

# BIOSAFETY LABORATORY MANUAL

## BIOSAFETY LEVEL 2 – BMBL 6<sup>TH</sup> EDITION



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## (BMBL 6<sup>TH</sup> EDITION)

### Table of Contents

Table of Contents .....	2
I. Introduction .....	4
1. What is Biosafety? .....	4
2. What is a Biohazardous Material? .....	4
3. Risk Groups (RG) - 4 levels.....	5
4. Biosafety Levels (BSL) - 4 levels .....	6
II. Standard Microbiological Practices .....	8
III. Special Practices .....	10
IV. Safety Equipment (Primary Barriers and Personal Protective Equipment) .....	11
V. Laboratory Facilities (Secondary Barriers).....	11
VI. Exposure Control .....	12
1. Administration .....	12
2. Containment.....	12
3. Practices and procedures.....	13
VII. Biohazards.....	13
1. What are biohazards?.....	13
2. Bloodborne Pathogens (BBP).....	13
3. Cell Culture/Recombinant DNA/Viral Vectors .....	13
VIII. Sharps .....	14
1. What are sharps? .....	14
2. Safe handling of sharps.....	14
IX. Biohazard Waste .....	14
X. Autoclave .....	14
XI. Training.....	15
XII. Other .....	16
Appendices.....	17
Appendix – A Standard Operating Procedures (SOP).....	17
1. BIOLOGICAL SPILL KIT.....	17

2. SOP - Spill Response and Clean-up Outside Biosafety Cabinet .....	17
3. SOP - Spill Response and Clean-up Inside Biosafety Cabinet .....	18
4. SOP - Processing Biohazardous Regulated Medical Waste in Biosciences .....	19
Appendix B – Laboratory Specific Biosafety Training .....	22
1. Autoclave Safety .....	22
2. Biological Safety Cabinet (BSC) .....	23
3. Laboratory Specific Training Form .....	25
Appendix C – Risk Assessment .....	26
PART A: Research/Investigation .....	26
PART B: Characterization of Agent .....	26
PART C: Characterization of Staff/Protocols .....	28
PART D: Characterization of Facilities/Equipment .....	29
PART E. Risk Assessment/Final Analysis/Approval .....	30
Appendix D – Administrative Information Form .....	31
Appendix E – Injuries .....	32
Appendix F – Biohazard Incident Report Form .....	33
Appendix G – Laboratory Specific Biosafety Manual .....	35

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### I. Introduction

#### 1. What is Biosafety?

Biosafety addresses the safe handling and containment of infectious microorganisms and hazardous biological materials. The practice of safe handling of pathogenic microorganisms and their toxins in biological laboratories is accomplished through the application of risk assessment, containment principles, and laboratory microbiological and special practices ([Biosafety in Microbiological and Biomedical Laboratories \(BMBL\)](#), 6<sup>th</sup> edition).

#### 2. What is a Biohazardous Material?

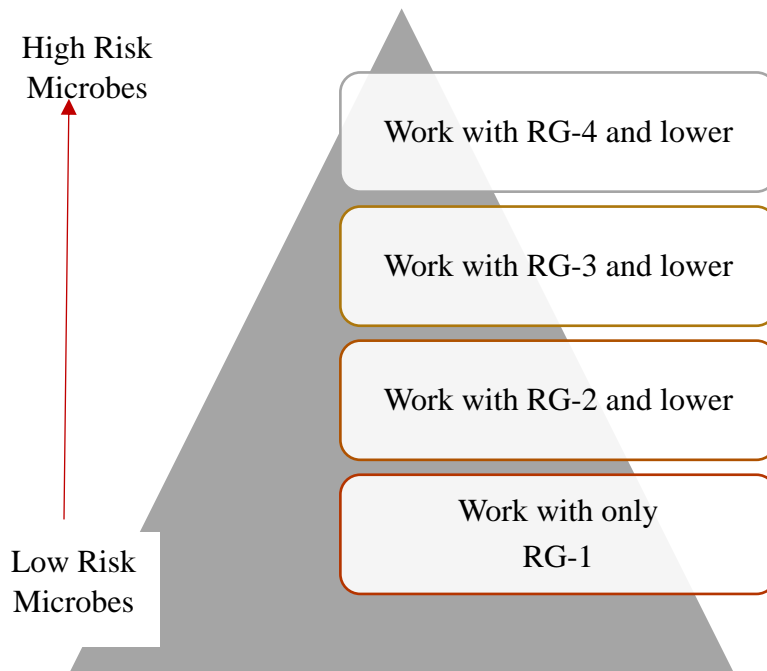
A biohazardous material is an infectious agent or hazardous material that presents a risk or potential risk to the health of humans, animals, or the environment ([CDC, 2011](#)).

Biohazardous agents include the following:

- a. Pathogenic agents including bacteria, fungi, viruses, protozoa, parasites, prions, and [toxins](#).
- b. Recombinant or synthetically derived nucleic acids, including those that are chemically or otherwise modified analogs of nucleotides (e.g., morpholinos) or both. The [NIH](#) defines synthetically derived nucleic acid molecules as follows:
  - i. Molecules that are constructed by joining nucleic acid molecules that can replicate in a living cell (i.e., recombinant nucleic acids).
  - ii. Nucleic acid molecules that are chemically or otherwise modified but can pair with naturally occurring nucleic acid molecules (i.e., synthetic nucleic acids).
  - iii. Molecules that result from the replication of those described in (a) or (b) above.
- c. Recombinant DNA molecules, organisms, vectors (e.g., plasmids, viral vectors), and viruses containing recombinant DNA molecules.
- d. Human and non-human primate blood, tissue, body fluid, and cell culture (primary and established cell lines).
- e. Plants, animals, or derived waste which contains or may contain pathogenic hazards.

### 3. Risk Groups (RG) - 4 levels

Defining the agents in use is a starting point for risk assessment and assignment of Biosafety Levels (BSL).



\*Exceptions may be made based on risk assessment

**RG-4- Are associated** with potentially lethal human disease for which preventatives or therapeutics are not readily available.

**RG-3- Are associated** with serious or lethal human disease for which preventatives or therapeutics may be available.

**RG-2- Are associated** with diseases that are rarely serious and for which preventatives or therapeutics are often available.

**RG-1- Are not associated** with disease in healthy adult humans or animals.

#### 4. Biosafety Levels (BSL) - 4 levels

<b>BSL</b>	<b>Agents</b>	<b>Special Practice <sup>a</sup></b>	<b>Primary Barrier and Personal Protective Equipment <sup>a</sup></b>	<b>Facilities (Secondary Barriers) <sup>a</sup></b>
1	Well-characterized agents not known to consistently cause disease in immunocompetent adult humans and present minimal potential hazard to laboratory personnel and the environment	Standard microbiological practices	No primary barriers required; protective laboratory clothing; protective face, eyewear, as need	Laboratory doors; sink for handwashing; laboratory bench; windows fitted with screens; lighting adequate for all activities
2	Agents associated with human disease and pose moderate hazards to personnel and the environment	Limited access; occupational medical services including medical evaluation, surveillance, and treatment, as appropriate; all procedures that may generate an aerosol or splash conducted in a BSC; decontamination process needed for laboratory equipment	BSCs or other primary containment device used for manipulations of agents that may cause splashes or aerosols; protective laboratory clothing; other PPE, including respiratory protection, as needed	Self-closing doors; sink located near exit; windows sealed or fitted with screens; autoclave available

3	Indigenous or exotic agents; may cause serious or potentially lethal disease through the inhalation route of exposure	Access limited to those with need to enter; viable material removed from laboratory in primary and secondary containers; opened only in BSL-3 or ABSL-3 laboratories; all procedures with infectious materials performed in a BSC	BSCs for all procedures with viable agents; solid front gowns, scrubs, or coveralls; two pairs of gloves, when appropriate; protective eyewear, respiratory protection, as needed	Physical separation from access corridors; access through two consecutive self-closing doors; hands-free sink near exit; windows are sealed; ducted air ventilation system with negative airflow into laboratory; autoclave available, preferably in laboratory
4	Dangerous and exotic agents that pose high individual risk of aerosol-transmitted laboratory infections and life-threatening disease that are frequently fatal, for which there are no vaccines or treatments; and related agents with unknown risk of transmission	Clothing change before entry; daily inspections of essential containment and life support systems; all wastes decontaminated prior to removal from laboratory; shower on exit	BSCs for all procedures with viable agents; solid front gowns, scrubs, or coveralls <sup>b</sup> gloves <sup>b</sup> full-body, air-supplied, positive-pressure suit	Entry sequence; entry through airlock with airtight doors <sup>c</sup> walls, floors, ceilings form sealed internal shell; dedicated, non-recirculating ventilation system required; double-door, pass-through autoclave required

- a. Each successive BSL contains the recommendations of the preceding level(s) and the criteria in the cell.
- b. Applies to Cabinet Laboratory.
- c. Applies to Suit Laboratory.

## II. Standard Microbiological Practices

1. The laboratory supervisor enforces the institutional policies that control safety and access to the laboratory.
2. The laboratory supervisor ensures that laboratory personnel receive appropriate training regarding their duties, potential hazards, manipulations of infectious agents, necessary precautions to minimize exposures, and hazard/exposure evaluation procedures (e.g., physical hazards, splashes, aerosolization) and that appropriate records are maintained. Personnel will receive annual updates and additional training when equipment, procedures, or policies change. All persons entering the facility are advised of the potential hazards, are instructed on the appropriate safeguards, and will read and follow instructions on practices and procedures. The laboratory supervisor ensures that the [CMU Laboratory Entry and Access Policy](#) is followed.
3. Personal health status may affect an individual's susceptibility to infection and ability to receive available immunizations or prophylactic interventions. Therefore, all personnel, and particularly those of reproductive age and/or those having conditions that may predispose them to increased risk for infection (e.g., organ transplant, medical immunosuppressive agents), are provided information regarding immune competence and susceptibility to infectious agents. Individuals having such conditions are encouraged to self-identify to the institution's healthcare provider for appropriate counseling and guidance.
4. A safety manual specific to the facility is prepared or adopted in consultation with the facility director and appropriate safety professionals. The safety manual is available, accessible, and periodically reviewed and updated as necessary.
  - a. The safety manual contains sufficient information to describe the biosafety and containment procedures for the organisms and biological materials in use, appropriate agent-specific decontamination methods, and the work performed.
  - b. The safety manual contains or references protocols for emergency situations, including exposures, medical emergencies, facility malfunctions, and other potential emergencies. Training in emergency response procedures is provided to emergency response personnel and other responsible staff according to institutional policies.
5. A sign incorporating the universal biohazard symbol is posted at the entrance to the laboratory when infectious materials are present. Posted information includes: the laboratory's Biosafety Level, the supervisor's or other responsible personnel's name and telephone number, PPE requirements, general occupational health requirements (e.g., immunizations, respiratory protection), and required procedures for entering and exiting the laboratory. Agent information is posted in accordance with the institutional policy.
6. Long hair, loose clothing and any other material (bracelets, etc.) that could pose a safety hazard should be restrained so they cannot contact hands, specimens, containers, or equipment.
7. Gloves are worn to protect hands from exposure to hazardous materials.
  - a. Glove selection is based on an appropriate risk assessment.
  - b. Gloves are not worn outside the laboratory.
  - c. Change gloves when contaminated, glove integrity is compromised, or when otherwise necessary.
  - d. Do not wash or reuse disposable gloves, and dispose of used gloves with other contaminated laboratory waste.



8. Gloves and other PPE are removed in a manner that minimizes personal contamination and transfer of infectious materials outside of the areas where infectious materials and/or animals are housed or manipulated.
9. Persons wash their hands after working with potentially hazardous materials and before leaving the laboratory.
10. Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption are not permitted in laboratory areas. Food is stored outside the laboratory area.
11. Mouth pipetting is prohibited. Mechanical pipetting devices are used.
12. Policies for the safe handling of sharps, such as needles, scalpels, pipettes, and broken glassware are developed, implemented, and followed; policies are consistent with applicable state, federal, and local requirements. Whenever practical, laboratory supervisors adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions are always taken with sharp items. These include:
  - a. Plasticware is substituted for glassware whenever possible.
  - b. Use of needles and syringes or other sharp instruments is limited in the laboratory and is restricted to situations where there is no alternative (e.g., parenteral injection, blood collection, or aspiration of fluids from laboratory animals or diaphragm bottles). Active or passive needle-based safety devices are to be used whenever possible.
    - i. Uncapping of needles is performed in such a manner to reduce the potential for recoil causing an accidental needlestick.
    - ii. Needles are not bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
    - iii. If absolutely necessary to remove a needle from a syringe (e.g., to prevent lysing blood cells) or recap a needle (e.g., loading syringes in one room and injecting animals in another), a hands-free device or comparable safety procedure must be used (e.g., a needle remover on a sharps container, the use of forceps to hold the cap when recapping a needle).
    - iv. Used, disposable needles and syringes are carefully placed in puncture-resistant containers used for sharps disposal immediately after use. The sharps disposal container is located as close to the point of use as possible.
  - c. Non-disposable sharps are placed in a hard-walled container for transport to a processing area for decontamination, preferably by autoclaving.
  - d. Broken glassware is not handled directly. Instead, it is removed using a brush and dustpan, tongs, or forceps.
13. Perform all procedures to minimize the creation of splashes and/or aerosols.
14. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant. Spills involving infectious materials are contained, decontaminated, and cleaned up by staff who are properly trained and equipped to work with infectious material. A spill procedure is developed and posted within the laboratory.
15. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method, consistent with applicable institutional, local, and state requirements. Depending on where the decontamination will be performed, the following methods are used prior to transport:

- a. Materials to be decontaminated outside of the immediate laboratory are placed in a durable, leak-proof container and secured for transport. For infectious materials, the outer surface of the container is disinfected prior to moving materials and the transport container has a universal biohazard label.
  - b. Materials to be removed from the facility for decontamination are packed in accordance with applicable local, state, and federal regulations.
16. An effective integrated pest management program is implemented.
  17. Animals and plants not associated with the work being performed are not permitted in the laboratory.

### **III. Special Practices**

1. Access to the laboratory is controlled when work is being conducted.
2. The laboratory supervisor is responsible for ensuring that laboratory personnel demonstrate proficiency in standard microbiological practices and techniques for working with agents requiring BSL-2 containment.
3. Laboratory personnel are provided medical surveillance, as appropriate, and offered available immunizations for agents handled or potentially present in the laboratory.
4. Properly maintained BSCs or other physical containment devices are used, when possible, whenever:
  - a. Procedures with a potential for creating infectious aerosols or splashes are conducted. These include pipetting, centrifuging, grinding, blending, shaking, mixing, sonicating, opening containers of infectious materials, inoculating animals intranasally, and harvesting infected tissues from animals or eggs.
  - b. High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory using sealed rotors or centrifuge safety cups with loading and unloading of the rotors and centrifuge safety cups in the BSC or another containment device.
  - c. If it is not possible to perform a procedure within a BSC or other physical containment device, a combination of appropriate personal protective equipment and administrative controls are used, based on a risk assessment.
5. Laboratory equipment is decontaminated routinely; after spills, splashes, or other potential contamination; and before repair, maintenance, or removal from the laboratory.
6. A method for decontaminating all laboratory waste is available (e.g., autoclave, chemical disinfection, incineration, or other validated decontamination method).
7. Incidents that may result in exposure to infectious materials are immediately evaluated per institutional policies. All such incidents are reported to the laboratory supervisor and any other personnel designated by the institution. Appropriate records are maintained.

## **IV. Safety Equipment (Primary Barriers and Personal Protective Equipment)**

1. Protective laboratory coats, gowns, or uniforms designated for laboratory use are worn while working with hazardous materials and removed before leaving for non-laboratory areas (e.g., cafeteria, library, and administrative offices). Protective clothing is disposed of appropriately or deposited for laundering by the institution. Laboratory clothing is not taken home.
2. Eye protection and face protection (e.g., safety glasses, goggles, mask, face shield or other splatter guard) are used for manipulations or activities that may result in splashes or sprays of infectious or other hazardous materials. Eye protection and face protection are disposed of with other contaminated laboratory waste or decontaminated after use.
3. The risk assessment considers whether respiratory protection is needed for the work with hazardous materials. If needed, relevant staff are enrolled in a properly constituted respiratory protection program.
4. In circumstances where research animals are present in the laboratory, the risk assessment considers appropriate eye, face, and respiratory protection, as well as potential animal allergens.

## **V. Laboratory Facilities (Secondary Barriers)**

1. Laboratory doors are self-closing and have locks in accordance with the institutional policies.
2. Laboratories have a sink for handwashing. It should be located near the exit door.
3. An eyewash station is readily available in the laboratory.
4. The laboratory is designed so that it can be easily cleaned.
  - a. Carpets and rugs in laboratories are not appropriate.
  - b. Spaces between benches, cabinets, and equipment are accessible for cleaning.
5. Laboratory furniture can support anticipated loads and uses.
  - a. Benchtops are impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
  - b. Chairs used in laboratory work are covered with a non-porous material that can be easily cleaned and decontaminated with an approved disinfectant.
6. Laboratory windows that open to the exterior are not recommended. However, if a laboratory does have windows that open to the exterior, they are fitted with screens.
7. Illumination is adequate for all activities and avoids reflections and glare that could impede vision.
8. Vacuum lines in use are protected with liquid disinfectant traps and in-line HEPA filters or their equivalent. See Appendix A, Figure 11. Filters are replaced, as needed, or are on a replacement schedule determined by a risk assessment.
9. There are no specific requirements for ventilation systems. However, the planning of new facilities considers mechanical ventilation systems that provide an inward flow of air without recirculation to spaces outside of the laboratory.
10. BSCs and other primary containment barrier systems are installed and operated in a manner to ensure their effectiveness. See Appendix A.

- a. BSCs are installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSCs are located away from doors, windows that can be opened, heavily traveled laboratory areas, and other possible airflow disruptions.
- b. BSCs can be connected to the laboratory exhaust system by either a canopy connection (Class IIA only) or directly exhausted to the outside through a hard connection (Class IIB, IIC, or III). Class IIA or IIC BSC exhaust can be safely recirculated back into the laboratory environment if no volatile toxic chemicals are used in the cabinet.
- c. BSCs are certified at least annually to ensure correct performance.

## VI. Exposure Control

### 1. Administration

- a. Before starting work, register work with the [Office of Research Compliance](#).
- b. Depending on the nature of the proposed research, approval may be required by IBC, IRB, and/or IACUC.
- c. Prior to operations, Principal Investigator (PI) and Laboratory Safety Specialist perform risk assessment (Appendix C).
- d. Laboratory Specific Biosafety Manual is available (Section II-4, Appendix G).
- e. Pathogen Safety Data Sheets are available (Appendices B and G).
- f. Standard Operating Procedures are available (Appendices A and G).
- g. Administrative Information is available (Appendix D)

### 2. Containment

- a. Primary containment for biological agents/aerosols (Section VI).  
Personal Protective Equipment,
  - 1) Nitrile gloves.
  - 2) Lab coat/apron.
  - 3) Eye protection.
  - 4) Closed Toe Shoes.
  - 5) Heat gloves for autoclave (Appendices A and B).
  - 6) See Appendix D-5 for lab specific requirements on PPE.
- b. Secondary containment for biological agents/aerosols (Section V).
  - 1) Approved lab facility/directional airflow.
  - 2) Annually certified Biosafety Cabinet (BSC) (Section X, Appendices A and B).
  - 3) Do not restrict the airflow of the cabinet.
  - 4) Do not use volatile chemicals unless ducted outside the lab.
  - 5) Do not use flames in the BSC.
  - 6) BSC Training Exam Required (Appendices B and G).
  - 7) Procedures with a potential for creating infectious aerosols or splashes must be conducted in the BSC.

### 3. Practices and procedures

- a. No food or drink in any lab.
- b. Appropriate attire and behavior.
- c. Long hair is restrained.
- d. Aseptic techniques.
- e. Sharps protocols.
- f. Labeling completed.
- g. Knowledge/training/SOPs.
- h. See Sections II and III for detailed recommendation on laboratory practice.

## VII. Biohazards

### 1. What are biohazards?

Biohazards are disease producing agents (pathogens) that can be transmitted to individuals through various routes of exposure (modes of transmission). Exposure to these hazards may result in acute or chronic health conditions ([CDC, 2022](#)).

### 2. Bloodborne Pathogens (BBP)

- a. Pathogenic microorganisms that may be present in human blood, fluids, tissues and cells and can cause disease in humans. Examples of Potential Pathogens are Hepatitis B and C, HIV.
- b. [CMU BBP Exposure Control Plan](#).
- c. Protection:
  - i. BBP training (Section XI).
  - ii. Aseptic technique & good lab practices.
  - iii. Exposure control and PPE/Barriers.
  - iv. Sharps protocols/safety sharps if available.
  - v. Vaccination option if available.
  - vi. Hand washing and housekeeping.

### 3. Cell Culture/Recombinant DNA/Viral Vectors

- a. Genetic engineering routinely involves in vitro incorporation of non-native genetic material into cells that may be of human origin.
- b. Main classes of viral vectors are retroviruses (RV), adenoviruses (AV), adeno-associated viruses (AAV), lentiviruses (LV), and herpes simplex viruses (HSV)
- c. Protection:
  - i. Aseptic techniques/good laboratory practice/aerosol control.
  - ii. BSC use for viruses and human cell lines.
  - iii. Disinfection of surfaces.
  - iv. Sharps protocols/safety sharps if available.
  - v. PPE/Barriers.
  - vi. Hand Hygiene:
    - Upon entering and leaving the lab,
    - Before and after cell culture work,
    - Before touching an unprotected part of your body,

- As needed.

## **VIII. Sharps**

### **1. What are sharps?**

Sharps are needles (and attached syringes), lancets, scalpel and razor blades (even if not contaminated) or any item (e.g., razor blades, cover slips, slides, glass, broken plastic & glassware or other devices) sharp enough to cutting or piercing skin.

### **2. Safe handling of sharps**

- a. Glass or Pasteur pipettes are considered glass sharps and should be placed in a cardboard box or suitable rigid container.
- b. Approved plastic biohazard sharps container must be used for needles, syringes, razor blades, & scalpel blades.
- c. Sharps container must have biohazard label.
- d. Sharps containers are dated when put into use, must be discarded within 90 days, are no more than 2/3 full, and must display proper labeling.
- e. Never bend, break or recap needles or blades.
- f. Never remove a needle from a syringe or blade from a scalpel by hand.
- g. Always discard used sharps in approved sharps container immediately after use.
- h. Never reach inside, overfill, or force items in sharps container.
- i. See the Section II - 12 for detailed recommendations on sharps.

## **IX. Biohazard Waste**

1. Proper labeling of all biohazard containers must include the universal biohazard symbol and/or the words “Biohazardous Waste” in letter 1 inch higher or greater.
2. Biohazardous waste is collected in appropriate containers, closeable, puncture-resistant, leak-proof, and properly labeled.
3. Solid, non-sharp biohazard waste is placed in appropriate containers lined with a biohazard bag.
4. Animal carcasses and pathological waste must be removed by a licensed waste hauler and incinerated.
5. Waste must be stored so it does not putrefy.
6. Biohazard waste containers must be closed when not in use.
7. Dispose of noninfectious waste in regular trash.

## **X. Autoclave**

1. Sterilize glassware, equipment and media.
2. Decontamination of used cultures, stocks and media, laboratory and regulated medical waste.
  - a. Get proper training from PI.
  - b. Watch ASU video and take exam (Appendices A and B).
  - c. Use only approved autoclave bags.
  - d. Do not overfill bags.

- e. Do not autoclave bleach, volatiles, solvents, or corrosives.
3. Protection.
  - a. Wear heat resistant PPE and eye protection.
  - b. Awareness of scalding liquids and hot surfaces.
  - c. Stand back and open door slowly.
  - d. Do not agitate liquids.
4. Solid Waste.
  - a. Contain waste in an autoclave bag and place in an autoclavable tub.
  - b. Use indicator tape or biological indicators periodically to validate waste cycles.
  - c. Waste should be autoclaved for at least 45 minutes.
5. Liquid Waste.
  - a. Treat with 10% bleach for 30 minutes or use other validated decontamination procedures.
  - b. Dispose to sanitary sewer as long as no heavy metals, solvents, alcohols or chemotherapeutics, or any other hazardous waste is in use.
6. Once appropriately autoclaved, biohazard waste is no longer considered hazardous and can be disposed of in the dumpster or liquids in the sanitary sewer. All decontaminated red biohazard bags are placed in a black trash bag prior to disposal.
7. See autoclave training and exam in Appendices A and B.

## **XI. Training**

Before starting to work in BSL2 laboratory, all laboratory workers are required to completed required safety trainings,

1. Read Laboratory Specific Biosafety Manual (Appendix G).
2. [Laboratory Safety Training - SuccessFactors](#) is required for working in laboratory.
3. [Biosafety training - CITI](#) is required for working in BSL2 laboratory.
4. [Bloodborne Pathogens \(BBP\) training - SuccessFactors](#) is required for exposure to sharps, blood, human tissues, cell lines, bodily fluids, or other potentially infectious materials.
5. Laboratory specific training (Appendix B) is required to be conducted by supervisor/PI in the laboratory.
6. Any other trainings recommended by your supervisor/PI and Laboratory Specific Biosafety Manual.
7. Consult the [Office of Laboratory and Field Safety \(OLFS\)](#) for help with the requirements of safety training.

## **XII. Other**

1. Carefully plan out all procedures and experiments.
2. Notify other users of unsafe practices before they hurt themselves or others.
3. Contact a facility manager if there is a dangerous situation.
4. In case of life-threatening emergency, please call 911.
5. Other Emergencies, please call emergency contact numbers listed on door signs or the biosafety manual (Appendix D – Administration Information and Appendix E – Injuries)
6. Reporting: In the event of any accidents, incidents, or spills please let lab supervisors, the Office of Research Compliance or the Office of Laboratory and Field Safety know immediately (Appendix F – Biohazard Incident Report Form).
7. Don't be afraid to ask for help.



# Appendices

## Appendix – A Standard Operating Procedures (SOP)

### 1. BIOLOGICAL SPILL KIT

In a 5-gallon bucket with lid, place the following items:

- Spill response and cleanup procedures (SOP)
- 1 Notepad
- 1 Pen
- 1 Permanent marker
- 1 trash bag
- 6 Biohazard stickers
- 1 roll duct tape
- 1 roll/pack absorbent paper towels
- 4 Absorbent pads (each holds 500ml)
- 1 Small sharps container
- 4 Biohazard waste bags and closures
- 1 hand broom with dustpan
- 1 squeegee
- 1 pair tongs
- Antibacterial soap
- Hand sanitizer
- 250 ml concentrated bleach less than 1 year old
- 1 Spray bottle with distilled water to mix bleach solution
- 6 pair nitrile gloves
- 2 Disposable waterproof apron
- 3 pair disposable shoe covers
- Splash goggles
- 2 N-95 respirators (NOTE: medical clearance & fit-testing required for respirator use)

Assemble the materials listed above in the biological spill kit before taking action. After the 30-minute evacuation period clean up the spill as soon as possible.

### 2. SOP - Spill Response and Clean-up Outside Biosafety Cabinet

#### RESPONSE

- REMAIN CALM!
- Alert people who are in the immediate area there has been a spill.
- Remove contaminated clothing as you leave the area (evacuate).
- Also try to isolate the spill as much as possible by closing doors, etc., if it does not delay your evacuation.
- Immediately wash your hands and any exposed areas with soap and warm water.

- Post a “DO NOT ENTER” sign outside the spill area and restrict access.
- Allow aerosols to settle (~30 minutes).
- Evaluate the agent’s specific hazards.
- Contact emergency services and the Office of Laboratory and Field Safety for assistance.
- Seek medical treatment if warranted (Appendix E).

### **SPILL CLEAN-UP**

- Assess the extent of the spill and formulate a plan for decontamination.
- Assemble a spill response kit and clean-up team.
- Put on gowns, gloves (double if needed), shoe covers, and appropriate personal protective equipment from spill kit.
- Starting at the perimeter of the spill and working toward the center, cover the spill with paper towels or other absorbent material.
- Slowly pour disinfectant over the absorbent material being careful to avoid splashing and spill starting around the edges and working toward the center. Saturate the area with the disinfectant.
- Allow sufficient contact time for the disinfectant to inactivate biohazardous agents; typically 15-20 minutes, but double the contact time for viscous materials or body fluid.
- Use a squeegee and paper towels to wipe up the spill, working from the edges to the center.
- Use tongs/forceps/dust pan to pick up sharp objects (broken glass, etc.) that may puncture gloves.
- Discard absorbent material used to clean-up the spill in biohazard waste bag. Use a bucket for large sharp objects or saturated absorbent materials.
- Clean the the spill area again using with fresh paper towels soaked in disinfectant. Thoroughly wet the spill area and allow to disinfect for approximately 15-20 minutes.
- Discard cleanup materials in biohazard bag, along with any contaminated PPE.
- Any re-useable and autoclavable materials that are contaminated should be placed in a separate bag or autoclavable container for sterilization.
- Close and secure bag, then place bag in second biohazard bag. Secure outer bag and disinfect by autoclaving (steam sterilization).

### **3. SOP - Spill Response and Clean-up Inside Biosafety Cabinet**

#### **DO NOT TURN OFF THE BSC!**

Have a complete biological spill kit ready to go before you start the clean-up. Initiate clean-up as soon as possible. Allow cabinet to operate during clean-up.

#### **RESPONSE**

- REMAIN CALM!
- Alert people in the immediate area of spill.
- Remove contaminated clothing.
- Identify the agent’s specific hazards.

## **SPILL CLEAN-UP ROUTINE PROCEDURES**

- Use tongs/forceps to pick up sharp objects (broken glass, etc.) that may puncture gloves.
- Starting at the perimeter and working toward the center of the spill, cover the spill with paper towels or other absorbent material.
- Carefully pour disinfectant over the absorbent material and spill starting around the edges and working toward the center. Saturate the area with the disinfectant.
- Allow sufficient contact time typically 15-20 minutes and double the contact time for body fluids or viscous substances.
- Wipe up spill with a squeegee and/or paper towels, working from edge to center.
- Re-disinfect the spill area with fresh paper towels soaked in appropriate disinfectant. Wipe down all reachable cabinet surfaces with disinfectant.
- Place disposable contaminated materials into a biohazard bag and autoclave.
- Place contaminated reusable items in biohazard bags or heat resistant pans or containers with lids before autoclaving and further clean-up.
- Expose non-autoclavable materials to disinfectant, 20 minutes contact time, before removing them from the BSC.
- Remove protective clothing used during clean-up and place in biohazard bag for autoclaving.
- The cabinet should be run 15 minutes after clean-up before resuming work or turning off the cabinet.
- Inform all users of the BSC as well as the laboratory supervisor/principal investigator about the spill and successful clean-up as soon as possible.

**Notes:** Medical evaluation, surveillance, and treatment are provided as appropriate, and written records are maintained.

## **4. SOP - Processing Biohazardous Regulated Medical Waste (example)**

**AUTOCLAVE TRAINING:** [ASU Autoclave Training Video](#). Complete Biosafety training form signed by your supervisor and give to the Office of Laboratory and Field Safety.

### **SUPPLY LIST**

**Please return all used items the room!**

- Biohazard Waste Bin
- Autoclaved Waste Cart
- Auxiliary Biohazard Bin
- Large Nalgene Autoclavable Tubs (6)
- Small Nalgene Autoclavable Tubs (6)
- Supply Cart
- Supply Shelf
- Biohazard Bags/twist ties
- Waste Tags and Wires
- Autoclave Gloves
- Waterproof Gloves

- Nitrile Gloves (M-L)
- Disposable aprons
- Safety glasses
- Autoclave tape
- Sharpies
- Black Opaque Trash Bags
- Disinfecting wipes
- Disinfectant spray
- Bleach
- Paper towel
- Plunger
- Spatula
- Dust pan/Broom
- Squeegee
- Logbook and pen

### **WEAR APPROPRIATE PPE**

#### **For unsterilized waste:**

- Lab Coat/Apron
- Eye protection
- Nitrile gloves
- Shoes

#### **For Autoclaved waste:**

- Lab Coat/Apron + long sleeves
- Eye protection
- Heat resistant gloves

### **LABELING LABORATORY REGULATED MEDICAL WASTE**

Fill out the Biohazard waste tag with a Sharpie and secure to the bag with a twist tie or wire. The label should include:

- Your Name:
  - Lab room #:
  - Date:
  - Description of waste:
  - Cycle number:
- (Refer to the example provided)

### **PROCESSING BIOHAZARD WASTE**

- Place tagged and unsterilized waste in the Red Biohazard Waste bin to be sterilized or the auxiliary biohazard bin (small bin with no wheels) if the large wheeled bin is full.
- If the red bins are full please run a waste cycle if you have been trained on using the autoclave and processing the waste or please report full waste bins to your PI/supervisor.
- The autoclave cycle for waste will run for 45 minutes and tested periodically with biological indicators to validate removal of viable agents.
- Be sure the waste bag is contained in a Nalgene autoclave tub before placing it in the autoclave.

- If the majority of the waste material is “dry”, be sure to not seal the bag tightly or add ½ liter of water to the bag before sealing to assure steam penetration/generation.
- Tubs are to be placed on the large rolling autoclave rack. Roll the rack to the front of the autoclave and be sure the pins align with the slots inside the autoclave. The rack with the tubs can then be rolled into the autoclave.
- Close the door, start the waste cycle and fill out the logbook provided.
- After the cycle is complete and wearing heat protective PPE, roll out the autoclave rack, place a “STERILE & HOT” sign on the tubs and allow the bags to cool. Released agar will solidify in the Nalgene tub and will quickly grow microbes so be sure discard as soon as it has cooled.
- Write the cycle number on all the waste tags for the sterilized waste.
- After the waste cools, place the sterilized red bag inside a black opaque garbage bag. Any agar that has solidified should be scooped out of the tub using a dust pan or spatula and placed into the black trash bag with the waste bag. Place the sealed black bags in the rolling cube truck to go to the dumpster.
- Rinse the spatula, dust pans and autoclave tubs thoroughly and store so they will dry. Molten agar in the sink must be flushed by running hot water in the drain a minimum of 5 minutes so it does not solidify in the drains.
- Remember the processed materials often contain nutrients that grow bacteria and fungi. If they have water they will rapidly putrefy.
- CLEAN UP! Thank you.

# Appendix B – Laboratory Specific Biosafety Training

## 1. Autoclave Safety

Autoclave Training 20 Minute Video from [Arizona State University](#).

### Training Exam – Autoclave Safety True or False

1. Skin and eye protection are required due generation of high pressure and steam. T or F
2. Most plastic materials can be safely autoclaved. T or F
3. Glass with liquids should be tightly capped for sterilization. T or F
4. It is important to quickly remove liquids once the autoclave is opened. T or F
5. When sterilizing red bag waste is best to place the bag directly on the rack to assure sterilization. T or F
6. Waste bags should be loosely closed especially if the waste are mostly dry goods. T or F
7. Liquid cycles use slow exhaust cycles to slowly release pressure to keep liquids from boiling out. T or F
8. Heat gloves always protect your hands even when wet. T or F
9. You should keep your face away and behind the door when you open the door. T or F
10. All materials will be sterile after a 15-minute cycle at 121 degrees C. T or F

Please keep a copy of the quiz in the laboratory biosafety manual for a staff using the autoclaves. The principal investigator or OLFS should sign the form.

Employee Signature \_\_\_\_\_ Date of Training \_\_\_\_\_

PI Signature \_\_\_\_\_ Date \_\_\_\_\_

## 2. Biological Safety Cabinet (BSC)

### General information

- BSC used for bacteria, viruses, or potentially infectious substances.
- Primary barrier especially to reduce exposure to infectious aerosols and respiratory transmission of infectious agents.
- Required for BSL2 work/aerosol generating activities of RG2 agents or higher.
- BSC certified annually, when moved, or malfunction/repair occurs, ANSI/NSF STD. 49 or manufacturers specifications.
- BSC should not be placed near doors fume hoods, windows, ductwork. etc.
- Only one person should use the BSC at any given time.
- Avoid rapid movements in and out of BSC.

### Types of BSCs and Usage

- Class II A2 (Formerly called A/B3) air in from sash, HEPA, exhaust into room, limit chemicals test tube quantities.
- Class II B1 air in from sash, HEPA, 30% recirculated, 70% ducted, small volumes of chemicals.
- Class II B2 air from above unit/outside fully ducted, but **not** a chemical hood.
- UV light use requires eye/skin protection and a closed sash.
- Do **not** use open flames in BSC.

### Operation of BSC

1. Turn blower on unit and run 5-10 minutes prior to use to stabilize and purge.
2. Assure sash is in the proper position (check with alarm).
3. Be sure certification is current.
4. Check magnehelic meter with current certification + or – 10%.
5. Purge for 2 minutes after use.
6. Never use if in alarm mode.

### Procedures

1. Proper PPE with gloves, lab coat, eye protection.
2. Disinfect inside of hood with adequate contact time for agent being used.
3. Rinse with sterile water or 70% ethanol if bleach disinfectant is used.
4. Use good aseptic technique/avoid generation of aerosols.
5. Plan your work, disinfect materials before placing in BSC.
6. Avoid tall and bulky items, minimal amounts of materials in BSC.
7. Designate a clean area, work area, and contaminated area and work clean to dirty.
8. Slow and deliberate movements. Move arms straight in and out.
9. Do not block grates, work 4-6” back from grate in center of surface.
10. Bag and secure waste inside BSC before disposal outside of BSC.

11. Leave BSC blower on if a spill occurs, cover with absorbent, add disinfectant, 15-30 minutes contact, wipe area, discard outside gloves.
12. Disinfect items as removed.
13. Disinfect the cabinet, discard PPE.
14. Wash hands and forearms with antibacterial soap.

**Training Exam – Biological Safety Cabinet (BSC)** **True or False**

- |   |        |
|---|--------|
| 1. Use of pathogens in the BSC can begin as soon as the blower is turned on.      | T or F |
| 2. You should move slowly and deliberately while working in the BSC.              | T or F |
| 3. Aseptic technique is not needed in the BSC.                                    | T or F |
| 4. The interior of the BSC should be disinfected before and after use.            | T or F |
| 5. The operation of the BSC is improved if placed near a doorway.                 | T or F |
| 6. The BSC is a primary barrier for aerosolized potentially infectious materials. | T or F |
| 7. It is best to place commonly used supplies on the grate for easy access.       | T or F |
| 8. Using large volumes of flammables is okay in a Class II B2 BSC                 | T or F |
| 9. If you spill a pathogen in a BSC, immediately shut off the blower.             | T or F |
| 10. The BSC requires certification annually.                                      | T or F |

Employee Signature \_\_\_\_\_ Date of Training \_\_\_\_\_

PI Signature \_\_\_\_\_ Date \_\_\_\_\_



### 3. Laboratory Specific Training Form

Name of Laboratory Personnel: \_\_\_\_\_

Laboratory Location: Building \_\_\_\_\_ Room # \_\_\_\_\_

Principal Investigator: \_\_\_\_\_

Biohazardous Agent(s) or Other Potentially Infectious Material (OPIM):  
\_\_\_\_\_

Do you know the location of the Emergency Information/Equipment listed below?

Emergency Contact Numbers	Yes <input type="checkbox"/>	No <input type="checkbox"/>
Nearest Telephone	Yes <input type="checkbox"/>	No <input type="checkbox"/>
Fire Extinguisher	Yes <input type="checkbox"/>	No <input type="checkbox"/>
Eyewash Station/Shower	Yes <input type="checkbox"/>	No <input type="checkbox"/>
Disinfectant/Spill Kit/Dust pan for broken glass	Yes <input type="checkbox"/>	No <input type="checkbox"/>
Applicable Safety Data Sheets (SDS)	Yes <input type="checkbox"/>	No <input type="checkbox"/>

Have you completed lab specific safety training? Yes  No

Have you been trained in control of exposure to blood borne pathogens? Yes  No

Have you read the Standard Operating Procedures (SOP) for the hazardous materials?  
Yes  No

Have instructions been given on what to do in case of a spill or emergency? Yes  No

Are working alone policies in place for your lab? Yes  No

Do you have experience with aseptic technique and universal precautions? Yes  No

Do you know how to properly segregate the biohazardous & other types of lab waste?  
Yes  No

Are there sharps protocols in place? Yes  No

Is Personal Protective Equipment (PPE) required and available? Yes  No

Do you know the procedures to minimize the generation of aerosols? Yes  No

Does the work require use of a Biological Safety Cabinet (BSC)? Yes  No

Have you been trained on the safe use the BSC and other specialized equipment?  
Yes  No

Does the lab generate waste requiring autoclaving?  
Yes  No

Have you been trained on how to autoclave/disinfect waste materials for disposal?  
Yes  No

**Please sign and date this sheet and keep in a folder in your laboratory.**

Employee Signature \_\_\_\_\_ Date of Training \_\_\_\_\_

PI Signature \_\_\_\_\_ Date \_\_\_\_\_

# Appendix C – Risk Assessment

## PART A: Research/Investigation

Principal Investigator:

Department:

Location of Research:

Funding agency:

Agent Used:

Material Safety Data Sheet (MSDS) available:

Risk Group Level of Agent:

Biological Safety Level Used:

**Title & Brief Description of Research Activity:**

## PART B: Characterization of Agent

1. Is the agent a living microorganism?

Yes  No  NA

*If no, go to question #2*

Is the agent pathogenic based on the wild type strain?

Yes  No  NA

What is the host range of the agent?

Healthy humans,  Animals,  Immunocompromised humans,  Plants

Is the agent transmissible?

Yes  No  NA

If yes, what is the route of transmission? Airborne, ingestion, broken skin, mucous membranes, vectors, other \_\_\_\_\_

Is the agent attenuated?

Yes  No  NA

Does the attenuation reduce the risk?

Yes  No  NA

Lab strain? Yes  No  NA

Source \_\_\_\_\_

Is the organism well characterized? Yes  No  NA

Will the agent be genetically modified? Yes  No  NA

***If yes, answer question #2***

NIH Risk Group:  RG1,  RG2,  RG3,  RG4,  NA

Other/Comments:

**2. Are recombinant DNA constructs used or created?** Yes  No  NA

***If no go to question #3***

Is a viral vector being used? Yes  No  NA

***If yes, answer question #1***

What is the host range of the viral vector?

Healthy humans,  Immunocompromised humans,  Animals,  Bacteria (phage),  Plants

Is there a risk of the target cells becoming oncogenic? Yes  No  NA

Does the DNA code for production of a human toxin? Yes  No  NA

Where will the DNA construct be inserted?

Human,  Animal,  Plant,  Bacterium,  Tissue,  Cells,  Fungi/yeast,  
 Other \_\_\_\_\_

**3. Are human or non-human primate materials involved?** Yes  No  NA

***If no, answer question #4 in Part C on page #3***

Human blood cells or tissue? Yes  No  NA

Non-human primate (NHP) blood cells or tissue? Yes  No  NA

Other human bodily fluids?

Yes  No  NA

- Other NHP fluids?
- Human derived cell lines or tissue?
- NHP cell lines or tissue?
- Are any of the materials fixed or preserved?  
**If yes,** fixative used? \_\_\_\_\_

Yes  No  NA

Yes  No  NA

Yes  No  NA

Yes  No  NA

Other/Comments:

### **PART C: Characterization of Staff/Protocols**

4. Does the principal investigator have experience with this agent? Yes  No  NA

5. Do workers require special training to safely work with the agent? Yes  No  NA

6. Is the training documented? Yes  No  NA

7. Increased risk for exposure for certain workers or activities? Yes  No  NA

8. Are there risks to maintenance or custodial staff in the lab? Yes  No  NA

9. Are there procedures in place to minimize exposure? Yes  No  NA

10. Are there alternative activities that may reduce the risk? Yes  No  NA

11. Is there a vaccination available against the agent? Yes  No  NA

12. Is medical surveillance appropriate for monitoring exposure? Yes  No  NA

13. Does the research involve a large scale operation? (>10 Liters) Yes  No  NA

14. Are vertebrate animals used in the research? Yes  No  NA

***If no, skip to question #20 in PART D***

15. Are animals infected or exposed to the agent? Yes  No  NA

16. Is shedding of the agent possible? Yes  No  NA

17. Is the animal infectious to other animals or humans? Yes  No  NA

18. Will bites/scratches increase the risk of exposure to the agent? Yes  No  NA

19. Has the vertebrate animal protocol been approved by IACUC? Yes  No  NA

Other/Comments:

**PART D: Characterization of Facilities/Equipment**

20. Are there sharps protocols? (plastic, safe-sharps, disposal, etc.) Yes  No  NA
21. Are there proper waste disposal arrangements in place? Yes  No  NA
22. Is there an autoclave available for biohazardous waste? Yes  No  NA
23. Is the waste autoclaved correctly to assure sterility? Yes  No  NA
24. Is the biohazardous labeling of the sterile waste concealed before disposal in the dumpster? Yes  No  NA
25. Is the laboratory waste properly transported? Yes  No  NA
26. Is biohazardous waste properly segregated? Yes  No  NA
27. Is a Class II Biological Safety Cabinet (BSC) recommended? Yes  No  NA
28. Is an effective and appropriate disinfectant in use? Yes  No  NA
29. Is the disinfectant contact time sufficient? Yes  No  NA
30. What types of personal protective equipment are recommended?  
gloves  eye protection  lab coats/aprons   
face protection  respiratory protection  Other  \_\_\_\_\_
31. Are laundry and decontamination facilities or services available? Yes  No  NA
32. Is there a contingency plan in case of exposure/accident? Yes  No  NA
33. What Biosafety level is recommended for the work?  
Laboratory work BSL1 BSL2 BSL3 BSL4  
Animal Work: ABSL1 ABSL2 ABSL3 ABSL4

Other/Comments:

**PART E. Risk Assessment/Final Analysis/Approval**  
*(To be completed by biosafety coordinator or appointee)*

Date of risk assessment: \_\_\_\_\_

Risk assessment conducted by: \_\_\_\_\_

IBC approval required for research based on risk assessment?      Yes  No  NA

Submitted to IBC (date): \_\_\_\_\_

Reviewed by IBC on (date): \_\_\_\_\_

Corrective action (s) required for approval of research?:      Yes  No  NA

*If yes, describe below:*

Corrective actions completed?      Yes  No  NA  Date: \_\_\_\_\_

IBC approval granted:      Yes  No  NA  Date: \_\_\_\_\_

## **Appendix D – Administrative Information Form**

1. Key personnel's contact information
  - a. Principle Investigator (PI):
  - b. Co-Investigator:
  - c. Laboratory Manager/Technicians:
  - d. Date Manual Prepared/updated:
  - e. Containment (Biosafety) Level Assigned: **BSL2**
  
2. Biological Agent Information:
  - a. Biological / rDNA agent in use and biosafety level:
  
  - b. Susceptible population:
  
  - c. Route of infection:
  
  - d. Infective dose:
  
  - e. Clinical sign of infection:
  
  - f. Treatment:
  
  - g. Prophylaxis:
  
3. Agent Location and Storage: (specify location and containment equipment for each agent by room, freezer, incubator, hood, etc.)
  
  
4. Location of spill clean-up kit:
  
  
5. Lab Specific PPE required: (Include PPE requirements for lab access with no agent manipulation, agent manipulation, agent manipulation with potential aerosol production, animal handling, etc.)
  
  
  
  
  
  
6. Laboratory specific emergency notification, Call primary investigator, phone #:

## Appendix E – Injuries

On-the-job injuries of Central Michigan University Employees/Student employees, based on severity of the injury, are either immediate attention emergencies or non-emergencies. **Call 911 and seek medical attention immediately in the event of an emergency.**

All injuries that occur at work should be reported as soon as possible (preferably within 24 hours) to the Workers' Compensation office at 989-774-7177. [Here](#) is the CMU's injury reporting procedure.

Faculty, staff and student employees requiring medical treatment for work-related injuries/illnesses should seek treatment at one of the following CMU designated medical providers:

### **COMP - Occupational Medicine**

- 1523 S. Mission (next to Rally's)
- 989-773-2339
- Monday - Friday, 8:00 am – 4:00 pm
- Closed Saturday and Sunday

**Ready Care** - Use this facility when COMP is closed (*except for bloodborne pathogen exposures and emergencies*).

- 1523 S. Mission (next to Rally's)
- 989-773-1166
- Monday - Friday, 9:30 am – 5:30 pm
- Closed Saturday and Sunday

**Emergency Facility** - Use this facility for critical emergencies or when Ready Care is closed.

- McLaren - Central Michigan Fast Track or Emergency Department
- 1221 South Drive
- 989-772-6777
- 24/7
- Follow-up visits must be scheduled at COMP.

Note: If an injured employee/student employee chooses to see his/her own medical provider or seek treatment at facilities other than those listed above, the employee may be responsible for any incurred expense.



# Appendix F – Biohazard Incident Report Form

## BIOHAZARD INCIDENT REPORT FORM

**Instructions:** This form should be completed by lab supervisors, principal investigators (PIs), lab instructors, or the person involved in the incident. Complete the appropriate information below and provide the [Office of Laboratory and Field Safety](mailto:labfieldsafety@cmich.edu) (labfieldsafety@cmich.edu). Please forward this information as soon as possible and preferably within 24 hours of the incident.

### General Information

Your Name:  
Email:  
Phone number:  
Report Date:

### Incident Information

Name of person in incident:  
Role (employee, student, visitor, etc.):  
Lab Location (building and room #):  
Witness Names:

---

### Lab Information: (check all that apply)

- |                                  |  |   |
|----------------------------------|--|---|
| <input type="checkbox"/> BSL1    | <input type="checkbox"/> Research Lab            | <input type="checkbox"/> Controlled access        |
| <input type="checkbox"/> BSL2    | <input checked="" type="checkbox"/> Academic lab | <input type="checkbox"/> Hand washing sink        |
| <input type="checkbox"/> BSL3    | <input type="checkbox"/> Insectary               | <input type="checkbox"/> Biosafety cabinet in use |
| <input type="checkbox"/> Eyewash | <input type="checkbox"/> Animal room             | <input type="checkbox"/> Safety shower            |
| <input type="checkbox"/> SOPs    | <input type="checkbox"/> Training Documents      | <input type="checkbox"/> Sharps protocols         |
| <input type="checkbox"/> Signage | <input type="checkbox"/> Emergency contact info. | <input type="checkbox"/> Lab Manual               |

Other \_\_\_\_\_ (specify)

### Nature of Incident (check all that apply)

- |   |   |   |       |          |
|---|---|---|-------|----------|
| <input type="checkbox"/> Needle stick     | <input type="checkbox"/> Puncture wound                       | <input type="checkbox"/> Laceration     |       |          |
| <input type="checkbox"/> Aerosol exposure | <input type="checkbox"/> Animal bite                          | <input type="checkbox"/> Animal scratch |       |          |
| <input type="checkbox"/> Spill            | <input type="checkbox"/> Environmental release outside of lab |   |       |          |
| <input type="checkbox"/> Splash to:       | eyes  | nose                                    | mouth | (circle) |

Unknown exposure \_\_\_\_\_ (specify)

### Agents Involved (check all that apply)

- Human Derived Materials

- Bloodborne pathogens
- Recombinant DNA
- Viral Vectors
- Biological toxin
- Infectious or Pathogenic Agent

Other \_\_\_\_\_ (specify)

**Personal Protective Equipment Used**

- Nitrile Gloves       Lab coat       Shoe/Head covers
- Safety Glasses       Safety Goggles       Face shield

Respiratory Protection \_\_\_\_\_ (specify)

Other \_\_\_\_\_ (specify)

**Description of Incident: (Use separate page if needed)**

*(What happened, immediate response/first aid measures taken, medical treatment provided, cause of incident if known, etc. (include dates and times if possible))*

Signature:

Date:

*Biosafety Program Use Only:*

Date Received:

Reviewed by:

Follow-up:

Reported to:

Attached Documents:

## **Appendix G – Laboratory Specific Biosafety Manual**

The Laboratory Specific Biosafety Manual should be a living and working document that is an important resource for personnel engaged in the activities using biological and/or rDNA materials. Its primary focus should be to provide pertinent information to help execute the operations of the laboratory in a safe and professional manner.

This document along with other applicable information can serve as a Laboratory Specific Biosafety Manual which is required by the Centers for Disease Control (CDC) for working with biohazardous materials. Please include as appropriate:

### **1. Training**

- Laboratory Safety Training certificate (XI Training).
- Biosafety training certificate(s) (XI Training).
- BBP training certificate (XI Training).
- Autoclave Training Documents (X Autoclave, Appendix B).
- BSC Training Documents (Appendix B).
- Documents for completing trainings that supervisor/PI recommended, and/or Laboratory Specific Biosafety Manual required (XI Training).

### **2. Approval**

- Committee approvals, IBC, IRB, and/or IACUC.
- IBC Registration Document Title, Number(s), and Date Approved:
  - IBC Registration Document.
  - IACUC Protocol Number.
  - BSC Certification Documents.

### **3. Documents**

- Standard Operating Procedures including all applicable SOPs for decontamination, disposal, security, emergency procedures, handling procedures, shipping, and transport, etc.
- Occupational Health Information including targeted fact sheets that specify the agent and information on occupational risks that may be posed by the agent.
- Laboratory specific agent spill clean-up procedure.
- Risk Assessment for Specific Agent and Project. A comprehensive risk assessment is a critical element of the biosafety manual. The PI is responsible for identifying what the risks are for lab personnel or others working with the agent in the laboratory. Examples might be sharps hazards, aerosol hazards, animal bites or exposures, spills, splashes, etc.
- Laboratory specific sharps protocol.
- Laboratory specific aerosol reduction procedure.
- Laboratory agent-specific decontamination procedure.
- Agent security provisions/procedure.
- Laboratory specific emergency procedures. List procedures to be followed in case of an accident such as a spill, injection, ingestion, aerosolization, splash, etc.

#### 4. Regulations

- [Biosafety in Microbiological and Biomedical Laboratories \(BMBL\) 6<sup>th</sup> Edition.](#)
- [National Institute of Health Biosafety Guidelines.](#)
- [ASM Guidelines for Biosafety in Teaching Laboratories.](#)
- [OSHA Blood Borne Pathogen Standard.](#)
- [Pathogen Safety Data Sheets.](#)