



Safety and Risk
Management

Biological Safety Manual

Revision Date: January 2024

Table of Contents

Forward.....	1
Section 1: Responsibilities.....	1
Safety and Risk Management Office.....	1
Department Heads.....	1
Principal Investigators or Supervisors.....	1
Laboratory Personnel.....	2
Section 2: Laboratory Registration	3
Registration of Biological Agents	3
Chemical Inventory	3
Section 3: Laboratory Inspections.....	3
Inspection Reports.....	4
Lab Inspection Follow-up Process.....	4
BSL-2 Lab Inspections.....	4
Section 4: Laboratory Close-Out Procedure	4
Section 5: Introduction to Biosafety	5
Routes of Exposure	5
Biosafety Levels (BSL).....	5
Pathogen Risk Group (RG).....	8
Biosafety Levels & Recommended Safety Practices	8
Section 6: Standard Laboratory Practice and Technique.....	10
Personal Protective Equipment (PPE).....	10
Handwashing.....	10
Hand to Face Contact.....	10
Housekeeping	10
Pipetting.....	10
Sharps Safety.....	11
Decontamination Procedures	12
Transportation Procedures within the Campus.....	12
Section 7: Standard Operating Procedures (SOPs)	13
High Risk Procedures	13
Section 8: Recombinant DNA (rDNA).....	14

NIH Guidelines	14
rDNA Descriptions.....	14
Institutional Biosafety Committee (IBC)	14
rDNA Registration Process	18
General Laboratory Procedures.....	18
Incident Response and Reporting.....	18
rDNA Research Training	18
Section 9: Biological Waste Management	19
Autoclave Safety	19
Section 10: Biohazard Spill Cleanup.....	19
Section 11: Safety Training Requirements.....	19
Section 12: Facility Design	20
Furniture Selection.....	20
Cold Rooms	22
Section 13: Laboratory Equipment	23
Biological Safety Cabinet (BSC)	23
Standard Practices when utilizing BSCs	23
BSC Open Flame Policy.....	24
Certification of BSCs.....	24
Centrifuges.....	24
Blenders and Homogenizers	25
Furniture and Equipment Surplus or Disposal	26
Section 14: Bloodborne Pathogens.....	26
Appendix A: Emergency Contacts	27

Forward

The Biological Safety Manual is intended to highlight general university wide laboratory practices that are obligatory for protecting employees from exposure to hazardous biological materials. The guidelines provide methods for evaluating the risks involved with biological materials and for the proper handling practices that will effectively minimize the risk of an injury or illness. Laboratories designated at Biosafety Level 2 (BSL2) are required to have a copy of this manual available for lab personnel.

Section 1: Responsibilities

Safety and Risk Management Office

- Maintain records of biological agents used on campus, their risk classification, location, and responsible Principal Investigator (PI).
- Assist PI/Laboratory Supervisors in the selection of appropriate safety control requirements, which include laboratory practices, personal protective equipment, engineering controls, and training.
- Maintain in functional working order appropriate workplace engineering controls (e.g., fume hoods) and safety equipment (e.g. emergency showers/eyewashes, fire extinguishers), with emphasis on controls for particularly hazardous substances.
- Assist with hazard assessments and provide advice on laboratory SOPs, upon request.
- Maintain area and personal exposure-monitoring records.
- Provide technical consultation and investigation, as appropriate, for laboratory accidents and injuries.
- Help to determine medical surveillance requirements for laboratory personnel, when applicable.
- Review plans for installation of engineering controls and new laboratory construction or renovation projects, as requested.
- Review and evaluate the effectiveness of the Biological Safety Manual at least annually and update as appropriate.
- Additional responsibilities regarding Institutional Biosafety Committee (IBC) involvement.

Department Heads

- Ensure compliance with Biological Safety Manual and IBC requirements for biological safety within their respective departments.
- Provide direction on departmental approach to development and implementation of laboratory specific Chemical Hygiene Plans.
- Ensure that proper storage areas are provided.

Principal Investigators or Supervisors

- Identify biohazardous conditions or operations in the lab, determine safe procedures and controls, and implement and enforce standard safety procedures.
- Establish standard operating procedures (SOPs) for higher risk materials and tasks.
- Develop and document the Laboratory Specific CHP and provide access to the WCU Chemical Hygiene Plan if hazardous chemicals are utilized or stored in the laboratory.

- Provide laboratory personnel under his/her supervision with access to the Biological Safety Manual, and all applicable SOPs and SDSs.
- Train laboratory personnel he/she supervises to work safely with biological materials and operations, and maintain records of training provided locally.
- Maintain in functional working order appropriate personal protective equipment (e.g., gloves, goggles).
- Promptly report laboratory accidents and injuries to the Office of Safety and Risk Management.
- Submit an annual inventory of potentially infectious agents to the Office of Safety and Risk Management.
- Make available required medical surveillance or medical consultation/examination for laboratory personnel, when applicable.
- Inform facilities personnel, other non-laboratory and any outside contractors of potential lab-related hazards when they are required to work in the laboratory environment. Identified potential hazards should be minimized to provide a safe environment for repairs and renovations.
- Ensure training and coordinating audits are completed.
- Additional responsibilities regarding IBC involvement.

Laboratory Personnel

- Follow the guidelines in all laboratory safety documents.
- Follow oral and written laboratory safety rules, regulations, and standard operating procedures required for the tasks assigned.
- Keep the work areas safe and uncluttered.
- Review and understand the hazards of materials and processes in the laboratory research prior to conducting work.
- Utilize appropriate measures to control identified hazards, including consistent and proper use of engineering controls, personal protective equipment, and administrative controls.
- Understand the purpose, capabilities, and limitations of personal protective equipment.
- Promptly report accidents and unsafe conditions to the PI/Laboratory Supervisor.
- Complete all required health, safety and environmental training.
- Participate in the medical surveillance program, when required.
- Inform the PI/ Laboratory Supervisor of any work modifications ordered by a physician as a result of medical surveillance, an occupational injury, or exposure.

Additional Duties of Laboratory Personnel Working Autonomously:

In addition to the above responsibilities, laboratory personnel working autonomously or performing independent research are also responsible for the following:

- Provide the PI/Laboratory Supervisor with a written scope of work for their proposed research.
- Notify and consult with the PI/Laboratory Supervisor, in advance, if they intend to deviate from their written scope or scale of work.

- Provide appropriate oversight, training and safety information to laboratory personnel they supervise or direct.

Generally, it is prudent to avoid working in a laboratory building alone. Under normal working conditions arrangements should be made between individuals working in separate laboratories outside of working hours to crosscheck periodically. Experiments known to be hazardous should not be undertaken by a worker who is alone in a laboratory.

Section 2: Laboratory Registration

Every laboratory at WCU must be registered with the Safety and Risk Management Office. This process ensures that the Safety Office has a current list of occupied labs and responsible parties, a survey of the type of work being conducted in the labs, and an accurate list of lab emergency contacts.

The Principal Investigator (PI) or designated lab supervisor is required to submit a “Lab Registration Form” to initially register the space and at any time in the future if any of the following apply:

- You are relocating to a new lab space or become responsible for an additional lab space
- You are using a new hazardous chemical, biological agent, or new hazardous procedure

The Safety Office will use the information provided on the form to develop door signs for the lab. This provides a necessary reference in the event of an emergency.

Registration of Biological Agents

An annual inventory of biological agents used in research and teaching labs is required to be submitted to the Office of Safety and Risk Management. The inventory should include a list of the biological agents (bacteria, virus, fungi, etc.), the risk level of the agent, the storage location, use location (teaching lab, research lab), and the responsible person (Lab Supervisor, PI, Lab Coordinator, etc.).

Chemical Inventory

A system for maintaining an accurate chemical inventory on campus is essential for compliance with local and state regulations and any applicable building codes. Every lab should maintain an up-to-date chemical inventory and a physical chemical inventory must be performed at least annually and submitted to the Safety and Risk Management Office. The chemical inventory should include the chemical name, chemical abstract number (CAS), manufacturer & product number (reorder #, catalog #, etc.), quantity and location.

A laboratory storing and/or using hazardous chemicals is required to have the current WCU Chemical Hygiene Plan (CHP) available for lab personnel. The CHP highlights laboratory practices that are obligatory for protecting employees from exposure to hazardous chemicals.

Section 3: Laboratory Inspections

As required by State and Federal law, the Safety and Risk Management Office will conduct laboratory inspections to determine individual laboratory compliance with WCU’s Biosafety Manual, Chemical Hygiene Plan (CHP), and other relevant safety policies. These surveys are comprehensive and address record keeping, fire safety, egress, engineering controls, personal protective equipment, work practices,

and where appropriate, chemical, biological, and radiation safety. At least one annual inspection will be announced in order to work directly with the PI or laboratory supervisor to address specific items, such as inventories of particularly hazardous materials or processes, biosafety compliance, and any other safety concerns that arise. Other inspections may be unannounced to provide a snapshot of laboratory safety and compliance and help to continually improve the safety program.

Inspection Reports

An inspection report identifying deficiencies and areas for corrective action will be directed to the laboratory's principal investigator or supervisor. These items must be corrected within 30 days of receipt of the laboratory inspection report. If the items cannot be corrected in that timeframe, the principal investigator must submit a written corrective action plan detailing the expected corrections and estimated date of completion within the same 30 days. Any inspection finding deemed an imminent danger (likely to cause a serious hazard, injury, disability, or death) must be corrected immediately.

Lab Inspection Follow-up Process

1. If no response is received within 30 days of the initial report, then the Safety Office as a courtesy will contact the Principal Investigator of the laboratory with a reminder. If the laboratory conducts research, additional department designees may also be notified.
2. If no response is received and/or corrective actions are not completed after 60 days from receipt of the initial inspection report, the laboratory will be deemed noncompliant, and information will be forwarded to the Dean's Office.

Previous inspection reports are a good measure for addressing safety issues and eliminating laboratory risks. To help prepare for future inspections, please review the "Laboratory Inspection Checklist" and perform self-inspections on a regular basis. The self-inspection process is an excellent learning tool for students and other lab personnel and should be documented as part of the lab specific safety training requirement.

BSL-2 Lab Inspections

Laboratories designated as BSL-2 have some additional regulatory requirements; therefore, PIs operating at BSL-2 should perform their own laboratory self-inspections using the "BSL-2 Laboratory Inspection Checklist" available on the Safety & Risk Management website.

Section 4: Laboratory Close-Out Procedure

The Safety Office must be notified prior to a laboratory move, relocation, or vacancy for any reason in order to perform a lab check-out assessment. This procedure will ensure that all hazardous materials are properly accounted for or disposed of and will prevent the next occupant from inheriting "unknown" or potentially hazardous materials. Contact the Safety and Risk Management Office (828-227-7443) to begin the closeout procedure.

Lab equipment that is broken or unwanted, such as refrigerators, freezers, incubators, centrifuges, etc., may be discarded through WCU Surplus. Equipment that could possibly be contaminated with biological,

chemical, or radioactive materials must be decontaminated prior to disposal. Contact SRM to request an “Equipment Clearance” before scheduling pickup with the Surplus Department.

Section 5: Introduction to Biosafety

Over the past two decades, *Biosafety in Microbiological and Biomedical Laboratories (BMBL)* has become the code of practice for biosafety in addressing the safe handling and containment of infectious microorganisms and hazardous biological materials. The fundamental objective of any biosafety program is the containment of harmful biological agents. This is achieved through microbiological practices, safety equipment, and facility safeguards that protect laboratory workers, the environment, and the public from exposure to infectious microorganisms that are handled or stored in the laboratory. Individual workers who handle pathogenic microorganisms must understand the containment conditions under which infectious agents can be safely manipulated and secured. The subsequent sections of this manual address the principals outlined in the BMBL for containment and risk assessment.

Routes of Exposure

There are four main routes of exposure employees need to be aware of and attempt to avoid when working with biohazardous agents in the laboratory.

- **Ingestion:** Accidental ingestion typically results from improper personal hygiene in the laboratory. Hands must be washed after removing gloves, immediately upon visible contamination, and before leaving the laboratory. Food, drink, tobacco products (including electronic cigarettes), and cosmetic application are prohibited in all areas of the laboratory.
- **Percutaneous Injuries (Injection):** Results from needle sticks, cuts, or abrasions from contaminated items. This is a particularly serious route of entry because of the possibility for immediate entry of the agent into the bloodstream.
- **Inhalation:** Various laboratory procedures can cause aerosolization of infectious agents. Appropriate work practices must be implemented to minimize the aerosolization of materials, especially those known to be transmitted via aerosols (Tuberculosis, Adenovirus, Brucellosis).
- **Mucous membrane absorption:** Exposure of mucous membranes to infectious agents can lead to occupationally acquired infections. Mucocutaneous exposures typically result from splashes to the face or inadvertent inoculation from contaminated hands. Face protection should always be worn if there is potential for splash or spray.

Biosafety Levels (BSL)

Biological agents are assigned to four biosafety levels (BSL) according to the risk they pose to human health and the environment.

BSL-1 is appropriate for work involving well-characterized agents not known to regularly cause disease in immunocompetent adult humans and present minimal potential hazard to laboratory personnel and the environment. All bacterial, viral, fungal, rickettsial, parasitic, and chlamydial agents that have been assessed for risk and do not belong to a higher risk group may be safely handled at BSL-1. It is important to be aware that numerous agents not ordinarily associated with disease are opportunistic pathogens that may cause infection in the elderly, the young, and immunocompromised individuals and still need to be

treated as though they are pathogenic. Examples of agents handled at BSL-1 include: Bacillus subtilis, non-pathogenic E. coli, yeast, canine hepatitis, etc.

BSL-2 is appropriate for work involving agents that pose a moderate threat to humans in that they may cause mild disease or are difficult to contract via aerosol. BSL-2 differs from BSL-1 in that the laboratory personnel have specific training in handling pathogenic agents, access to the laboratory is restricted, and any procedure that could create infectious aerosols is conducted within a biosafety cabinet (BSC) or other physical containment equipment.

A sign incorporating the universal biohazard symbol must be posted at the entrance to the laboratory when infectious agents are present. For Biosafety Level 2 (BSL-2) the door sign **must** include the name of the agent(s) in use, an indication that the area is a BSL-2 lab, and the PI's name (or other responsible personnel), telephone number, and required procedures for entering and exiting the laboratory.

Written standard operating procedures (SOP) for agents used at BSL-2 are required and supplement this general lab biosafety manual for the lab-specific training. SOP templates are available from the Safety & Risk Management website.

BSL-2 Agents (including, but not limited to the following)

BSL-2 Viral Agents	
<ul style="list-style-type: none"> • Adenovirus • Creutzfeld-Jacob agent • Cytomegalovirus • Eastern equine encephalitis • Epstein-Barr virus • Hepatitis A, B, C, D, E • Herpes simplex viruses • HTLV types I and II 	<ul style="list-style-type: none"> • Human Blood & Blood Products • Kuru • Monkeypox virus • SIV • Spongiform encephalopathies • Vaccinia virus • HIV • VSV (lab adapted strains)
BSL-2 Bacterial/Rickettsial Agents	
<ul style="list-style-type: none"> • <i>Campylobacter fetus</i>, <i>coli</i>, <i>jejuni</i> • <i>Chlamydia psittaci</i>, <i>trachomatis</i> • <i>Clostridium botulinum</i>, <i>tetani</i> • <i>Corynebacterium diphtheriae</i> • <i>Legionella</i> spp • <i>Neisseria gonorrhoeae</i> • <i>Neisseria meningitidis</i> • <i>Pseudomonas aeruginosa</i>, <i>pseudomallei</i> 	<ul style="list-style-type: none"> • <i>Proteus mirabilis</i>, <i>vulgaris</i> • <i>Salmonella</i> spp • <i>Shigella boydii</i>, <i>dysenteriae</i>, <i>flexneri</i>, <i>sonnei</i> • <i>Treponema pallidum</i> • <i>Vibrio cholera</i> (including El Tor) • <i>Vibrio parahemolyticus</i> • <i>Vibrio vulnificus</i> • <i>Yersinia pestis</i>
BSL-2 Fungal Agents	
<ul style="list-style-type: none"> • <i>Blastomyces dermatitidis</i> • <i>Cryptococcus neoformans</i> • <i>Microsporum</i> spp • <i>Exophiala dermatitidis</i> (<i>wangiella</i>) 	<ul style="list-style-type: none"> • <i>Fonsecaea pedrosoi</i> • <i>Sporothrix schenkii</i> • <i>Trichophyton</i> spp
BSL-2 Parasitic Agents	
<ul style="list-style-type: none"> • <i>Entamoeba histolytica</i> • <i>Cryptosporidium</i> spp • <i>Giardia</i> spp • <i>Naegleria fowleri</i> • <i>Plasmodium</i> spp 	<ul style="list-style-type: none"> • <i>Strongyloides</i> spp • <i>Tania solium</i> • <i>Toxoplasma</i> spp • <i>Trypanosoma</i> spp

Only BSL-1 and BSL-2 pathogens and research are permitted at WCU.

BSL-3 is applicable to clinical, diagnostic, teaching, research, or production facilities where work is performed with agents that may cause serious or potentially lethal disease through inhalation route exposure. Laboratory personnel must receive specific training in handling pathogenic and potentially lethal agents and special engineering and design features are required.

BSL-4 is required for work with dangerous agents that pose a high individual risk of life-threatening disease, aerosol transmission, or related agent with unknown risk of transmission.

The table below summarizes BSL laboratory practices and safety equipment for each BSL level. If a particular agent is not listed below, or if further assistance is needed in interpreting the BSL requirements for your laboratory, please contact the Safety and Risk Management Office.

Pathogen Risk Group (RG)

The pathogen Risk Group (RG) is a number assigned based on the inherent risk of the microorganism. It is important to note that the Risk Group and Biosafety Level are not synonymous. The Biosafety Level represents the containment and procedures used when working with an organism. To identify the appropriate BSL to use for a microbe, a risk assessment of multiple factors is used which includes, but is not limited to, the Risk Group of the microorganism.

- RG-1 are not associated with disease in healthy adult humans or animals.
- RG-2 are associated with disease which is rarely serious and for which preventative or therapeutics is often available.
- RG-3 are associated with serious or lethal human disease for which preventative or therapeutics may be available.
- RG-4 are associated with lethal human disease for which preventative or therapeutics are not readily available.

The American Biological Safety Association (ABSA) has a [Risk Group Database](#) that can be used to provide the RG for an organism.

Biosafety Levels & Recommended Safety Practices

BSL	Agents	Practices	Safety Equipment (Primary Barriers)	Facilities (Secondary Barriers)
1	Not known to regularly cause diseases in immunocompetent adults.	Standard Microbiological Practices. Integrated pest management program.	PPE as needed: Lab coats, gloves, closed-toed shoes, face protection.	<ul style="list-style-type: none"> • Open chemical resistant bench top • Non-porous furniture • No rugs/carpets • Doors for access control • Windows that open to the exterior must be fitted with screens • Sink for Handwashing

BSL	Agents	Practices	Safety Equipment (Primary Barriers)	Facilities (Secondary Barriers)
2	May cause disease but are difficult to contract via aerosol.	BSL-1 practices plus: <ul style="list-style-type: none"> • Restricted access to areas • sharps precaution • defined waste decontamination • medical surveillance policies • Laboratory specific biosafety BSL-2 SOP • Biohazard warning sign 	Biological Safety Cabinet (BSC) or other physical containment device. PPE (lab coats, gloves required and face/eye protection as needed)	BSL-1 facilities plus: <ul style="list-style-type: none"> • Autoclave available • Eyewash readily available • Doors should be self-closing and lockable
WCU does not have BSL-3 or BSL-4 facilities				
3	May cause serious or potentially lethal disease via inhalation, but for which treatment exists.	BSL-2 practices plus: <ul style="list-style-type: none"> • Controlled access to areas at all times • Decontamination of all waste • Decontamination of laboratory clothing before laundering 	BSL-2 safety equipment plus: BSC for all work with microbes. Respirators may be required.	BSL-2 plus: <ul style="list-style-type: none"> • Self-closing, double door access. • Exhausted air not recirculated • Negative air pressure in lab • Entry through airlock • Hands-free sink at laboratory exit
4	Highest level of biological safety. Microbes are dangerous and exotic, posing a high risk of aerosol-transmitted infection. Cause serious or potentially lethal disease for which treatment does not exist.	BSL-3 practices plus: <ul style="list-style-type: none"> • Clothing change before entering • Shower on exit • All material decontaminated before leaving 	Manipulation of agents must be performed in a Class III BSC. Personnel must wear a positive pressure supplied air suit.	BSL-3 plus: <ul style="list-style-type: none"> • Separate building or isolated zone • Dedicated supply and exhaust, vacuum, and decontamination systems • Additional requirements detailed in CDC text

Section 6: Standard Laboratory Practice and Technique

Personal Protective Equipment (PPE)

When a biological hazard has been identified, the supervisor/principal investigator (PI) must assign appropriate PPE. Supervisors are responsible for training those exposed in their laboratories on proper selection and use. Appropriate PPE must be donned before handling potentially hazardous biological materials and replaced immediately if excessively contaminated or damaged. PPE must be removed before exiting the laboratory.

- Gloves must always be worn when handling biohazardous materials. Disposable gloves (nitrile or latex) typically provide an adequate barrier to most biohazardous materials.
- Lab Coats/Gowns: Long sleeved lab coats or gowns (preferably cuffed) must be worn to protect skin and personal clothing from contamination. If the potential for splash or spray exists, the garment must be resistant to liquid penetration. Reusable clothing needs to be laundered and personnel should not take laboratory clothing home.
- Face Protection: Including but not limited to goggles, side-shielded safety glasses, and face shields. Must be used when splash or spray of potentially hazardous biological materials is anticipated and the work is being performed outside of a biological safety cabinet.
- Disposable Booties: When significant splash and spray are anticipated, shoe covers/booties should be utilized. Covers/booties must be removed and disposed of before leaving the laboratory.

Handwashing

Hands must be washed thoroughly for 20 seconds with mild soap and as soon as possible after coming in contact with potentially infectious materials. Hands should also be washed after glove removal, and before exiting the laboratory.

Hand to Face Contact

To minimize potential exposure, eating, drinking, smoking (including electronic cigarettes), applying cosmetics, and handling contact lenses is prohibited in laboratory areas. Food and drink may not be stored in refrigerators in which laboratory materials are also stored, unless they are for lab use only, and are labeled as such.

Housekeeping

Work benches will be maintained as organized and clutter-free as practical. Benches must be wiped down with a freshly prepared 10% bleach solution or other approved disinfectant at least once a day and immediately after a spill of potentially infectious materials.

Pipetting

In order to prevent the accidental inhalation, contact, or ingestion of pipetted infectious agents please use the following safety precautions when pipetting in the laboratory:

- Never pipette by mouth, always use mechanical pipetting aids.

- Release contents so that they may run down the wall of the container, do not release them from a height (to reduce risk of splatter).
- Place absorbent paper on benchtops to absorb any dripped infectious materials from pipette tips and reduce the risk of aerosol generation.
- Place disposable pipettes into pipette disposal boxes that have been lined with an autoclave bag and follow disposal guidelines.

Sharps Safety

Needles, scalpels, lancets, glass slides, cover slips, and glass pipettes all pose potential sharps hazards in laboratories. Personnel must take the following precautions to prevent injury during use, cleaning, and disposal of a sharp device:

- Substitute plastic ware whenever possible (plastic graduated cylinders, funnels, aspirators, etc.).
- Use appropriate Personal Protective Equipment (PPE). The supervisor should assess the exposure potential for procedures performed and identify the necessary PPE for personnel.
- Sharp items must be disposed of immediately after use in an appropriate puncture-resistant container labeled as “biohazardous sharps” or “non-biohazardous sharps”.
- Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs, or forceps and placed in a glass disposal box.
- Needles must not be recapped, purposely bent or broken, removed from disposable syringes, or otherwise manipulated by hand. Exceptions (such as when needles must be recapped for sterility, i.e., re-use of needle on the same patient) for specific procedures must be approved by the Safety Office.
- Any approved recapping procedures must be done either by using a recapping device or a one-handed scoop method for recapping. The one-handed scoop technique uses the needle itself to pick up the cap, and then the cap is pushed against a hard surface to ensure a tight fit onto the device. The cap may also be held with tongs or forceps and placed over the needle. Immediately after use, these sharps must be placed into appropriate containers.
- All personnel must be trained on the availability and use of approved safety devices where appropriate for their work responsibilities. Where feasible, sharps with engineered sharps injury protection, such as self-sheathing needles, retractable needles, or needleless systems must be used.
- All personnel handling sharps when working with human blood or other potentially infectious material (OPIM) must review the Bloodborne Pathogens Exposure Control Program and complete the required training.
- Follow Universal Precautions when working with potentially infectious material. This is an approach to infection control to reduce the potential for exposure to bloodborne pathogens by treating all human blood and body fluids (OPIM) as if they are known to be infected.

Decontamination Procedures

The following describes the three main types of physical and chemical means of decontaminating hazardous materials to ensure safety for additional handling:

Radiation: Ultraviolet radiation (UV) may be used in biological safety cabinets (BSC) to decontaminate surface contamination. UV does not have a very high penetrating power and therefore is not effective when used in dirty/dusty areas. UV poses a burn hazard to eyes and skin and the following precautions must be followed to prevent injury:

- Activate the UV light only when the area is unoccupied.
- Turn the fan off and close the sash, if possible, when the UV lamp is on.
- Post a warning sign on the front of the BSC indicating the presence of UV light hazards. The sign must indicate "Caution: Turn OFF the UV light before working".
- UV should also be used in conjunction with another disinfection process and not relied upon as the sole means of decontamination.

Note: UV light does not work in shadowed areas (cracks, grill plates, spill area) and is ineffective on microbes covered in dust, dirt, or organic matter.

Heat: Wet heat (steam) is the most reliable method of sterilization. Autoclaves are to be utilized to sterilize glassware and media and decontaminate BSL-1 and BSL-2 level waste (aside from human blood/tissues which will be sent to a 3rd party for incineration). Autoclaves should be monitored for efficacy by the use of biological indicators. The generator of the waste is responsible for performing and documenting this testing.

Liquid Disinfectant: Liquid disinfectants are used for surface decontamination. Any EPA registered disinfectant can be used as long as it is effective for the biological agents in use and the manufacturer's instructions are followed regarding the amount of disinfectant to use and the length of time it must remain wet on the contaminated surface. Employees must be trained in the proper use of the disinfectant and adhere to the instructions on the label.

Tuberculocidal disinfectant or a freshly prepared 10% bleach solution should always be used for decontamination when human materials are handled. Fresh bleach solutions must be prepared daily (every 24 hours) as they quickly deteriorate and become ineffective. The contact time for bleach is generally considered to be the time it takes the product to air dry. Solutions of bleach should not be stored in glass containers, but in materials such as the plastic containers the consumer bleach product is packaged in.

Transportation Procedures within the Campus

When transporting biological agents between laboratories or buildings on campus, special care must be taken. Biological agents should be placed in a closed, leak proof container that is labeled with the contents, responsible researcher, and date. The container must be shatter-proof and closed with an appropriate lid (do not use an easily breakable seal like parafilm). Then, the primary container must be placed in a secondary container that has a closed lid and is shatter-proof and leak proof. Ensure that the

secondary container is clean and disinfected before placing the primary biological agent container inside. The secondary container must be labeled with the word "BIOHAZARD" and must include the biohazard symbol.

The container must be taken directly to the intended laboratory. Do not enter any offices, break rooms, cafeterias, or other inappropriate locations with the container. Upon delivery, the biological agent should be properly stored in the receiving laboratory. Lastly, inspect the primary and secondary containers for evidence of spillage.

Laboratories that receive biological agents that were not previously stored there should contact the Laboratory Safety Officer so that the campus biological agent inventory can be updated.

No biological agent should be transported via mail without the relevant DOT/IATA training. Contact the Safety and Risk Management Office if you have questions about shipping or receiving biological agents.

Section 7: Standard Operating Procedures (SOPs)

Written SOPs for any particularly hazardous substance (PHS) or procedures that pose unique health risks must be developed and made available for all lab members. The SOP describes how your lab will handle a hazardous material safely, including the amount and concentration of material you will use, how you obtain or create the working solution, special handling procedures, engineering controls, personal protective equipment, and waste disposal considerations. The SOP must be read and signatures documented by all lab personnel who may be exposed to the hazardous situation. A BSL-2 SOP template and chemical SOP templates are available from the Safety & Risk Management website.

High Risk Procedures

High risk procedures are lab procedures which are likely to require engineering controls beyond those found in the standard laboratory. These include the use of chemicals or toxins which require medical surveillance, vaccination, special antidotes, or exposure monitoring, and operations that pose significant risk of fire, explosion, or exposure to personnel if a malfunction were to occur (such as a utility outage, runaway reaction, broken container, or chemical spill).

Contact the Safety and Risk Management Office (828-227-7443) if you have questions regarding High Risk Procedures or if you need to obtain permission for any of the following procedures:

- Large quantities of liquid nitrogen or other cryogenics which could deplete oxygen in the air. Large quantities would be more than one freezer and one attached Dewar per room, filling a cryocart or cooler, or liquid nitrogen (or other cryogen) piped in from a tank located outside the building.
- Heat concentrated perchloric acid (requires a perchloric acid approved fume hood).
- Use pyrophoric gases or other reactive Particularly Hazardous Substances (PHS).
- Use hydrofluoric acid or other chemicals for which an antidote or specific first-aid treatment is required.
- Use formaldehyde at the level which may require exposure monitoring or respirator use.
- Create or synthesize nanomaterials.
- Use botulinum toxin, tetanus toxin, or other toxins for which vaccination is recommended.

- Perform a procedure or use other equipment that is likely to require engineering controls beyond those found in the standard laboratory.

Section 8: Recombinant DNA (rDNA)

NIH Guidelines

Researchers at Western Carolina University who construct and/or handle materials containing recombinant DNA molecules must comply with the requirements of the [National Institutes of Health \(NIH\) Guidelines for Research Involving Recombinant DNA Molecules](#). The following information and procedures are developed to assist Western Carolina University researchers with the documentation of this compliance. The NIH Guidelines provide practices for handling recombinant or synthetic nucleic acid molecules based on the risk group of the agent. Generally, experiments requiring the use of recombinant biological agents should be handled under the same BSL requirements as the highest risk group (RG) agent. For example, handling of adenoviral vectors (RG2) should be performed under BSL-2 conditions. Researchers should review Appendix B in the NIH Guidelines for the Classification of Biohazardous Agents by Risk Group to conduct a preliminary assessment of their research.

NIH Guidelines for Research Involving rDNA molecules are applicable to all rDNA research conducted or sponsored by an institution that receives any support for rDNA research from the NIH. **All recombinant DNA research at WCU must be registered with the WCU Institutional Biosafety Committee (IBC) whether or not the Principal Investigator received funding from NIH for the project.**

rDNA Descriptions

The NIH rDNA Guidelines define rDNA as

- Molecules that are constructed outside living cells by joining natural or synthetic DNA segments to DNA molecules that can replicate in a living cell AND molecules that result from the replication of those described above.

At WCU, rDNA work may include:

- Any RNA produced from rDNA, including messenger RNA (mRNA), small interfering RNA (siRNA), micro RNA (miRNA), etc.
- Genetically-modified organisms (animals, plants, bacteria, viruses, fungi, etc.). This includes creation, cross-breeding, or manipulation of transgenic animals and plants.
- Any such material obtained from another researcher or source.
- Plasmids and viral vectors.
- Any synthetic DNA or RNA.

Institutional Biosafety Committee (IBC)

The NIH rDNA Guidelines require that an IBC be established at any institution receiving NIH funding for rDNA research to oversee all rDNA research at that institution and ensure that such work is compliant with the guidelines. Additionally, WCU Policy #50 requires that an IBC be established to review all

recombinant DNA research conducted by WCU faculty, staff, or students whether NIH funded or non-funded.

The mission of the WCU IBC is to:

- Ensure that all recombinant DNA research conducted at the institution or sponsored by the institution is conducted in compliance with the National Institutes of Health Recombinant DNA Guidelines.
- Ensure that protocols of research involving Select Agents (defined by the Centers for Disease Control and Prevention), including but not limited to recombinant DNA, are reviewed and found to comply with all national, state, and local requirements.

The IBC, in conjunction with the Office of Safety and Risk Management, is authorized to inspect research facilities, approve research practices and procedures, and to take action such as enforcement of cessation of laboratory or clinical research activities, in the event of an unsafe workplace situation.

Research that requires IBC approval includes:

- The use of recombinant or synthetic nucleic acid molecules
- The use of human or animal pathogens
- Use of soil, seed or plant pathogens, field releases of plant pests, or field releases of genetically modified organisms received under a USDS APHIS permit
- The use of biological toxins (any toxic substance produced by a living organism)
- Use of experimental biological products administered to animals

The IBC responsibilities are broken down as follows:

Chairperson, Institutional Biosafety Committee

- Ensure that the Institutional Biosafety Committee is properly constituted and fulfills its requirements under the appropriate regulations, rules, etc.
- Ensure that all members of the IBC are adequately trained in appropriate containment practices, secondary containment procedures, and accidental spill containment procedures to fulfill their responsibilities as a member of the IBC.
- Call and preside over meetings of the IBC.
- In conjunction with the Safety Office and Research Compliance Officer, review and ensure compliance of all authorized researchers utilizing biohazardous materials.
- Review all instances of noncompliance and recommend corrections to the University.

Institutional Biosafety Committee

- Upon request, advise the Chancellor, Provost, Associate Provosts, Deans, and Department Chairs on matters related to biohazards and biosafety within their respective areas of responsibility.
- Review and recommend policies and procedures for biological risk assessment and biological risk reduction throughout the University.

- Review emergency plans for the containment and resolution of accidental spills and other related emergencies with an emphasis on risk reduction, personnel protection, and environmental protection.
- Oversee all research activities involving recombinant and synthetic nucleic acid molecules including review and approval prior to initiation, annual reviews, and updates. In conjunction with the Office of Safety and Risk Management, review laboratory safety equipment, safety procedures, and certification of compliance with all applicable rules and regulations governing the use of recombinant DNA. Approve those research projects that are found to conform with NIH Guidelines, OSHA and the CDC including (a) an independent assessment of containment levels required by the NIH Guidelines for the proposed research; and (b) assessment if applicable, of the facilities, procedures, practices, training and expertise of personnel involved in the proposed use of infectious biological agents.
- Review of Curriculum Vitae to ensure that all Principal Investigators are sufficiently trained and experienced in appropriate containment practices, secondary containment practices, and their responsibilities as Principal Investigators as applicable to the submitted protocol.
- Advise and provide technical expertise as necessary to the Safety Officer on matters involving biosafety.
- Conduct investigation of serious violations or problems and make recommendations to the Chancellor for the resolution of continued non-compliance or serious infractions.

Office of Safety and Risk Management

- The Office of Safety and Risk Management staff will support the IBC in carrying out its mission.
- Investigate laboratory accidents and report problems, violations, and injuries or illnesses associated with biohazardous research activities to the IBC.
- Develop, recommend, and implement policies and procedures for biological risk assessment and biological risk reduction throughout the University.
- Develop emergency plans for the containment and resolution of accidental spills and other related emergencies with an emphasis on risk reduction, personnel protection, and environmental protection.
- Provide advice and assistance to the IBC and PI concerning containment procedures and practices, laboratory security, recommended laboratory containment equipment, rules, regulations, and other matters as may be necessary.
- Provide oversight and assurance that laboratory safety containment is functioning properly.
- Serve as a member of the IBC.
- Provide industrial hygiene and safety support for all laboratory operations.
- Conduct periodic inspections of laboratories to ensure compliance with established procedures.
- Ensure transportation and disposal of all infectious waste in compliance with all applicable federal, state, and local ordinances.
- Assist, as necessary, in the emergency response, cleanup, and decontamination of biological spills and accidents.
- Provide occupational health and safety training.

Office of Research Administration

- Provide the necessary liaison between PIs, the IBC, granting agencies, and regulatory agencies.
- Serve as the administrator for the IBC and as a non-voting member.
- Maintain documentation of IBC reviews, approvals, and meeting minutes.
- Provide all necessary documentation, forms, regulatory guidelines and regulations for the Principal Investigator.
- Notify the Principle Investigator of the results of the IBC review.
- Provide annual reports to institutional leadership.

Principal Investigators (PI)

- Ensure compliance with appropriate National Institute of Health guidelines and all conditions stated in the protocol approved by the IBC.
- Submit protocol applications for all activities or modifications of activities involving biohazardous materials and obtain approval by the IBC prior to initiation of the activities or modifications.
- Ensure that all laboratory staff, including students, are trained in the accepted procedures, laboratory practices, containment methods, disinfectant and disposal practices, and required actions in the event of an accidental spill.
- Develop a Laboratory Safety Plan, including an emergency action plan for accidents and spills, in accordance with the Laboratory Specific Chemical Hygiene Plan (refer to Western Carolina Universities Chemical Hygiene Plan for more information).
- Ensure compliance with all shipping requirements for biological agents and toxins.
- Ensure proper handling and disposal of all infectious wastes.
- Request immunizations for laboratory personnel when working with biological agents for which there is an effective vaccine available. When required, maintain documentation of immunizations or titers. Agent specific plans and vaccines should be included in the Lab Specific Plan. Specific treatment plans or vaccines, if needed, should be made available to employees through University Health Services.
- Maintain all biosafety equipment in appropriate operating condition. Decontaminate laboratory equipment prior to maintenance or disposal.
- Maintain records of microorganisms and toxins used in the laboratory and biosafety cabinets.

Laboratory Safety – All other employees and students

- Follow all procedures and containment methods established for the activities conducted.
- Properly utilize all laboratory protective equipment including proper clothing, personal protective equipment, and containment devices.
- Report all accidents and spills to the PI or the Institutional Biosafety Officer as soon as possible.
- Report unsafe conditions, near misses, or opportunities to improve safety to the PI, IBC, or Safety Office immediately.

rDNA Registration Process

All research involving exempt and non-exempt rDNA experiments must be registered with and approved by the IBC. All approved projects must be renewed annually. Further guidance and the application for recombinant DNA research at WCU is available from the [WCU Institutional Biosafety](#) webpage.

General Laboratory Procedures

Review the general laboratory procedures for biosafety and rDNA work found in Appendix G of the NIH Guidelines.

- Strict adherence to good microbiological practices, aseptic techniques, and knowledge of the biology of the organisms used in the experiments so that the potential biohazards can be understood and appreciated.
- Any research group working with potential biohazards shall have an emergency plan describing the procedures to follow if an accident contaminates personnel or the environment, as well as treatment protocols and immunizations effective against the biohazard agents in use.
- For particularly infectious agents, it is recommended that personnel carry a card in their wallet indicating “I work with the following infectious agents.....” if they are found unconscious.
- Physical containment to confine organisms containing recombinant or synthetic nucleic acid molecules to reduce the potential for exposure.
- The level of biological containment is dependent on the host-vector system used in the experiment; therefore, laboratory practices, containment equipment, and special laboratory design guidelines should be followed based on the biosafety level required, and are detailed in Appendix G of the NIH Guidelines.

Incident Response and Reporting

The NIH requires institutions to report incidents involving rDNA materials including loss, theft, or release. This includes both NIH exempt and non-exempt rDNA materials.

- Report any loss, theft, or release involving rDNA materials to the Office of Research Administration Compliance Officer.
- Report any human exposure to infectious biological agents to the Safety and Risk Management Office and complete the Report of Occupational Injury or Illness form.

rDNA Research Training

The following training is available for employees working with rDNA:

- Lab-specific orientation and training is provided directly by the Principal Investigator or Supervisor.
- Biosafety training is for those who handle infectious material or other potentially infectious material (OPIM) that poses a splash, splatter, or percutaneous exposure hazard.
- Recombinant DNA training is required for those who conduct research with recombinant or synthetic nucleic acid molecules.

- Bloodborne Pathogens training is required for those who handle materials of human origin (i.e. primary and well-established cell lines).
- Animal risk training (IACUC) for personnel handling vertebrate animals.

Section 9: Biological Waste Management

Appropriate biological waste handling practices at Western Carolina University are based on compliance with OSHA regulations to protect employees who handle the waste and the North Carolina Medical Waste Regulations to ensure appropriate disposal.

Biological waste disposal guidelines are detailed in WCU's Laboratory Waste Management Plan (LWMP).

Autoclave Safety

Autoclaving, or steam sterilization, is the most dependable procedure for the destruction of all forms of microbial life. Autoclave safety guidelines are detailed in WCU's Laboratory Waste Management Plan (LWMP).

Section 10: Biohazard Spill Cleanup

During spill cleanup, be especially cautious of sharps. Always remove sharps with mechanical means (pieces of cardboard, tongs, etc.) and do not pick them up with your hands.

Blood or Body Fluids

- Don all appropriate PPE. Disposable gloves are required, shoe covers and face masks may be necessary.
- Absorb fluids with disposable towels. Place materials in a red biohazard bag.
- Clean area of all visible fluids with soap and water.
- Decontaminate area with a freshly prepared 10% bleach solution or Tuberculocidal disinfectant.

BSL-2 Microorganism

- Alert people in immediate area of the spill and request that they leave.
- Don all appropriate PPE. Disposable gloves are required, shoe covers and face masks may be necessary.
- Cover spill with disposable absorbent (towels or inert loose material).
- Carefully pour a freshly prepared 10% bleach solution around the edges of the spill and then into the center of the spill. Do not splash. Leave for 20 minutes.
- Using disposable paper towels, wipe up the spill, working from the outside towards the center. Dispose of materials in a red biohazard bag.
- Clean spill area with fresh towels soaked in an approved disinfectant or 10% bleach solution and allow to air dry. Place these materials in a red biohazard bag.

Section 11: Safety Training Requirements

Safe use of hazardous materials requires knowledge of risks to the researcher, campus community, and environment. Researchers learn to handle hazardous materials safely during their scientific training and

experience, as well as through information and training provided by their supervisors and the University's Safety and Risk Management Office. Principal Investigators and Lab Supervisors are responsible for the safety training of their lab personnel and must ensure that they document the training provided.

General Lab Safety Training: Each laboratory employee and student is required to take the general laboratory safety course before beginning work in the lab and every two (2) years thereafter.

Laboratory Specific Safety Training: The lab supervisor shall conduct laboratory-specific hazard awareness training for each employee or student working in the lab before that person begins work. Training must cover all items specified in the Laboratory Specific Chemical Hygiene Plan. This hazard awareness training must be documented and shall be reviewed and updated any time a new hazard is introduced in the laboratory.

Biological Safety Training: Employees and students working with infectious agents in a BSL-2 lab are required to complete biosafety training. Hands-on training provided directly by the PI/Supervisor for safe microbiological practices should also be documented.

Bloodborne Pathogens Training: Employees and students working with human derived potentially infectious materials (blood, body fluids, unfixed cells and tissues, etc.) are required to complete Bloodborne Pathogens training.

Section 12: Facility Design

Furniture Selection

The following guidelines should be used when selecting new furniture for laboratories. If there is a question on furniture selection, contact the Safety and Risk Management Office. No furniture used in laboratories that has been exposed to corrosive, toxic, or flammable chemicals or biological hazards may be repurposed for use in an office setting. Furniture that is no longer needed must be disposed of or relocated to another laboratory that its design and construction is suitable for.

Casework Materials:

- Metal or hardwood (such as oak or other approved equivalent) may be used in general research and teaching laboratories where humidity and temperature will be normal (standard for occupied rooms), and where biohazardous, flammable, corrosive, or toxic substances will not be absorbed into the surface.
- Plastic laminate may be used in miscellaneous storage and workrooms requiring base or wall storage facilities, and where the infusion of appropriate colors may be architecturally desirable.
- Only non-combustible and non-reactive chemical resistant laminates and resins may be used where biohazardous, flammable, corrosive, or toxic chemicals are to be used or stored.
- Millwork shall not be considered for new construction. Variances may be considered on renovation projects on a case-by-case basis.

Counter Tops:

Type 1	Type 2	Type 3	Type 4	Type 5	Type 6	Type 7
Composition Stone with a chemical resistant resin finish	Natural Quarry Stone with a chemical resistant resin finish	Solid Resin for chemical resistant surfaces	Wood fiber or wood particle board core, with chemical resistant finish on all exposed surfaces	Plastic Laminate with a wood particle core; may be self-edged or post-formed	Stainless Steel. Type 316 polished stainless steel, approved on a case-by-case basis.	Composition Stone with a low gloss vinyl sealer

Chemical Reaction and Abuse Resistance:

Type 1: Composition Stone with a chemical resistant resin finish

Type 2: Natural Quarry Stone with a chemical resistant resin finish

Type 3: Solid Resin for chemical resistant surfaces and in the bottom of general purpose fume hoods.

General Purpose Areas where neither chemical nor physical abuse is expected and where no liquids or biological hazards are to be used (such as writing surfaces, instrument support surfaces, or storage areas) shall use either of the following:

Type 4: Wood Core, a wood fiber or wood particle board core, with chemical resistant finish on all exposed surfaces

Type 5: Plastic Laminate with a wood particle core; may be self-edged or post-formed.

Radiation and Other Special Uses: Areas where radioactive materials or other special uses are approved shall use the following:

Type 6 – Stainless Steel – Type 316 polished stainless steel counter top surfaces may be approved on a case-by-case basis.

Physical Abuse Resistance: Areas where abrasive physical abuse is expected; Physics, Earth Sciences, Geology shall use:

Type 3 – Solid Resin with a chemical resistant surface, or

Type 7 – Composition Stone with a low gloss vinyl sealer

Fume Hood Work Surfaces:

General Purpose Hoods – Type 3: Solid Resin (chemical resistant)

Radiation Hoods – Type 6: Type 316 Stainless Steel

Perchloric Acid Hoods – Type 6: Type 316 Stainless Steel

Special Purpose Hoods – Type 3, Solid Resin (chemical resistant)

Chairs: Laboratory seating should be upholstered with vinyl or be constructed of solid materials such as plastic or wood that has been sealed to render it non-porous. Finishes shall be as resistant as possible to the corrosive chemical activity of chemicals used in the laboratory, as well as disinfectants.

Natural or synthetic fabric upholstery is not acceptable for use in a laboratory.

Chairs for working at laboratory benches, computer workstations, or biological safety cabinets should have the following adjustments:

- Pneumatic height adjustment
- Adjustable lumbar support
- Adjustable foot ring
- Adjustable seat pan depth

If the chair has arms, the arms should have the following adjustments:

- Adjustable height arms (small “T” style)
- Adjustable arm width

Cold Rooms

Cold rooms are commonly shared by multiple researchers; therefore, it is important that users take responsibility in maintaining the area to ensure a clean and safe environment.

Mold Growth: If mold growth occurs it can quickly spread throughout the space and into other areas of the building. Mold can directly contaminate research samples.

Minimize mold growth: Remove all wood, cardboard and paper products. These materials can absorb moisture and, because they are composed of cellulose, become a perfect breeding ground for mold.

Mold can thrive on any organic medium. Promptly clean up any spilled liquids. Keep surfaces clean. Never use bleach on metal surfaces as this can result in pitting corrosion.

For minor cleaning use a wet cleanup method (e.g., dampen cloth with a non-ammoniated soap or detergent). Dry surfaces to ensure moisture has been removed. If mold reappears soon after cleaning, then remove contaminated items and use any hospital approved disinfectant on mold contaminated surfaces.

Chemical hazards: Most cold rooms have closed air circulation and thus are not adequately vented. Improperly stored chemicals, particularly volatile toxic chemicals, could present inhalation risks to the workers in the space. Cold rooms may contain ignition sources, another risk if flammable materials are present. Furthermore, corrosive chemicals can damage the refrigeration system resulting in loss of cooling capability and subsequent research sample degradation.

Structural damage: Excess moisture or spilled chemicals can cause rust, corrosion, or other damages to the building and lab equipment resulting in costly repairs and loss of research productivity.

To prevent common problems from occurring, users should follow the cold room usage guidelines:

- Never store food/beverages in a cold room.
- Never use compressed gas in a cold room due to oxygen displacement and risk of asphyxiation.
- Never store dry ice in a cold room. Dry ice can create an oxygen deficient atmosphere when it sublimates and releases carbon dioxide gas.
- Keep the cold room door firmly shut at all times.
- Do not store flammable, toxic, or corrosive chemicals in a cold room.
- Do not store items in cardboard boxes. Store any paper products in air-tight plastic containers.
- Clean up spills promptly.
- Organize the cold room to establish ownership and responsibility for materials being stored.
- Keep electrical cords to a minimum and ensure GFCIs are installed on electrical outlets.
- Keep all surfaces clean and dry.
- Regularly remove items which are no longer needed or have expired.
- Report condensation buildup or water leaks immediately to Facilities Management 828-227-7442.

Section 13: Laboratory Equipment

Biological Safety Cabinet (BSC)

These cabinets are primarily intended to protect employees from biological hazards and should not be used for chemical hazards. BSCs are the most commonly used primary containment devices in microbiological laboratories. There are three classes of BSCs (Class I, II, and III). Each class provides different levels of protection. Because WCU utilizes BSL-1 and BSL-2 agents, it is our policy to default to the use of Class II BSCs to ensure consistent protection. A Class II Biological Safety Cabinet (BSC) uses HEPA filtered, laminar airflow to provide operator, environmental and sample protection. For the purpose of sterility, HEPA filters are typically rated at 99.99+% efficiency for particles 0.3 micron in size.

Standard Practices when utilizing BSCs

- Keep front and rear perforated grills free of clutter.
- Avoid installing BSCs near windows or doorways.
- Avoid sudden movements in and out of the cabinet. Also, always enter straight forward, without sweeping motions.
- Do not use gas burners or volatile chemicals inside the BSC.
- Do not store items on top of the cabinet.
- Disinfect the interior of the cabinet before and after use.
- Change HEPA filters as necessary.

BSC Open Flame Policy

Typical microbiological procedures often utilize Bunsen burners or other open flames to sterilize and/or reduce cross contamination; however, the use of such open flames inside of a BSC is NOT recommended for several reasons:

- The Class II BSC maintains sample protection through delivery of downward laminar airflow (volumes of air traveling in a single direction at a constant speed, without turbulence) over the work area of the cabinet. Hot air rises, so any open flame causes air to rise against the laminar down-flow, creating turbulence and foiling the BSC's ability to protect the samples in the work area.
- If an open flame gets too hot, it also has the capacity to melt the bonding agent holding the HEPA filter media to its frame. This destroys the HEPA filter's effectiveness, leading to loss of containment in the positively pressured plenum.
- If the flame goes out and the gas supply valve remains open, flammable gas would be introduced to the cabinet unabated. In a Biosafety Cabinet, where 70% of the air within is recirculated, concentrations of the flammable gas could reach explosive potential and pose a serious risk to not only the BSC, but to the user and the laboratory it occupies.

Several measures can be taken to reduce the chance for cross-contamination of materials when working in a BSC. Opened tubes or bottles should not be held in a vertical position. Investigators working with Petri dishes and tissue culture plates should hold the lid above the open sterile surface to minimize direct impaction of downward air. Bottle or tube caps should not be placed on the towelings. Items should be recapped or covered as soon as possible.

Open flames are not required in the near microbe-free environment of a biological safety cabinet; however, if a flame is deemed absolutely necessary, there are types of equipment widely available that are safer alternatives to the Bunsen burner. Some of these employ low profile, pedal attenuated flames, and others detect motion. Small electric "furnaces" are available for decontaminating bacteriological loops and needles and are preferable to an open flame inside the BSC. Disposable or recyclable sterile loops should be used whenever possible.

Certification of BSCs

BSCs are to be certified by a Safety Office approved vendor. All cabinets in which human materials and infectious (or potentially infectious) materials are being manipulated should be certified annually. All newly purchased or recently moved cabinets must be certified before they can be used. Costs associated with certification are the responsibility of the department that the BSC belongs to.

Inhalation of infectious aerosols is a significant route of laboratory acquired infection and warrants special consideration. Aerosolization can occur through lab activities including, but not limited to pipetting, centrifuging, opening of ampoules, and shaking.

Centrifuges

During centrifugation, aerosols can escape during the high-speed spin process if not adequately contained within the unit. Filling centrifuge tubes, removing caps or lids after centrifugation, and removing

supernatant liquid and resuspension of pellets can also lead to the release of aerosols into the laboratory environment. The following precautions must be considered to minimize aerosol formation during the centrifuging process:

- Use a centrifuge with aerosol-tight rotors and buckets when handling potentially infectious substances.
- Always follow manufacturer guidelines for use and maintenance of the centrifuge.
- Over time the aerosol-tightness of rotors will deteriorate. Visually inspect the equipment for unusual cracks, irregularities, and wear prior to each use.
- Ensure the vessel's material is resistant to the liquid being centrifuged. Inspect that the tube is not damaged and ensure that it fits properly into the rotor bore. Use aerosol-tight caps or lids.
- Use plastic tubes instead of glass tubes as plastic is less likely to break and can be closed more tightly.
- Don't exceed the maximum filling volume for the vessel. If filled too high, the liquid may touch the lid and form a droplet that could lead to aerosol formation.
- Clearly label all tubes for identification. It is best to label the tube directly and not use labels that can fall off during the spin.
- Wipe the outside of the tube with an approved disinfectant before placing it in the centrifuge.
- Always verify proper loading of specimens to maintain balance. Incorrect loading can reduce the lifetime of the rotor and heavy vibrations can lead to permanent damage. Balance the tubes according to weight. Load the rotor symmetrically and ensure the opposing tubes are the same type and are filled with the same mass. Counterbalance with water tubes if necessary.
- During operation, listen for unusual noises or vibrations until the programmed speed is reached.
- Don't exceed the maximum speed tolerance of the vessel as this could lead to tube breakage.
- Always wait at least 10 minutes before opening the bucket or rotor to allow any aerosols to settle.
- If breakage occurs, wait at least 30 minutes before opening the aerosol tight lid or rotor cap to allow the aerosols to settle.
- If possible, load and unload the rotor in a Biological Safety Cabinet (BSC) to contain potential aerosols inside the cabinet.
- Clean up spills and disinfect immediately if visible contamination is evident. Use cleaning and decontaminants that are recommended by the manufacturer to prevent equipment damage from aggressive chemicals.
- Routinely decontaminate buckets, caps, rotors, lids, and interior surfaces with an approved disinfectant and follow manufacturer directions.

Blenders and Homogenizers

These items are frequently used in laboratories, and both can potentially produce aerosols. Safety seals homogenizers and blenders are available and should be used when working with agents that could be transmitted via aerosols. They may be used on an open benchtop; however, they must be opened in a BSC. All non-sealed devices must be used exclusively in a BSC.

Furniture and Equipment Surplus or Disposal

Review the Laboratory Waste Management Plan (LWMP) section for Laboratory Equipment Disposal and Surplus Property. The University has procedures in place to ensure the safety of personnel picking up surplus equipment as well as requirements to ensure the proper disposal regulations are followed.

Section 14: Bloodborne Pathogens

Any research, diagnostic, or teaching activity conducted with material derived from humans including blood, body fluids, unfixed tissues, primary or established cell lines, and other potentially infectious materials (OPIM) requires the PI to implement the University's Bloodborne Pathogens Exposure Control Plan. This plan addresses all of the provisions of the Occupational Safety and Health Administration's (OSHA) Occupational Exposure to Bloodborne Pathogens Standard (29CFR 1910.1030) and is implemented by the Office of Safety and Risk Management. To be compliant with OSHA Bloodborne Pathogens requirements the program provides the following:

- Information about Bloodborne Pathogens and risks for potential exposure
- Universal Precautions approach and safe work practices
- Hepatitis B Vaccination Program
- Annual Bloodborne Pathogens training
- Post-Exposure procedures and follow-up

Information and forms for the Bloodborne Pathogens Program are available on the [Safety and Risk Management](#) website.

Appendix A: Emergency Contacts

In the event of an emergency affecting campus, the [Campus Emergencies](#) webpage is the official source for WCU emergency related information.

Emergency Telephone Numbers

	Normal Business Hours	Evenings/Weekends
EMERGENCY Fire/Police/Medical	828-227-8911 or 911	828-227-8911 or 911
University Police Department NON-EMERGENCY	828-227-7301	828-227-7301
Safety and Risk Management	828-227-7443	828-227-7443
Chemical Spill	828-227-7443	828-227-7443
Biological Spill	828-227-7443	828-227-7443
Radiation Exposure	828-227-7443	828-227-7443
Workers' Compensation	828-227-7443	828-227-7443
NC Poison Control Center	1-800-84 TOXIN (1-800-848-6946)	1-800-84 TOXIN (1-800-848-6946)
N.C. Radiation Protection Section	919-814-2250	800-858-0368 Emergency after hours
Jackson County Department of Public Health	828-586-8994	8:00am - 5:00pm Monday-Friday
Work Management Centers		
Facilities Management	828-227-7442	828-227-7224
WCU Health Services	828-227-7640	828-227-8911 EMS