condition, any defects, and operator's name and date should be included.
- Always inspect tubes and bottles for cracks or stress marks before and after use.
- Inspect the gasket seal on the rotor or safety bucket before each use to use. If gaskets (e.g., O-rings) appear worn or damaged in any way (ripped, torn, etc.) they must be replaced immediately before using the equipment. Proper lubrication of threads and gaskets will help contain any leakage and may also extend the lifespan of the gaskets.
- Never overfill centrifuge tubes as leakage may occur. The recommended maximum for most centrifuge tubes is 3/4 full.

Note: Some ultra-centrifuge tubes must be filled to the top to prevent the tube from collapsing - always refer to the product literature.

- Always balance tubes, in weight and distribution, prior to spinning.
- Work within a BSC when re-suspending sedimented material. Use a swirling rotary motion rather than shaking. If shaking is necessary, wait a few minutes to permit the aerosol to settle before opening the tube.
- Always decant supernatants within the BSC. If possible and if the supernatant is not needed, use a vacuum aspirator with appropriate in-line reservoirs and filters to remove the supernatant.

d. *Inoculation Loops*

- Flaming inoculating loops can result in spatter and release of aerosols and droplets. Use of an electric incinerator will effectively control spatter resulting from sterilization of inoculating loops. Alternatively, the use of disposable loops that are discarded after each and every use can be utilized to prevent this issue.

Note: Open flames (e.g., Bunsen burners) must never be used within a biosafety cabinet as they will disrupt airflow and could potentially damage the HEPA filter and cause a fire. Contact the BSO for alternative solutions to open flames.

e. *Use of Absorbent Materials*

- Always cover work surfaces with absorbent paper to collect splashes and splatters, and to minimize the spread of contamination. The absorbent paper should be changed regularly, such as at the completion of work, always after a

spill, and at least daily.

Personal protective Equipment

Personal protective equipment (PPE) must be provided without cost to personnel. PPE is considered to be the last-line of defense against prevention of exposure in the laboratory, and should never be a substitute for the use proper engineering controls and good laboratory work practices. PPE is a primary barrier to infectious agents and their proper use, in conjunction with work practices and use of BSCs, safety centrifuge cups/rotors, and other containment devices, will reduce the likelihood of exposure.

Types of Personal Protective Equipment

a. Laboratory Coat

Laboratory coats must always be worn within the laboratory whenever handling biological or chemical reagents. They should never been worn outside of the laboratory.

There are different types of laboratory coats that can be used for specific needs:

The most common lab coat is usually made of cotton material and closes in the front. Preferably, lab coats should have tight-fitting cuffs around the wrists. These coats are reusable and must be decontaminated (e.g., washed, autoclaved) on a regular basis (e.g., weekly) or when they become soiled. Each laboratory on campus should develop decontamination procedures for their re-usable lab coats.

Disposable lab coats also close in the front, but have the benefit of being disposable and do not require lab coat decontamination procedures to be established. Disposable lab coats can be re-used, but must be discarded when they become contaminated or whenever they are torn, ripped, punctured, etc.

b. Gloves

Gloves must be selected on the basis of the hazards involved and the activities to be conducted.

Standard latex examination gloves provide protection against microbiological hazards, but do not generally provide adequate protection against certain liquid chemicals. Additionally, some people may have a latex allergy or be more prone to irritant contact dermatitis when using latex gloves. Nitrile gloves are a suitable alternative to latex gloves that provide similar dexterity, protection, and increased chemical resistance, without the latex allergy hazard.

- Gloves must always be worn when working with biohazardous and/or toxic materials and chemicals.
- When working within a biological safety cabinet with infectious materials, gloves should be removed or decontaminated prior to removal of hands from the cabinet.
- Gloves must always be removed and changed when they become contaminated, or

when they are torn, punctured, or whenever the integrity of the barrier is compromised.

- Always wash hands with soap and water after the removal of gloves and before exiting the laboratory.

When removing gloves, always do so with strict attention to avoid contact between the outside of gloves and bare skin/clothing.

Figure 10 below illustrates a glove removal technique which, when followed correctly, avoids all contact between the outside of potentially contaminated gloves and any bare skin or clothing.

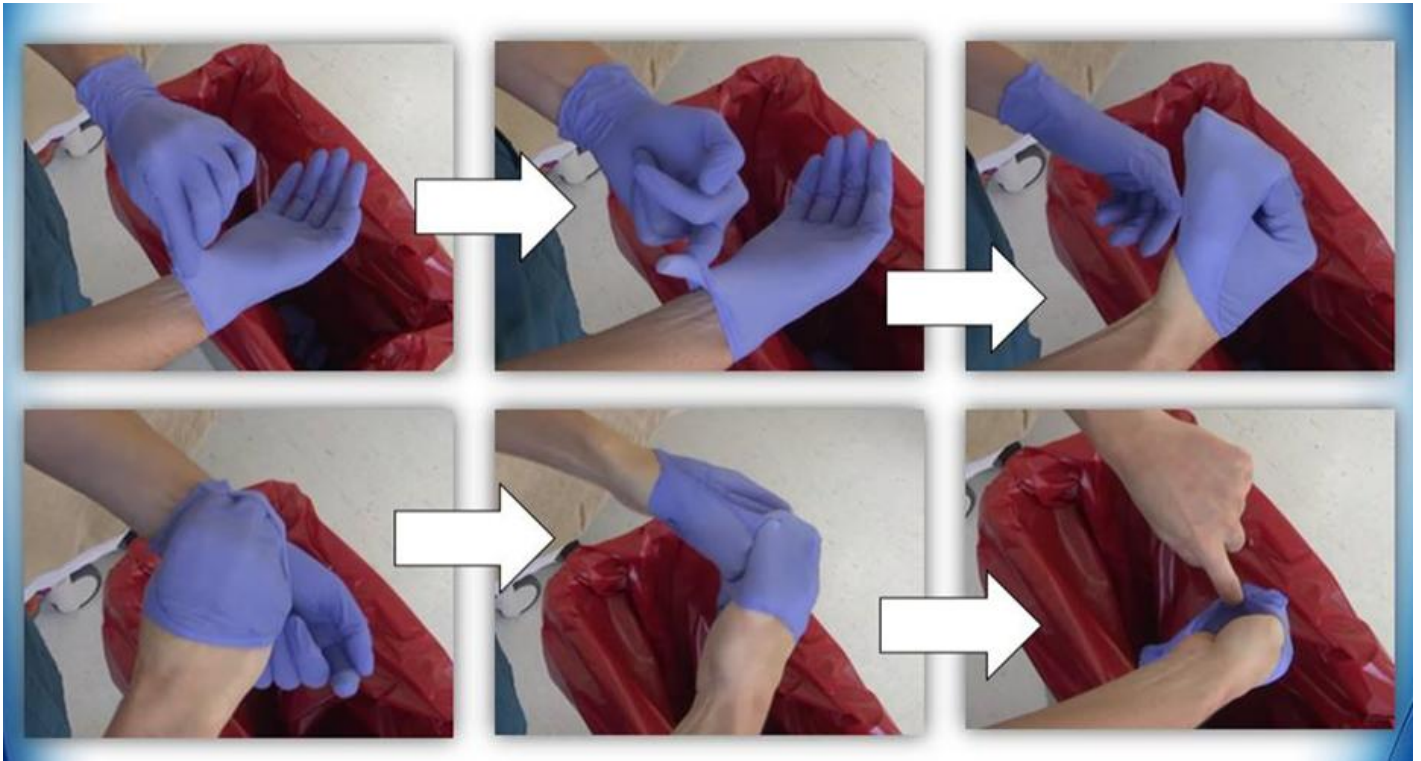


Figure 10: This image shows one technique for proper glove removal, which avoids contamination of bare skin and/or clothing.

Source: Sean Kaufman, Behavioral-Based Improvement Solutions

c. *Eye Protection*

Safety glasses or goggles are to be worn whenever there is the potential for a splash, splatter, or spray of hazardous materials. Eye protection must have solid-side shields, seeing eye-glasses are not suitable alternatives to proper safety glasses or goggles. In addition, full-face shields that cover the entire face can be utilized when splashes, sprays, or splatters are anticipated.

d. *Respiratory Protection*

Respirators are selected based on the hazard involved and the protection factor

required. Certain laboratory situations may require respiratory protection to prevent inhalation of infectious agents. Regulations, as well as good safety practice, require that personnel be medically evaluated, specifically trained, and fit tested **prior** to wearing respiratory protective equipment.

Contact EH&S if respiratory protection is required or desired, as they will ensure that all requirements are satisfied prior to permitting the use of the respirator.

e. *Clothing and Footwear Guidelines*

Shorts or other clothing that exposes the legs and open-toed shoes or sandals are not proper laboratory attire and must never be worn in the laboratory. Good laboratory practices require that personal clothing or the equivalent, such as scrubs, should cover all exposed skin. It is also important to avoid loose, dangling jewelry that may get caught in equipment or make incidental contact with hazardous materials.

Storage and Labeling of Biological Agents

Biological agents must be stored using leak proof and sealed containers. These containers must be clearly labeled with the identity of the agent. It is recommended that all samples be stored in secondary containers to help prevent leakage. Secondary containers should list the identities of the agents inside and if applicable (see below), the universal biohazard symbol. All laboratories must have proper laboratory door signage on all entry doors. The information contained on these signs includes:

- The name of the PI and any alternate individuals, if applicable, and their contact information
- The hazards associated with the laboratory
- Laboratory specific information for emergency responders

The door signs must be requested through EH&S via the [Laboratory Door Sign Request form](#)

Universal Biohazard Symbol

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



As mentioned above, each laboratory must have a sign at the entrance that provides safety information to visitors, service personnel, and emergency responders. Biohazard signs must be posted at the following:

- a. Entrances to all laboratories and animal rooms that are classified as BSL-2 or ABSL-2
- b. Cages and animal rooms used for housing animals infected with human cells or tissues, risk group 2 or 3 agents, or biological toxins.

Within each of these areas, pieces of equipment and certain waste items will also require to be labeled with the biohazard symbol. Refrigerators, freezers, and any equipment or storage area requires the biohazard symbol whenever they are used to store human source materials, OPIM, risk group 2 or 3 agents, and/or biological toxins. Laboratory equipment where these types of materials are used or processed must also be labeled (e.g., centrifuges, microwaves, incubators, biosafety cabinets, water baths, etc.).

Personal items (e.g., food and drinks), are absolutely prohibited in laboratory refrigerators, cold rooms, freezers, or incubators.

IX. Laboratory Training

All laboratory workers (faculty, staff, students, volunteers, and visitors) must be properly trained for working in the laboratory environment. The specific training required for each particular individual will depend on the hazards to which they are exposed. It is the responsibility of the Principal Investigator to ensure that all personnel receive training that is appropriate for their job duties and exposure potential. Anyone who is to work in any ODU laboratory must complete the ODU Laboratory Safety training offered through EH&S. The list of training courses offered by EH&S can be found on their [Safety Training website](#).

As stated above, the PI is ultimately responsible for ensuring each member of their laboratory staff is adequately trained to safely work with the hazards associated with the laboratory. Safety training consists of both online and in-person instructional training but must also involve laboratory-specific hands-on training that is to be conducted by the PI or a competent member of the laboratory (see below). The online training is completed through the Collaborative Institutional Training Initiative (CITI), and instructions on how to create an account and register for courses can be found under the “Forms and Templates” section of IRBNet. A list of training courses offered through CITI and EH&S can be found below:

a. Laboratory Safety (In-person or Zoom)

This course is offered through Environmental Health and Safety (EHS) and is mandatory for employees/TAs/GAs who work with chemicals in laboratories. The training involves classroom lecture, a self-study workbook and graded quiz, and on the job training conducted by the PI which must be laboratory specific. For more information, refer to the [ODU Safety Training](#) webpage. This training is only required initially.

b. Basic Biosafety (CITI)

This course must be completed by all individuals listed on any approved IBC protocol. Completion of this course is required every three (3) years.

c. OSHA Bloodborne Pathogens (VectorSolutions and CITI)

This course must be completed by all individuals listed on an approved IBC protocol if the protocol involves human blood, blood products, or other potentially infectious materials (OPIM) covered by the [OSHA Bloodborne Pathogen Standard](#). Bloodborne Pathogen (BBP) training through Environmental Health and Safety or CITI is mandatory for all employees who come in contact with blood or OPIM during the course of their work. It is the position of the CDC and OSHA that all cell lines of human origin are considered potentially infected with bloodborne pathogens, and these materials are to be handled using a minimum of BSL-2 containment and procedures. All persons working with human and non-human primate cell lines are therefore required to complete BBP training before beginning work and on an annual basis. For more information, refer to the [ODU Safety Training](#) webpage. The CITI aspect of this training must be completed annually.

d. *NIH Recombinant DNA (rDNA) Guidelines (CITI)*

This course must be completed by all individuals listed on an approved IBC protocol if the protocol involves rsNA work. Completion of this course is required every three (3) years.

e. *Additional CITI Training Courses*

Depending on the work to be conducted, CITI offers additional training courses which may be required before a protocol will be approved by the IBC. These additional courses include:

- Nanotechnology
- Animal Biosafety
- Shipping and Transport of Regulated Biological Materials

*Note: Completion of this training **does not** certify an individual to ship or transport any biological materials. Before shipping any materials, EH&S must be notified and will work with the individual to assist in preparing the item(s) for shipment in accordance with all local, state, and federal regulations.*

- Select Agents, Biosecurity, and Bioterrorism

Note: Although there may be no usage, storage, or transportation of select agents or toxins at ODU, those laboratories working with exempt strains or toxins below the permissible level will be required to complete this training.

- Emergency and Incident Response to Biohazard Spills and Releases

Laboratory-specific Training

Individual laboratories must develop specific training for the particular agents and procedures that personnel will perform in that laboratory. This training must be specific to the hazards in the laboratory and to each person's laboratory duties. Each person must understand the hazards associated with the agent, signs and symptoms of infection, laboratory operations, spill response procedures, how to prevent exposures to biological and chemical agents, and be trained on the laboratory standard operating procedures. Laboratory-specific training should be designed to supplement the general laboratory training course, not duplicate it.

Each laboratory should maintain training records which include the names of personnel in the laboratory and their most recent dates of laboratory safety training, including CITI, EH&S offered training, and the laboratory specific training offered by the PI or other competent laboratory member. Training records are also kept electronically by the Biosafety Officer in the Office of Research for all individuals listed on an approved protocol.

To avoid complacency in the laboratory, training should be a continuous process for all members of the laboratory. As new hazards and procedures are introduced into the laboratory, all individuals must receive an update to their training.

X. Decontamination and Sterilization

Definitions

Decontamination

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

Sterilization

The use of a physical or chemical procedure to destroy all microbial life, including highly resistant bacterial endospores. However, sterilization may not destroy prions.

Disinfection

A less lethal process than sterilization which eliminates nearly all recognized pathogenic microorganisms, but not necessarily all microbial forms on inanimate objects.

Effectiveness is influenced by the kinds and numbers of organisms, the amount of organic matter, and the object to be disinfected and chemical exposure time, temperature, and concentration.

Types of Decontamination

Decontamination of cultures and objects contaminated by biological agents must be routinely performed within microbiological laboratories. This is a vital component of laboratory safety practice and serves to protect laboratory personnel, as well as others, from infection, and prevents the release of infectious organisms to the outside environment. The reduction of cross-contamination in the laboratory is an added benefit of decontamination.

Depending on the circumstances and tasks, decontamination of a surface (e.g., lab bench) is accomplished with a disinfectant, while decontamination of biomedical waste is done by sterilization in an autoclave.

In order to properly select an appropriate method and tools for decontamination, it is important to consider the following aspects:

- a. Type of biohazardous agents, concentration, and potential for exposure
- b. Physical and chemical hazards to personnel, products, materials, and environment

Physical and chemical means of decontamination fall into four main categories: heat, liquid chemicals, vapors and gases, and radiation.

Disinfection is typically accomplished by applying a liquid chemical or wet heat during boiling or pasteurization. To sterilize, vapors, gases, radiation, and wet heat (steam

sterilization in an autoclave) are used. Some liquid chemicals can be used for sterilization, if used in the right concentration for the right amount of time.

Methods of Decontamination

1. Heat

In order to kill microbial agents, heat can be applied in either dry or wet form. The advantage of wet heat is a better heat transfer to and into the cell, which results in an overall shorter exposure time and lower temperature. Steam sterilization (e.g., autoclave) uses pressurized steam at 121-132°C for 30 to 90 minutes. This type of heat kills all microbial cells including spores, which are normally heat resistant. In order to accomplish the same effect with dry heat in an oven, the temperature needs to be increased to 160-170°C for periods of 2 to 4 hours.

Only autoclaves authorized by EH&S are permitted to be used for decontamination /treatment of waste (bagged or liquid)..

2. Liquid Chemicals and Disinfectants

The appropriate liquid disinfectant must be carefully selected by assessing the biohazardous agent(s) in use within the laboratory, and the type of material(s) to be decontaminated. Liquid disinfectants are preferred for routine decontamination of solid surfaces and equipment, but they vary greatly in their efficiency depending on the chemical constituents and the agents involved. When selecting a liquid disinfectant, consider the following:

a. Nature of surface being disinfected

Porous or smooth, the more porous and rough the surface, a longer contact time with the disinfectant will be needed in order to be effective.

b. Number of microorganisms present

Higher concentrations require a longer application time and/or higher concentration of the disinfectant.

c. Resistance of microorganisms

Microbial agents can be classified according to increase resistance to disinfectants and heat (see Table 5)

d. Presence of organic material

The proteins in organic materials such as blood, bodily fluids, and tissue can prevent or slow the activity of certain disinfectants.

e. Duration of exposure and temperature

Increased exposure time increases the effectiveness of disinfectants. Low temperature may slow down the activity requiring more exposure time.

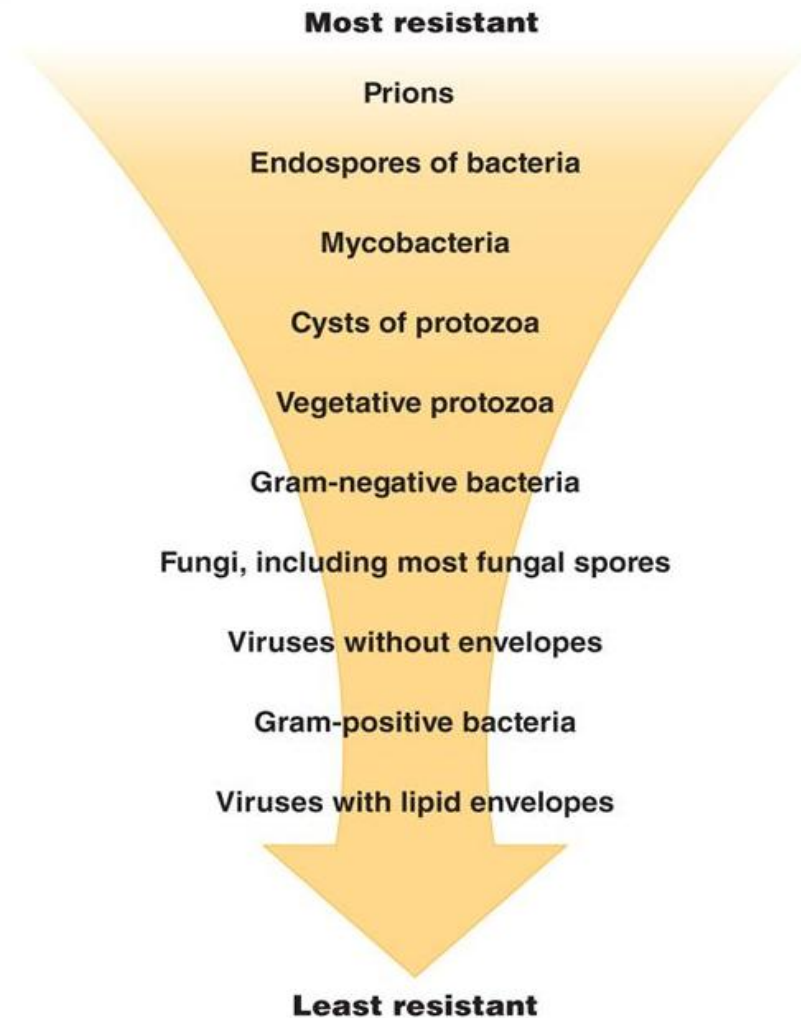


Figure 10: Decreasing order of resistance of microorganisms to chemical disinfectants

Tortora, G., Funke, B., Case, C. Pearson Education, Inc. *Microbiology: An Introduction*. 11th edition.

Figure 7.11: Decreasing order of resistance of microorganisms to chemical biocides. 2013.

There are many types of liquid disinfectants available under a variety of trade names. In general, these can be categorized as halogens, acids or alkalines, heavy metal salts, quaternary ammonium compounds, aldehydes, ketones, alcohols, and amines.

Unfortunately, the most effective disinfectants are often very aggressive (corrosive) and toxic. Some of the most commonly used disinfectants are described in this section and Table 6 provides a summary of the chemical disinfectants mentioned below.

a. *Alcohols*

Ethyl or isopropyl alcohol in concentrations of 70% to 90% are good general-use disinfectants. However, they evaporate fast and therefore have limited exposure time. They are less active against non-lipid viruses and ineffective against bacterial spores. Concentrations above 90% are less effective.

b. *Formalin*

Formalin is a 37% by weight of formaldehyde mixed with water and 10-12% methanol, which acts as a stabilizer. Dilution of formalin to 5% results in an effective disinfectant. Formaldehyde is a human carcinogen and creates respiratory problems at low levels of concentration.

c. *Glutaraldehyde*

This compound is chemically related to formaldehyde and is more effective against all types of bacteria, fungi, and viruses. Vapors of glutaraldehyde are irritating to the eyes, nasal passages, and upper respiratory tract. Glutaraldehyde should always be used in accordance with the instructions on the label and the appropriate PPE must always be worn.

d. *Chlorine Compounds*

Chlorine-containing solutions have broad-spectrum activity. Sodium hypochlorite is the most common base for chlorine disinfectants. Common household bleach is usually around 5% sodium hypochlorite (50,000ppm) and needs to be diluted 1/10 with water to yield a satisfactory disinfectant solution. Once diluted, it is recommended to make a fresh solution daily. If the diluted solution is in a closed container and protected from light, it will remain effective for a longer period of time. Excess organic materials inactivate chlorine-containing disinfectants.

Chlorine-containing disinfectants are strong oxidizers and very corrosive, therefore, appropriate PPE must be worn at all times while handling these chemicals. At high concentrations and extended contact times, hypochlorite solutions are considered cold sterilants since they inactivate bacterial spores.

Note: If using sodium hypochlorite, or other corrosive chemical, to disinfect the surfaces of a biosafety cabinet, or other metal items, always rinse with water and/or a 70% alcohol solution to remove any residual sodium hypochlorite which will rust and pit the metal.

e. *Iodophors*

Iodophors are compounds containing a combination of iodine and a solubilizing agent or carrier. The best-known and most widely used iodophor is povidone-iodine. An example of this type of disinfectant is Wescodyne®, which is a concentrated broad spectrum iodophor labeled as a one-step cleaner-disinfectant and no-rinse sanitizer. These disinfectants retain the germicidal efficacy of iodine but are generally non-staining and relatively free of toxicity or irritancy. Manufacturers' data demonstrate that commercial iodophors are not sporicidal, but they are tuberculocidal, fungicidal, virucidal, and bactericidal at their recommended use-dilution.

f. *Phenol and Phenol Derivatives*

Phenol based disinfectants come in various concentrations ranging mostly from 5% to

10%. Phenol itself is toxic and appropriate PPE must be worn when these types of disinfectants are being used. Phenolic disinfectants are used frequently for disinfection of contaminated surfaces and are effective against *Mycobacterium tuberculosis*, fungi, and lipid-containing viruses. They are not effective against spores or non-lipid viruses.

g. *Quaternary Ammonium Compounds*

Quaternary ammonium compounds (quats) are cationic detergents with strong surface activity, and are relatively non-toxic. They are acceptable for general-use disinfectants and are effective against gram-positive bacteria and lipid-containing viruses. However, they are not as effective against gram-negative bacteria and are not active against non-lipid-containing viruses. If quats are mixed with phenols, they are very effective disinfectants as well as cleaners.

3. Vapors and Gases

A variety of vapors and gases possess germicidal properties. The most commonly used are formaldehyde and ethylene oxide. Applied in a closed system under controlled conditions (e.g., humidity) these gases achieve sterility.

Vaporized hydrogen peroxide (VHP) is an effective sterilant capable of efficient and rapid elimination of contaminating organisms with a broad range of antimicrobial applications. Since this is a vapor, and not a gas, it does not achieve great penetration into tight spaces such as cracks and crevices within the space being decontaminated. However, this feature also makes it easier to contain the vapor to the designated area without it escaping to unwanted, potentially occupied areas.

Chlorine dioxide gas is an effective broad-spectrum, anti-inflammatory, bactericidal, fungicidal, and virucidal agent that is a sterilant as it will also inactivate bacterial endospores. Chlorine dioxide is much more effective at penetrating into tight spaces and is considered to provide a more thorough decontamination than VHP.

Both VHP and chlorine dioxide are oxidizers that are toxic and corrosive that require specialized equipment and training before using. If this level of decontamination is needed, it is typically recommended to hire a professional.

4. Radiation

Gamma and X-ray are two principal types of ionizing radiation used in sterilization. Their application is mainly centered on the sterilization of prepackaged medical devices.

Ultraviolet (UV) radiation is a practical method for inactivating viruses, mycoplasma, bacteria, and fungi; however, its capabilities are limited and its use as a method of decontamination, especially within biosafety cabinets, is not recommended.

For further information, refer to the *Guideline for Disinfection and Sterilization in Healthcare Facilities, 2008* (CDC).

Table 6: Summary of Chemical Disinfectants									
Disinfectant	Use parameters		Effective against ^a			Hydrophilic viruses	Bacterial spores	Important characteristics	Potential application
			Vegetative cells	Lipophilic viruses	Tubercle bacilli				
Alcohol	Conc.: 70-90%	Contact time: 10-30 min.	+	+	+	±		Eye irritant, toxic, flammable, inactivated by organic matter.	Surfaces: work and equipment
Chlorine compounds	Conc.: 0.05-0.5% Contact time: 10-60 min.		+	+	+	+	±	may leave residue; corrosive; skin, eye & respiratory irritant; inactivated by organic matter; makeup at least weekly	spills, equipment surfaces, instruments, glassware, water baths
Quaternary ammonium compounds	conc.: 0.1-2% contact time: 10-30 min.		+	+				toxic, inactivated by organic matter	surfaces (work & equip.), BSCs, floor maintenance, glassware, instruments
Phenolic compounds	conc.: 0.2-3% contact time: 10-30 min.		+	+	+	±		leaves residue; corrosive, skin, eye & respiratory irritant; toxic; inactivated by organic matter	surfaces (work & equip.), BSCs, floors, spills, glassware, instruments, water baths
Iodophor compounds	conc.: 0.47% contact time: 10-30 min.		+	+	+	±		leaves residue; corrosive, skin & eye irritant; toxic; inactivated by organic matter	surfaces (work & equip.), BSCs, glassware, water baths
Formaldehyde ^b (Formalin)	conc.: 4-8% contact time: 10-30 min.		+	+	+	+	±	leaves residue; skin, eye & respiratory irritant; toxic (carcinogen)	less effective than other disinfectants but can be used for equipment surfaces, glassware, instruments
Glutaraldehyde	conc.: 2% contact time: 10-60 min.		+	+	+	+	+	leaves residue; skin, eye & respiratory irritant; toxic	equipment surfaces, glassware, instruments

Source: Diane O. Fleming, John H. Richardson, Jerry J Tulis, and Donald Vesley, eds. *Laboratory Safety: Principles and Practices*, second

edition. American Society for Microbiology. Washington, D.C. 1995.

a: + = very positive response, $\pm$ = less positive response. A blank denotes a negative response or not applicable.

b: due to its irritating characteristics and status as a carcinogen, formaldehyde should not be used without good local exhaust ventilation.

XI. Incident Response

Personnel Exposures

Exposure to a biological agent, including recombinant or synthetic nucleic acid molecules, is considered an emergency and there must be immediate actions taken in order to help prevent an infection from occurring. When an exposure to a biological agent occurs in the absence of physical injury, the emphasis in responding to the incident is on the decontamination of affected persons to minimize the likelihood of infection. Rapidly identifying the route of exposure and the agent or toxin that the individual was exposed to is critical to minimizing the risk of infection. The effectiveness of post-exposure prophylactic measures decreases with time, therefore, it is recommended that a medical evaluation be obtained as soon as possible (within hours, not days) following all exposure incidents.

Individuals working in laboratories with lacerations or broken skin should not work with biohazardous materials unless the injury is completely protected, and they should receive permission from the PI prior to working in the lab. Those with compromised immune systems should self-identify, see #4 below for more details.

ODU Environmental Health & Safety number: 757-683-4495

Exposure First Aid Procedures

a. Percutaneous Inoculation

Examples: needle-stick, cuts or abrasions from contaminated items, animal bites, or other event that breaks the skin and introduces an agent or toxin into the body)

Wash the affected area thoroughly with soap and water and if bleeding, attempt to squeeze blood out of the wound while washing. If available, povidone-iodine, chlorhexidine, or 70% isopropyl alcohol may be used to cleanse the area, but these disinfectants have not been shown to be any more effective than soap and water. Immediately contact laboratory supervisor and EH&S.

b. Intact Skin Exposure

When an agent or toxin splashes or splatters onto intact skin, washing the affected area with soap and water will be sufficient to decontaminate the area. If the splash or splatter occurs onto non-intact skin, which includes sunburns, chapped lips, cracked skin, cuts, abrasions, etc., immediately contact the laboratory supervisor and EH&S.

c. Mucous Membrane Exposure (eyes, nose, and mouth)

Flood affected areas thoroughly with running water using an eye wash, sink faucet, or any other means available. Continue to rinse the area for a minimum of 10 minutes. If ingestion or contact with the mouth or oral cavity occurs, the mouth should be rinsed thoroughly several times using water or antiseptic mouthwash, if available. Immediately report the situation to the laboratory supervisor and EH&S.

d. *Inhalation*

If inhalation of infectious aerosols is suspected, there is no specific first-aid response beyond decontamination of exposed areas. Immediately contact the laboratory supervisor and EH&S for further investigation and to determine if a medical consultation is required.

Exposure Response Procedure

- a. Alert others in the immediate area that a known or potential exposure incident has occurred so that someone may assist in providing the first aid procedures, as described above.
- b. While the first-aid is being administered, have someone notify the laboratory supervisor and EH&S. If no one is around to assist, wait until after first-aid has been self-administered before notifying the appropriate personnel.
- c. Remove contaminated clothing as required, if a large area of skin was exposed, use the chemical shower to rinse the affected areas. All PIs should have spare clothing such as t-shirts and sweatpants in various sizes.

Note: All contaminated clothing must be thoroughly decontaminated or discarded as biological waste. All areas surrounding the incident must be surface decontaminated according to the procedures listed in Chapter XIII.

- d. Seek medical consultation and evaluation according to the procedure below.

Note: It is not mandatory to seek post-exposure medical care, however, it is strongly encouraged. As mentioned above, it is recommended that medical care is provided within two hours following an exposure.

Medical Care and Consultation

The University's contracted occupational health care provider is NowCare Medical Center. Following an exposure to blood or other potentially infectious materials, or to any biological agent or toxin, where medical attention is being sought, follow the steps below:

- a. After performing on-site first aid as described above, call NowCare (757-424-4300 OR 757-587-1700) to let them know you will be coming in for post-exposure medical care and consultation. Be sure to tell them that you are an ODU employee.
- b. Bring the [BBP-2](#) form to NowCare which will need to be completed and signed by the attending physician, as well as the affected employee. This form can also be found in Appendix E of ODU's BBP Exposure Control Plan, and within the [Forms and Pamphlets](#) section of the ODU EH&S website.
- c. Following the medical exam, forward a copy of the completed BBP-2 form to EH&S via campus mail or fax (757-683-6025).

Note: If you choose to not seek medical care, you must still complete the BBP-2 form

and select “Employee refuses post-exposure medical care”. Complete remaining applicable portions of the form (name, UIN#, and signature). If you choose not to seek immediate medical care, you may do so anytime thereafter.

- d. As soon as possible following the exposure, complete the [BBP-1](#) form. This form can also be found in Appendix E of ODU’s BBP Exposure Control Plan, and within the [Forms and Pamphlets](#) section of the ODU EH&S website. Once complete, forward a copy of the completed BBP-1 form to EH&S via campus mail or fax (757-683-6025).
- e. Contact the University’s [Worker’s Compensation](#) representative at 757-683-3051 to file a report of injury.

For an exposure incident that occurs after hours, refer to the instructions below to ensure appropriate medical attention is received:

- a. Immediately contact your supervisor and EH&S at 757-683-4495
- b. Contact a NowCare medical provider at 757-424-4300. NowCare will provide you with instructions as to which actions you should take next.
- c. Complete both BBP-2 and BBP-1 forms as previously instructed.

The NowCare medical office can be found at the addresses below.

NOWCARE
(0.3 Mile from Military Hwy)
6632 Indian River Road
VA. Beach, VA 23464
Phone: 424-4300
Hours of Operation:
M-F 8:00 AM - 8:00 PM
Sat. 9:00 AM - 3:00 PM

[Bloodborne Pathogens Post-Exposure Brochure](#) is available for download by clicking on the link provided or through the [ODU EH&S website](#).

Guidelines for Handling of Biological Agents by Immunodeficient or Immunosuppressed Employees

When an employee’s immune system is impaired by a condition such as those specified below, the risk of infection by biological agents increases. This results in many human or animal pathogens, which typically present little or no threat to a healthy individual, becoming a significant safety hazard. All individuals with any condition that may adversely affect the immune system are **strongly encouraged** to self-identify to their supervisors, EH&S, or student health. These conditions include, but are not limited to:

- a. Treatment with cytotoxic chemotherapeutic agents

- b. Treatment with adrenocorticosteroids
- c. Treatment with immunosuppressive agents/drugs or certain antibiotics
- d. Disease processes that suppress the immune system
- e. Pregnancy

Any individual who is aware of being immunodeficient/immunosuppressed are able to receive an appropriate medical evaluation and all communications are confidential.

These individuals should be counseled as to the advisability of working in areas where the potential for exposure to potentially hazardous organisms is present. Any limitations or restrictions relating specifically to the immunodeficient/immunosuppressed condition and laboratory work shall be reported to the employee's supervisor.

XII. Spill Response Procedures

The hazard associated with a biological spill is a function of the volume of the spill, the pathogenicity of the agent, and its concentration within the spilled material. When a spill occurs, the appropriate response should consider the protection of employees; preventing release of viable biological agents outside of the laboratory environment; and cleanup and disinfection of the area. A minor biological spill is one that the laboratory staff is capable of handling safely without any assistance from EH&S and/or the Biosafety Officer. All other biological spills are considered major and EH&S and/or the Biosafety Officer should be notified to assist.

Since biological spills will happen within the laboratory environment, it is important that each laboratory on campus be prepared to handle the situation. Laboratories working with biohazards must have a basic biological spill kit ready to use at all times. For most instances, the basic kit can be assembled from items already available within the laboratory. It is preferred to have the contents of the spill kit all in one location, such as within a dedicated bucket or drawer, where it is easily accessible to everyone in the lab, prior assembly is not always necessary. Each spill kit should contain the following items:

- Appropriate disinfectant for the biohazardous agents present in the laboratory
- Absorbent Material, such as paper towels
- Waste container (e.g., biohazard bags and sharps containers)
- PPE (e.g., extra lab coat, gloves, eye, and face protection)
- Disposable brush and dustpan or tongs/forceps for picking up broken glass

Alternatively, pre-assembled spill kits may be purchased online.

Biological Spill inside Laboratory

- a. Clear spill area of all personnel, remove PPE and exit the area.
- b. Call EH&S and laboratory supervisor to report the spill
- c. Wait 30 minutes for any aerosols to settle before re-entering the spill area.
- d. Upon re-entering the laboratory, don appropriate PPE (e.g., lab coat (preferably disposable), safety goggles or glasses, and gloves) and obtain the biological spill kit.
- e. Cover the spill entirely with absorbent materials (e.g., paper towels)
- f. Encircle the spill with an appropriate disinfectant, starting at the outside of the spill area and moving toward the center. Completely saturate the absorbent material with disinfectant and allow for a 30 minute contact time.
- g. Remove and discard absorbent material and dispose of into a biohazard bag.
- h. Remove any broken glassware with forceps, tongs, or broom and dustpan (preferably disposable), and dispose in designated sharps container. Never pick up contaminated

sharp objects with your hands.

- i. Decontaminate and remove all items within the spill area.
- j. Re-apply disinfectant to the spill area and wipe down the area, including nearby equipment.
- k. Dispose of all materials used during spill clean-up, including absorbent materials and PPE, into a red biohazard bag for autoclaving.
- l. Any contaminated re-usable items should either be chemically decontaminated or sent to the autoclave.
- m. Allow others to return to the area once the spill clean-up and decontamination is complete.

Biological Spill inside Biosafety Cabinet

- a. Assess the possibility that the spill splashed to the outside of the biosafety cabinet. If yes, then follow the spill protocol procedures outlined above for “Biological Spill inside BSL-2 laboratory”. If no, then follow contained spill protocol for biosafety cabinet work surface outlined below.
- b. If the spill leaked under the work surface of the cabinet, verify that the drain valve is closed and pour a small amount of disinfectant through front grill and allow for a 30 minute contact time.
- c. Do not turn off the cabinet or close the sash.
- d. Soak up the spill with absorbent material and pour disinfectant starting at the outside of the spill area and moving toward the center. Completely saturate the absorbent material and allow for a 30 minute contact time.
- e. After allowing for at least a 30 minute contact time, remove and discard the absorbent material and dispose into a biohazard bag within the biosafety cabinet.
- f. Decontaminate all interior surfaces of the BSC (side walls, back wall, working surface, underneath working surface) and any equipment within the cabinet.
- g. Note: Never place your head inside of a BSC in order to reach the back wall, or for any other reasons.
- h. Discard PPE worn during clean-up and place in a biohazard bag for decontamination (e.g., reusable lab coat for autoclaving) or disposal.
- i. Allow the cabinet to run for at least 10 minutes after clean-up and before resuming work.
- j. Inform the laboratory supervisor and BSC users of the spill and clean-up procedures conducted.

Biological Spill inside a Centrifuge

- a. If a spill is noticed to have occurred during centrifugation (e.g. sound of breaking glass tubes), DO NOT attempt to open the centrifuge after the cycle has completed. Notify personnel in the laboratory to evacuate and wait 30 minutes before attempting to clean-up the spill.
- b. If the spill is noticed upon opening the centrifuge, slowly close the lid and notify personnel to evacuate. Wait 30 minutes before attempting to clean-up the spill.
- c. Notify EH&S and the laboratory supervisor immediately.
- d. Don appropriate PPE (lab coat, safety goggles or glasses, gloves, and respiratory protection, if appropriate and certified to wear one)
- e. Slowly open centrifuge and remove rotor and buckets/safety cups and take them to the nearest biosafety cabinet for decontamination. If rotor cannot be removed, decontaminate in place by soaking absorbent materials with disinfectant and cover the rotor's entire surface area. Pour disinfectant into the wells of the rotor. Allow for a 30 minute contact time.
- f. Lay absorbent materials in the bottom of the centrifuge and pour disinfectant.
- g. Saturate additional absorbent material with appropriate disinfectant and stick them to the entire interior of the centrifuge. Allow for a 30 minute contact time.
- h. Use tongs or forceps to remove any broken glass. Discard glass or other sharp objects into a sharps container.
- i. Wipe down the interior of the centrifuge and discard all material as biohazardous waste.
- j. Wipe down the rotor and discard all material into the biohazard bag inside of the BSC (if the rotor was removable and decontaminated inside the BSC).

NOTE: Always inspect tubes for cracks and stress marks before centrifuging. Check rotors and seals, specifically the "o" rings on sealed rotors. Ensure all tubes are rated properly for the desired speed at which they will be centrifuged.

Biological Spill inside an Incubator

- a. If a spill has occurred inside an incubator that is contained within the incubator, slowly close the door and notify the laboratory supervisor and EH&S of the spill.
- b. If the spill has leaked outside of the incubator into the open laboratory, notify others in the laboratory to evacuate for at least 30 minutes and call the laboratory supervisor and EH&S.
- c. Before re-entering the lab, don appropriate PPE (lab coat, safety goggles or glasses, gloves, and respiratory protection, if appropriate and certified to wear one).

- d. If there is a water pan inside incubator, pour bleach into the pan to disinfect the water. Allow for a 30 minute contact time.
- e. Remove all materials from the incubator and decontaminate with an appropriate disinfectant.
- f. Wipe down all interior and exterior surfaces of the incubator with disinfectant.
- g. Pour disinfected water in water tray down the sink, wipe down with disinfectant, add clean water, and return to the incubator.
- h. Dispose of all materials used during the spill clean-up into a biohazard bag.

Biological Spill outside the Laboratory (e.g., during transport)

- a. Should a spill of biohazardous material occur outside the laboratory in a public area, immediately contact EH&S. Do not attempt to clean-up the spill without the proper personal protective equipment and spill response materials.
- b. Always transport biohazardous materials in accordance with packaging and transportation of biological materials on and off site as noted in the Section IV.

XIII. Biohazardous Waste

Management of Regulated Medical Waste

The [Virginia Department of Environmental Quality](#) (DEQ) governs biohazardous waste, termed regulated medical waste, through [regulated medical waste management regulations](#) (9VAC20-121). These regulations define regulated medical waste (RMW) as:

- Cultures and stock of microorganisms and biologicals. Discarded cultures, stocks, specimens, vaccines and associated items likely to have been contaminated by them are regulated medical wastes if they are likely to contain organisms likely to be pathogenic to healthy humans. Discarded etiologic agents are regulated medical waste. Wastes from the production of biologicals and antibiotics likely to have been contaminated by organisms likely to be pathogenic to healthy humans are regulated medical wastes.
- Human blood and human body fluids. Wastes consisting of human blood or human body fluids or items contaminated with human blood or human body fluids.
- Tissues and other anatomical wastes. All human anatomical wastes and all wastes that are human tissues, organs, or body parts are regulated medical waste.
- Sharps. Sharps likely to be contaminated with organisms that are pathogenic to healthy humans, and all needles, syringes with attached needles, suture needles, and scalpels are regulated medical wastes. This includes sharps generated through veterinary practice.
- Animal carcasses, body parts, bedding and related wastes. When animals are intentionally infected with organisms likely to be pathogenic to healthy humans for the purposes of research, in vivo testing, production of biological materials or any other reason; the animal carcasses, body parts, bedding material and all other wastes likely to have been contaminated are regulated medical wastes when discarded, disposed of or placed in accumulated storage.
- Any residue or contaminated soil, water, or other debris resulting from the cleanup of a spill of any regulated medical waste.
- Any solid waste contaminated by or mixed with regulated medical waste.

Proper handling and disposal of RMW is necessary to prevent infection of personnel (laboratory workers, custodians, laboratory visitors, etc.) and release to the environment. OSHA and Virginia DEQ regulations require that RMW be properly labeled, stored, and disposed of.

Labeling Requirements

At a minimum, all RMW must be clearly labeled with the universal biohazard symbol. Additional information, such as the type of waste (e.g., sharps, liquid waste) and origin of the waste, is recommended, along with the following required information:

- a. Generator's name
- b. Building and room number

Handling and Disposal of Regulated Medical Waste

Guidelines for Disposal of Regulated Medical Waste

Below are the guidelines for disposal of Regulated Medical Waste at approved locations (PDF), be sure to review prior to dropping off waste:

- [BSSF Medical Waste Guidelines](#)
- [Chemistry Medical Waste Guidelines](#)
- [IRP / CBE Medical Waste Guidelines](#) (No autoclave)
- [College of Health Sciences Medical Waste Guidelines](#)

Sharps

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

All sharps must be submitted in approved sharps containers and sealed (lids have locking tabs). Sharps containers must be sealed closed when they are 3/4 full; never fill a sharps container to the top.

The containers must be clearly labeled **prior** to the submission to the Biological Sciences Support Facility (BSSF), Health Sciences and Chemistry autoclaves with the following information:

- a. Generator's name
- b. Building and room number

Once the sharps have been autoclaved within the BSSF/Health Sciences/Chemistry autoclaves, they will be sent off campus to a contracted vendor for final disposal.

Approved sharps containers are rigid, leak-proof, puncture resistant boxes of various sizes (**Maximum size** not to exceed 14 quarts) with a lid that can be securely sealed, and which are clearly marked with the biohazard symbol. Examples of sharps containers can be found on the [ODU EH&S Regulated Medical Waste Guidelines](#) webpage.

Contaminated / Uncontaminated Laboratory Glassware and Broken Glass

- Collect contaminated glass/slides into an approved sharps containers and dispose

of through approved Autoclave location or vendor provided container (IRPII).

- Collect uncontaminated laboratory glassware and broken glass in safe, puncture resistant receptacles (e.g. rigid cardboard box lined with polypropylene bag) that is separate from other waste which will prevent cuts and punctures to personnel. Containers should be labeled “broken glass”, which can be disposed of as ordinary trash.

Plastic Pipettes (Serological Pipettes)

Pipette collection:

- Collect Plastic Pipettes in a leakproof, Poly container, double-bagged (Examples: Pipette washers tubes, 5-gallon buckets or Small trash can)
 - not mixed with other solid wastes
- When the bags are full, remove bags, form a “log/Roll shape”, close bags and place into a sterilization tray and dispose of at an approved autoclave location or Vendor provided waste cart (IRPII).
- Prior to disposal of bagged pipettes, be sure the bags are marked as follows:
 - PI’s name
 - Room Number

Pipettes and will be accepted for treatment per location schedule:

- BSSF: schedule waste drop-off
- CHEM: availability of Autoclave
- Health Sci: Discretion of Autoclave operator

Solid Regulated Medical Waste

Solid RMW includes microbial agents, tissue culture, and other contaminated material such as cultures, pipettes, gloves, and any other item falling under the RMW definition listed above. These materials must be collected in double red, autoclavable biohazard bags (*serological pipettes must be bagged separately from any other solid waste*) that are labeled with the universal biohazard symbol. References to autoclavable bags can be found on the [ODU EH&S Regulated Medical Waste Guidelines](#) webpage.

Unauthorized Material

The following materials should never be autoclaved – Unauthorized Material:

- Flammable, reactive, corrosive, toxic, even if mixed with biological waste (not including small spills/general disinfecting)
- Solvents or volatiles
- Radioactive materials
- Non-absorbed Bleach or any liquids containing bleach (sodium hypochlorite)
- Mixed waste materials (Radiological, Chemical)

- Liquids in sealed containers
- Resin plastics that will melt (e.g., high-density polyethylene (HDPE), low-density polyethylene (LDPE), polyethylene terephthalate (PET), or polyethylene terephthalate glycol-modified (PET-G))
- Animal carcasses/anatomical remains.

These items will be refused at RMW drop-off locations and PI/GA's will be told to contact EH&S for proper disposal.

Approved biohazard bags must be:

- a. Red in color
- b. Contain the universal biohazard label (at least 2" tall)
- c. Contain the words "Biohazard" or "Potentially Infectious Material"
- d. Contain the word "Autoclave Bag" or "Autoclavable"
- e. Made of Polypropylene
- f. At least 2.0 mil thick
 - Recommended bag: [FisherBrand 01-828D](#)

Unless specifically used to clean-up a biological spill, absorbent material (e.g., paper towels) should never be placed in the RMW bags. Absorbent material used to routinely clean a work surface before and after use, should be placed in the regular trash.

Loosely seal the bags with twist ties, rubber bands, or autoclave or laboratory tape when the bag is 3/4 full. Bags must **never** be sealed air-tight, as this will cause the bags to expand during autoclave and will also restrict steam from penetrating into the bag, significantly reducing the effectiveness of the autoclave.

All bags must be double bagged and placed in approved, autoclavable sterilization trays. Each lab is required to purchase their own sterilization trays and are responsible for retrieving them once their waste has been autoclaved.

BSSF recommends the following sterilization tray:

- [Thermo Scientific Nalgene Large Polypropylene Sterilizing Pan](#) (Catalog #13-359-20B)
- Any other type of tray should be cleared through BSSF staff prior to ordering.

RMW will be accepted for sterilization at the following locations:

- a. **BSSF (MGB207):** Schedule waste drop-off
- b. **Health Sciences:** At the discretion of the autoclave operator
- c. **Chemistry:** availability of autoclave (training required prior to use)

RMW must always be disposed of (e.g., brought to BSSF or Health Sciences) within 7 days of generation. Laboratories cannot serve as a storage facility for RMW. It is the responsibility of the RMW generator (i.e., the laboratory) for assuring that absolutely no chemical or radioactive waste is contained within the RMW bags.

Individual laboratory autoclaves cannot be used for the official decontamination of regulated medical waste. The only autoclaves on campus approved for treating regulated medical waste, which involving permitting and routine inspections, are in BSSF and Health Sciences. If laboratories have individual autoclaves, they may be used for sterilization of materials only. While laboratories may choose to use their autoclaves for decontaminating some of their waste, it must always still be brought to one of the approved autoclaves on campus for treatment and final disposition.

Liquid Biohazardous Waste

Liquid biohazardous waste includes blood, blood products, body fluids from human and animal research, and contaminated culture media. These liquids must be collected and stored in a closeable, rigid, leak-proof container labeled with the universal biohazard symbol and the word “Biohazard”. The collection vessels should be secured so that they cannot be tipped over. Secondary containment is strongly recommended and can be achieved by placing the container within a bucket or deep tray. If disinfectant is added to the container for the treatment of liquid waste, provide labeling to identify the chemical hazard as well (e.g., bleach-treated cell culture materials).

Liquid wastes must be treated and disposed of by either one or the other following methods:

a. *Chemical treatment with disinfectant*

****Laboratories located within MGB that choose to decontaminate their waste using this method must do so in accordance with the guidance below:**

- The mixed solution must sit out in an open container overnight to deactivate the bleach prior to dumping.
- When dumping, turn on water to rinse out the sink and let run for about 2-3 minutes to help flush pipes.

For all other laboratories:

EPA registered disinfectants effective at inactivating the agent(s) present in the collection vessel may be used for the treatment of liquid biological wastes. Most liquid wastes can be inactivated with bleach. Add bleach to collection vessel so that the final concentration is between 0.5 - 2% sodium hypochlorite. Household bleach typically contains approximately 5.25% sodium hypochlorite. Allow for a minimum contact time of 20 minutes. Carefully discharge the mixture to the sanitary sewer by way of the laboratory sink, then thoroughly rinse down the sink with copious amounts of water. Remember to always wear proper PPE for this procedure (goggles, gloves, and a lab coat).

The laboratory-specific protocol for chemically treating and disposing of wastes via the sanitary sewers are to be described in the IBC Registration Form.

NOTE: Many chemicals used for disinfection cannot be discarded down the drain. Contact EH&S to determine if sink disposal of disinfectants other than bleach is acceptable.

b. Autoclave treatment of liquids

Place the closed collection vessel in a secondary container and transport by cart to one of the two autoclave locations (BSSF or Health Sciences). Immediately prior to autoclaving, always loosen or remove the caps/closure on the container prior to placing in the autoclave (do not loosen the caps prior to transport as this will increase the chance of spilling). Once autoclaved and cooled, the decontaminated liquids can be poured down a laboratory sink for discharge into the sanitary sewer. Only personnel who have received training regarding the operation of the autoclave are to perform this procedure.

Note: NEVER autoclave materials containing solvents, volatile, or corrosive chemicals (e.g., phenols, chloroform, bleach, etc.), or radioactive materials.

Disposal of Animal Carcasses, Body Parts, Tissue, and Bedding

All animal carcasses and parts, regardless of infection status, are disposed of as pathological waste.

- a. All non-preserved carcasses should be stored in a freezer or cold storage area prior to disposal. Always secure limbs and sharp protrusions so they do not puncture the bag.
- b. Animal tissues and animal bedding must be disposed of as pathological waste if the source animal was infected **with a risk group 2 or higher agent.**

All animal carcasses and wastes are picked up by a waste contractor and transported to their facility for incineration.

Human Pathological Waste

Human cadavers and recognizable human anatomical remains require special handling. Please contact EH&S at 757-683-4495 for guidance.

Transport of Waste

All bagged or liquid waste and serological pipettes must be placed in an approved sterilization tray, covered with foil for pipettes, and placed on a cart or in a bin for transport to the autoclave.

Never hand carry any biological waste outside of the labs.

XIV. Transportation of Biological Materials

All shipments of biohazardous materials must be arranged through Environmental Health and Safety (757-683-4495).

Old Dominion University is legally required to comply with the Department of Transportation (DOT) and/or the International Air Transport Association (IATA) regulations as they apply to shipping dangerous goods and hazardous materials. The intent of the packaging and transportation regulations is to prevent accidental exposure of personnel who may handle the material during its shipment. There, certain general criteria apply to all possible transportation scenarios.

Federal regulations have outlined specific shipping requirements for DG/hazardous materials. Depending upon the mode of transportation and destination, these shipments are regulated by the 49 Code of Federal Regulation parts 171-180 and /or International Air Transport Association (IATA). To comply with shipping regulations, hazardous materials must be properly classified, documented, packaged, and handled.

The following controls must be in place for the transportation of any biological materials:

- Emergency procedures (e.g., spill clean-up & disinfection protocols) and contact information must be known to the person carrying the materials
- Container must be appropriate for the material being transported and must be properly labeled
- Material must be packed for that it will stay upright during transportation
- Proper PPE must be worn during the packaging of the material
- Hands should be washed after handling materials, after the removal of gloves.
- The person packaging the material must ensure that the exterior surfaces of each package are free of any potential contamination by the packed material.

Anyone who offers for shipment any hazardous materials **must** receive the appropriate DOT/IATA training **prior** to shipping any materials. Examples of these materials include:

- Laboratory chemicals, cryogenic materials, and samples containing flammable, toxic, explosive, radioactive, oxidizer, and/or corrosive materials
- Paints, stains, refrigerants, aerosols, medicines, pesticides, disinfectants, fuels
- Equipment containing hazardous materials, such as mercury, compressed gases, batteries (wet, lithium, and dry batteries containing sodium, potassium hydroxide)
- Dry ice
- Biological/infectious materials

Transportation of Materials on Campus

All individuals must adhere to the following requirements for the transportation of biological materials within on campus (e.g., between laboratories).

- a. At a minimum, all laboratory materials must be transported within a secondary container that is shatterproof and leak-proof. The secondary container must also be easy to decontaminate and absorbent material should be placed inside the secondary container to absorb any spills.
- b. Materials must never be carried in hands or pockets.
- c. Label information must include the biohazard symbol (if risk group 2 or above) and the PI's name and laboratory on the outside of the secondary container.
- d. The container must be carried directly to the intended location and not taken to offices, bathrooms, cafeterias, or other public or inappropriate locations.
- e. Upon delivery, the receiving laboratory personnel should be informed on the material being delivered.
- f. The package should be carefully inspected for signs of leakage or other contamination and, if necessary, decontaminated before opening.

Transportation of Materials off Campus

Contact ODU EH&S for guidance and assistance. EH&S has personnel with the appropriate DOT/IATA training to ensure the proper shipment of all hazardous materials. Individuals must submit a [Dangerous Good Shipping Request Form](#) at least 3 days in advance to allow ample time to obtain the supplies that may be needed for packaging and shipping.

Note: If shipping a hazardous material that is a commercially available product, it is more cost-effective in many cases to purchase the material at the location to which you wish to ship it. For example, if you are planning field-research, which required the use of hazardous chemicals, you may find it easier and less expensive to have a vendor ship the chemicals directly to the location where you will be working.

Once the proper packing has been obtained, EH&S staff will pick-up the material for shipping and will package, label, and document the shipment in accordance with all applicable regulatory requirements. Any costs related to the packaging and shipping of the material will be the responsibility of the shipper.

Penalties for non-compliance with the rules are significant and could result in fines up to \$500,000 and jail sentences up to 5 years.

ODU's Mail Service Department cannot handle/transport hazardous materials.

Appendix A - Policy on the Use of Human and Non-Human Primate Cells and Tissues

1. Purpose

The purpose of this Institutional Biosafety Committee (IBC) policy is to describe the procedures and precautions that must be taken to protect personnel from exposure to human bloodborne pathogens (BBP), specifically those found in human cells and tissues, and to prevent environmental contamination, to provide an environment for high quality research while maintaining a safe work place, and to comply with all applicable federal, state, local, and university requirements.

2. Definitions

2.1. Primary Cells

Cells, tissues, organs, or other human/non-human primate derived materials taken directly from a subject, which is not immortalized.

2.2. Established Cell Lines

An “immortalized” or “continuous” cell line which has acquired the ability to proliferate indefinitely either through random mutation or deliberate modification.

2.3. Biosafety Level 1 (BSL-1)

Biosafety Level 1 is suitable for work involving well-characterized agents not known to consistently cause disease in immunocompetent adult humans, and present minimal potential hazard to laboratory personnel and the environment.

2.4. Biosafety Level 2 (BSL-2)

BSL-2 is suitable for work involving agents that pose moderate hazards to personnel and the environment.

2.5. Animal Biosafety Level 1 (ABSL-1)

Animal Biosafety Level 1 is suitable for work in animals involving well-characterized agents that are not known to cause disease in immunocompetent adult humans, and present minimal potential hazard to personnel and the environment.

2.6. Animal Biosafety Level 2 (ABSL-2)

ABSL-2 is suitable for work involving laboratory animals infected with agents associated with human disease and pose moderate hazards to personnel and the environment. It also addresses hazards from ingestion as well as from percutaneous and mucous membrane exposure.

NOTE: Research requiring BSL-3/ABSL-3 or BSL-4/ABSL-4 containment cannot currently be conducted at ODU.

3. Background

In 1991, the Occupational Safety and Health Administration (OSHA) issued the Bloodborne Pathogen (BBP) standard to protect employees who have occupational exposure to human blood or other potentially infectious materials. While human blood, most body fluids, unfixed human tissues, and organs were clearly included within the scope and application of the standard, the inclusion of human cell lines was ambiguous.

In 1994, OSHA issued an interpretation of the applicability of the BBP standard towards human cell lines. According to the interpretation, human cell lines are considered to be potentially infectious and within the scope of the BBP standard unless the specific cell line has been characterized to be free of hepatitis viruses, HIV, Epstein-Barr virus, papilloma viruses, and other recognized bloodborne pathogens. The US Department of Health and Human Services publication, *Biosafety in Microbiological and Biomedical Laboratories* (BMBL), 5th edition, also describes the risks associated with handling human cells, tissues, blood, and body fluids and states that it is the responsibility of the institution to conduct a proper risk assessment based on the origin of the cells or tissues (e.g., species and tissue type), as well as the source (e.g., recently isolated or well-characterized).

The main risk presented by cell cultures is their ability to sustain the survival and/or replication of a number of adventitious agents. The major agents of concern are viruses, but other agents such as *Mycoplasma* spp. should also be considered. In addition, the components of cell culture media and cell products that could be biologically active are other hazards that should be assessed. The following factors should be addressed in a risk assessment involving the use of human or non-human primate cell lines:

3.1. *Origin of cell line and source population from which cell line was derived*

The risk from any cell line should be considered in terms of the likelihood of contamination and the ability of the cell line to support growth.

3.2. *Source of tissue*

This provides an indication of potential contamination and potential for expression/reactivation of latent viruses. Cells derived from peripheral blood and lymphoid cells present the greatest likelihood of contamination with serious human pathogens.

3.3. *Type of cell line*

Primary cell culture present the greatest risk of carriage, followed by continuous cell lines unless known to be persistently infected (e.g., B95-8 with EBV, and MT4 with HTLV), and well-authenticated/characterized cell lines such as those used for the manufacture of vaccines or recombinant proteins.

Note: Autologous cells or tissues (i.e., using one's own cells), must never be used in any laboratory setting. This presents a particular hazard as accidental self-inoculation could result in potentially serious consequences as they cells would circumvent the normal

protection of the immune system.

4. Policy

Based upon the regulations, guidelines, and recommendations for handling human and non-human primate cells and tissues, the ODU IBC has adopted the following policy:

4.1. *Biosafety Level*

- All human and non-human primate cells and tissues, including both primary material and established cell lines, must be handled using BSL-2 work practices and containment. All experiments involving these materials require IBC approval prior to initiation of work.

Note: The biosafety level designated to ATCC cell lines is strictly for the purposes of safe shipment, not laboratory containment, according to whether or not it is known that the cell line is harboring a known virus or any portion of a virus which causes human disease. ATCC further recommends handling all human and primate cultures under BSL-2 containment ([Biosafety level for ATCC cultures](#)).

4.2. *Animal Biosafety Level*

- All primary cells/tissues from human or non-human primate origin must be handled at ABSL-2, unless rigorously tested and shown to be free of human pathogens of concern. These pathogens include:
 - Mycoplasma spp., Human immunodeficiency virus 1 (HIV1), Human immunodeficiency virus 2 (HIV2), Human T-lymphotropic virus 1 (HTLV1), Human T-lymphotropic virus 2 (HTLV2), Hepatitis virus A, Hepatitis virus B, Hepatitis virus C, Hantaviruses (Hantaan, Seoul, Sin Nombre viruses), Herpes simplex 1, Herpes simplex 2, Human cytomegalovirus, Human herpes virus 6, Human herpes virus 8, Human adenovirus, Human papillomavirus 16 & 18 (HPV), Lymphocytic choriomeningitis virus, Varicella virus, and Epstein-Barr virus.
 - If negative for these pathogens, then ABSL-1 containment may be considered by the IBC and/or IACUC.
- Well-characterized established cell lines that are from a commercial vendor (or other source) with documentation of being free of pathogens (e.g., ATCC tests for HBV, HCV, HIV, CMV, and EBV for cell lines accessioned into their inventory since 2010), may be considered acceptable for ABSL-1 containment and work practices.
 - The level of containment is at the discretion of the IBC and/or IACUC. Each protocol will be risk assessed for any contributing factors that may warrant a higher level of containment.

- Established cell lines that do not have documentation of any testing or are not from a commercial vendor (i.e., received from a collaborator), ABSL-2 work practices and containment is required.
 - As with primary materials, if the cells are tested and negative for human pathogens of concern as listed above, then ABSL-1 may be considered at the discretion of the IBC and or IACUC.

References

- 4.3. American Type Culture Collection. Support. *Biosafety level of ATCC culture*. 2012.
- 4.4. Health and Safety Executive. *Biological agents: Managing the risks in laboratories and healthcare premises*. Advisory Committee on Dangerous Pathogens. 2005.
- 4.5. Occupational Safety and Health Administration. 1910.1030 - Bloodborne Pathogen standard. 1991
- 4.6. Occupational Safety and Health Administration. Bloodborne Pathogen standard interpretation. 1994.
- 4.7. Centers for Disease Control and Prevention. *Biosafety for Microbiological and Biomedical Laboratories*. 6th edition. 2020.

Appendix B: Summary of Biosafety Levels - Working with Viral Vectors

Viral Vector (<i>not wild-type virus</i>)	Biosafety Level		Additional Precautions	Disinfectant
	In vitro	In vivo		
Adenovirus	BSL-2	ABSL-1; ABSL-2	- ABSL-2 housing required for administration of adenovirus and for 72 hours post-inoculation - In the absence of human cells, animals may be downgraded to ABSL-2 containment after 72 hours	1% sodium hypochlorite, 2% glutaraldehyde, 0.25% sodium dodecyl sulfate
Adeno-Associated Virus (AAV)	BSL-1; BSL-2	ABSL-1; ABSL-2	- BSL-2 required in the presence of helper virus - Animals may be housed at ABSL-1 unless helper virus is present, which requires ABSL-2 containment	1% sodium hypochlorite, 2% glutaraldehyde, 0.25% sodium dodecyl sulfate
Epstein-Barr Virus (EBV)	BSL-2	ABSL-2	EBV vectors must be administered to animals under ABSL-2 containment and animals must remain at ABSL-2 for duration of the study.	1% sodium hypochlorite recommended disinfectant
Herpes Virus HSV-I and HSV-II	BSL-2	ABSL-2	HSV vectors must be administered to animals under ABSL-2 containment and animals must remain at ABSL-2 for duration of the study.	1% sodium hypochlorite recommended disinfectant
Lentivirus	BSL-2; BSL-2+*	ABSL-1; ABSL-2	- BSL-2 for non-human viruses, non-human pseudotyped viruses, or viruses that do not express transgenes with any known oncogenic potential or a biological toxin. - BSL-2+ for human viruses, amphotropic or VSV-g envelope pseudotyped viruses expressing transgenes with known oncogenic potential or a biological toxin. - ABSL-2 may be downgraded to ABSL-1 after 72 hours ONLY in animals that do not or will not contain any human cells or tissues	1% sodium hypochlorite, 2% glutaraldehyde, formaldehyde, 70% ethanol
Moloney Murine Leukemia Virus (MMLV)	BSL-1; BSL-2	ABSL-1; ABSL-2	- BSL-1/ABSL-1 for ecotropic vectors - BSL-2/ABSL-2 for amphotropic or VSV-g pseudotyped vectors containing biological toxin or gene with oncogenic potential - ABSL-2 may be downgraded to ABSL-1 after 72 hours ONLY in animals that do not or will not contain any human cells or tissues	1% sodium hypochlorite, 2% glutaraldehyde, formaldehyde, 70% ethanol
Pox Viruses / Vaccinia	BSL-1; BSL-2	ABSL-1; ABSL-2	- BSL-1/ABSL-1 for NYVAC, TROVAC, and ALVAC highly attenuated vaccinia strains - BSL-2/ABSL-2 for MVA highly attenuated strain and WR, NYCBOH, Copenhagen, Temple of Heaven, and Lister non-highly attenuated strains - Vaccinations recommended for non-highly attenuated strains	1% sodium hypochlorite, 2% glutaraldehyde, formaldehyde
Rabies Viral Vector	BSL-2	ABSL-2	ABSL-1 may be utilized for work with SAD-B19 vectors. No vaccination is needed for work with this vector. IBC will determine if vaccination is appropriate for personnel working with other recombinant rabies vectors	70% ethanol, phenol, formalin, ether, trypsin, β -propiolactone
Sendai Virus (SeV)	BSL-2	ABSL-2	BSL-2 required due to SeV causing respiratory disease in rodents and sometimes swine, but limited evidence of zoonotic transmission to humans. However, the virus is capable of infecting human cell lines and is similar to human parainfluenza virus type 1.	1% sodium hypochlorite, 70% ethanol, formaldehyde

*BSL-2+: Defined as utilizing BSL-3 work practices within BSL-2 containment. This includes, but is not limited to, conducting all work within a biosafety cabinet (no open bench work allowed), decontamination of all work surfaces with appropriate disinfectant immediately following work with biohazardous materials, required use of lab coats and other PPE, such as gloves, eye protection, and possibly respiratory protection. All waste must be inactivated through autoclaving or another appropriate method prior to disposal

Appendix C – Guidelines for Disposal of Regulated Medical Waste (per Autoclave location)

BSSF / MGB

- <https://www.odu.edu/sites/default/files/documents/bssf-medical-waste-guidelines.pdf>

Health Sciences

- https://www.odu.edu/sites/default/files/2024/documents/HS_medwaste_guidelines.pdf

Chemistry

- https://www.odu.edu/sites/default/files/documents/chem_medwaste_guidelines.pdf

IRP / CBE

- IRP / CBE Guidelines for disposal of Regulated Medical Waste