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| BSL- \_\_ Lab Biosafety Manual  PI Name:  Laboratory (Building(s) and Room(s): |
| |  |  |  | | --- | --- | --- | | University of Wisconsin-Milwaukee |  | Date of Revision: | |

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# Contact List

**In an emergency, contact: 911**

|  |  |  |  |
| --- | --- | --- | --- |
| **Name** | **Role** | **Office Phone #** | **E-mail Address** |
|  | Principal Investigator |  |  |
|  | Backup Contact |  |  |
|  | Facilities Management |  |  |
|  | Department of Public Safety |  |  |
|  | Non-emergency |  |  |
| Jill McClary-Gutierrez | Biosafety Officer | (414)588-4261 | [mcclary@uwm.edu](mailto:mcclary@uwm.edu) |
| Kim Axtman | Radiation Safety Officer | (414) 430-7507 | [axtman@uwm.edu](mailto:axtman@uwm.edu) |
| Jennifer Herriges | Laboratory Safety Coordinator | (414) 430-7508 | [herrigej@uwm.edu](mailto:herrigej@uwm.edu) |
| James Fay | Occupational Health and Industrial Hygiene Manager | (414) 313-9711 | [jamesfay@uwm.edu](mailto:jamesfay@uwm.edu) |
|  |  |  |  |

# Training

All personnel who work in the laboratory must receive adequate instruction from their supervisor prior to beginning work. Some training is required annually. Each lab will require different trainings. The UWM Department University Safety and Assurances provides training for biosafety in the following: (customize for your group, add in lab-specific training requirements done internally as well)

* **Biosafety Training (BSL-1 and BSL-2) (face-to-face and online)**
* **Animal Biosafety Levels 1 and 2 Training (online)**
* **Bloodborne Pathogens training (online)**
* **NIH Guidelines for Research Involving Recombinant/Synthetic Nucleic Acid Molecules (online)**
* **Shipping Infectious Materials (online)**

Training on lab-specific techniques and demonstration of competency should also be required before work. Some work may require an occupational health plan, including annual physicals, pulmonary function test and fit test for use of a respirator, vaccinations, serum testing, and/or other elements of a medical plan. Contact the Biosafety Program at (414) 588-4261 for guidance.

Please use this space to identify the training requirements for your facility.

The PI is required to inform laboratorians and animal handlers of any risks to immunocompromised individuals and should encourage those individuals to consult with their health care provider about these risks.

## Biological Safety Research Training Log

An up-to-date training log can be found below.

|  |  |  |
| --- | --- | --- |
| Name | Course | Training Completion Date |
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|  |  |  |

# Research Laboratory Description

Insert brief description of research conducted in this facility.

# List of Approved Biosafety Protocols

Include expiration dates, and any modification requests. Attach approved protocols in Appendix A.

# Biological Agents/ Organisms Used In This Facility

List all biological agents used in your facility here and clearly identify biohazards associated with them and their risk group.

Attach your completed and signed inventory in Appendix B.

(include bacterial, protozoal, viral, fungal, recombinant, human/non-human primate unfixed tissues or cells, prions, viroids, and any other parasites in your possession)

# BMBL BSL-2 Laboratory Criteria

*Biosafety in Microbiological and Biomedical Laboratories (BMBL)* 5th Edition, February 2009

Centers for Disease Control and Prevention and National Institutes of Health

**Biosafety Level 2** builds upon BSL-1. BSL-2 is suitable for work involving agents that pose moderate hazards to personnel and the environment. It differs from BSL-1 in that:

* laboratory personnel have specific training in handling pathogenic agents and are supervised by scientists competent in handling infectious agents and associated procedures;
* access to the laboratory is restricted when work is being conducted; and
* all procedures in which infectious aerosols or splashes may be created are conducted in BSCs or other physical containment equipment.

The following standard and special practices, safety equipment, and facility requirements apply to BSL-2:

**Standard Microbiological Practices**

1. The laboratory supervisor must enforce the institutional policies that control access to the laboratory.

2. Persons must wash their hands after working with potentially hazardous materials and before leaving the laboratory.

3. Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption must not be permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.

4. Mouth pipetting is prohibited; mechanical pipetting devices must be used.

5. Policies for the safe handling of sharps, such as needles, scalpels, pipettes, and broken glassware must be developed and implemented. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries.

Precautions, including those listed below, must always be taken with sharp items. These include:

1. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
2. Used disposable needles and syringes must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal.
3. Non-disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
4. Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs, or forceps. Plastic ware should be substituted for glassware whenever possible.

6. Perform all procedures to minimize the creation of splashes and/or aerosols.

7. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.

8. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method. Depending on where the decontamination will be performed, the following methods should be used prior to transport:

1. Materials to be decontaminated outside of the immediate laboratory must be placed in a durable, leak proof container and secured for transport.
2. Materials to be removed from the facility for decontamination must be packed in accordance with applicable local, state, and federal regulations.

9. A sign incorporating the universal biohazard symbol must be posted at the entrance to the laboratory when infectious agents are present. Posted information must include: the laboratory’s biosafety level, the supervisor’s name (or other responsible personnel), telephone number, and required procedures for entering and exiting the laboratory. Agent information should be posted in accordance with the institutional policy.

10. An effective integrated pest management program is required.

11. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures. Personnel must receive annual updates or additional training when procedural or policy changes occur. Personal health status may impact an individual’s susceptibility to infection, ability to receive immunizations or prophylactic interventions. Therefore, all laboratory personnel and particularly women of child-bearing age should be provided with information regarding immune competence and conditions that may predispose them to infection. Individuals having these conditions should be encouraged to self-identify to the institution’s healthcare provider for appropriate counseling and guidance.

**Special Practices**

1. All persons entering the laboratory must be advised of the potential hazards and meet specific entry/exit requirements.

2. Laboratory personnel must be provided medical surveillance and offered appropriate immunizations for agents handled or potentially present in the laboratory.

3. Each institution must establish policies and procedures describing the collection and storage of serum samples from at-risk personnel.

4. A laboratory-specific biosafety manual must be prepared and adopted as policy. The biosafety manual must be available and accessible.

5. The laboratory supervisor must ensure that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-2 agents.

6. Potentially infectious materials must be placed in a durable, leak proof container during collection, handling, processing, storage, or transport within a facility.

7. Laboratory equipment should be routinely decontaminated, as well as, after spills, splashes, or other potential contamination.

1. Spills involving infectious materials must be contained, decontaminated, and cleaned up by staff properly trained and equipped to work with infectious material.
2. Equipment must be decontaminated before repair, maintenance, or removal from the laboratory.

8. Incidents that may result in exposure to infectious materials must be immediately evaluated and treated according to procedures described in the laboratory biosafety safety manual. All such incidents must be reported to the laboratory supervisor. Medical evaluation, surveillance, and treatment should be provided and appropriate records maintained.

9. Animals and plants not associated with the work being performed must not be permitted in the laboratory.

10. All procedures involving the manipulation of infectious materials that may generate an aerosol should be conducted within a BSC or other physical containment devices.

**Safety Equipment (Primary Barriers and Personal Protective Equipment)**

1. Properly maintained BSCs (preferably Class II), other appropriate personal protective equipment, or other physical containment devices must be used whenever:

1. Procedures with a potential for creating infectious aerosols or splashes are conducted. These may include pipetting, centrifuging, grinding, blending, shaking, mixing, sonicating, opening containers of infectious materials, inoculating animals intranasally, and harvesting infected tissues from animals or eggs.
2. High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory using sealed rotor heads or centrifuge safety cups.

2. Protective laboratory coats, gowns, smocks, or uniforms designated for laboratory use must be worn while working with hazardous materials. Remove protective clothing before leaving for non-laboratory areas (e.g., cafeteria, library, administrative offices). Dispose of protective clothing appropriately, or deposit it for laundering by the institution. It is recommended that laboratory clothing not be taken home.

3. Eye and face protection (goggles, mask, face shield or other splatter guard) is used for anticipated splashes or sprays of infectious or other hazardous materials when the microorganisms must be handled outside the BSC or containment device. Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse. Persons who wear contact lenses in laboratories should also wear eye protection.

4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to latex gloves should be available. Gloves must not be worn outside the laboratory. In addition, BSL-2 laboratory workers should:

1. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary. Wear two pairs of gloves when appropriate.
2. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
3. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.

5. Eye, face and respiratory protection should be used in rooms containing infected animals as determined by the risk assessment.

**Laboratory Facilities (Secondary Barriers)**

1. Laboratory doors should be self-closing and have locks in accordance with the institutional policies.

2. Laboratories must have a sink for hand washing. The sink may be manually, hands-free, or automatically operated. It should be located near the exit door.

3. The laboratory should be designed so that it can be easily cleaned and decontaminated. Carpets and rugs in laboratories are not permitted.

4. Laboratory furniture must be capable of supporting anticipated loads and uses. Spaces between benches, cabinets, and equipment should be accessible for cleaning.

1. Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
2. Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.

5. Laboratory windows that open to the exterior are not recommended. However, if a laboratory does have windows that open to the exterior, they must be fitted with screens.

6. BSCs must be installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSCs should be located away from doors, windows that can be opened, heavily traveled laboratory areas, and other possible airflow disruptions.

7. Vacuum lines should be protected with High Efficiency Particulate Air (HEPA) filters, or their equivalent. Filters must be replaced as needed. Liquid disinfectant traps may be required.

8. An eyewash station must be readily available.

9. There are no specific requirements on ventilation systems. However, planning of new facilities should consider mechanical ventilation systems that provide an inward flow of air without recirculation to spaces outside of the laboratory.

10. HEPA filtered exhaust air from a Class II BSC can be safely re-circulated back into the laboratory environment if the cabinet is tested and certified at least annually and operated according to manufacturer’s recommendations. BSCs can also be connected to the laboratory exhaust system by either a thimble (canopy) connection or a direct (hard) connection. Provisions to assure proper safety cabinet performance and air system operation must be verified.

11. A method for decontaminating all laboratory wastes should be available in the facility (e.g., autoclave, chemical disinfection, incineration, or other validated decontamination method).

# Laboratory Signage

The sign found in Appendix C is a required sign for all BSL-2 facilities. This door sign outside of the laboratory is posted by the PI and will be checked bi-annually by the Biological Safety Officer, who will do walk-throughs of buildings to make sure BSL-2 facilities are complying with this requirement. Biosafety level 2 labs should have the following permanently-affixed decals: Biohazard symbol, “BSL-2” designation, Entry/ Exit Requirements, and emergency contacts. The specific agents worked with in the lab should **not** be listed, for biosecurity reasons. If any changes need to be made to the sign, including emergency contacts, please log in and update the information online. The UWM Biosafety Program will post any changes to the sign and let personnel know.

# Standard Operating Procedures

## Biological Safety Cabinet

The biosafety cabinet (BSC) is the primary means of protecting the researcher, the product, and the environment from biological hazards. All work with infectious agents should be manipulated in the BSC, especially those practices which could generate aerosols. Using the BSC properly includes the following:

* 1. Turn on cabinet fan 15 minutes before beginning work
  2. Disinfect the cabinet work surface with 70% ethanol or other disinfectant and wipe surfaces.
  3. Place supplies in the cabinet. Locate container inside the cabinet for disposal of pipettes. (Movement of hands in and out of the cabinet to discard pipettes into a container located outside of the cabinet creates turbulence and disrupts the air barrier that maintains sterility inside the cabinet.)
  4. Work as far to the back (beyond the air split) of the BSC work space as possible.
  5. Always use mechanical pipetting aids.
  6. Avoid using open flames inside BSCs. If a flame is necessary, use a burner with a pilot light and place it to the rear of the workspace. Flames disrupt the airflow and contribute to the heat load inside the BSC. Flames have burned holes through HEPA filters and have caused explosions in BSCs.
  7. Do not work in a BSC while a warning light or alarm is signaling.
  8. Keep the work area of the BSC free of unnecessary equipment or supplies. Clutter inside the BSC may affect proper airflow and the level of protection provided.
  9. Keep the front and rear grilles clear.
  10. When work is completed, remove equipment and supplies from the cabinet. Wipe the work area with 70% ethanol and allow cabinet to run for 15 minutes.
  11. Some BSCs are equipped with ultraviolet (UV) lights. If one is used, the tube should be wiped with 70% ethanol every two weeks, while turned off, to remove dust. UV radiation should not take the place of 70% ethanol for disinfection of the cabinet interior. The UV lamp is not an acceptable replacement for routine disinfection.
  12. The UV lamp should never be on while an operator is working in the cabinet.
  13. Minimize traffic around the biosafety cabinet and avoid drafts from doors and air conditioning.
  14. The BSC is certified annually by a contractor coordinated by your research lab.
  15. When using the house vacuum system, place a hydrophobic filter (C) between overflow flask (B) and vacuum port (D). Examples include Whatman Vacu-guard and Pall Gelman Vacushield in-line disk filters. Turn off the vacuum when not in use.

The biological safety cabinet in our facility was last certified on: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_.

The company that certified our cabinet was: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_.

## 

## Sharps Disposal

All researchers working with human cells/ tissues, human blood, or human bodily fluids are required to complete OSHA Bloodborne Pathogens Training annually and have a Bloodborne Pathogens Exposure Control Plan on file in their research facility that is available to the lab inspectors upon request. These are required with IBC registration/ approval. Include this in Appendix D: Exposure Control Plans

To prevent needle stick injuries:

* Avoid using needles whenever possible.
* Replace glass materials with plastic (such as Pasteur pipettes)
* Do not bend, break, or otherwise manipulate needles by hand.
* Do not recap needles by hand. Do not remove needles from syringes by hand.
* Immediately after use, discard needle and syringe (whether contaminated or not) into puncture resistant sharps containers. RECAPPING OF NEEDLES IS PROHIBITED.
* Never discard sharps into regular trash.
* Never discard sharps into bags of biological waste.
* Use care and caution when cleaning up after procedures that require the use of syringes and needles.
* Do not overfill sharps containers. Close completely when 3/4 full, request pickup from the DES webpage at www.des.umd.edu.
* Locate sharps containers in areas in which needles are commonly used. Make containers easily accessible.

In the event of a needle stick injury:

Wash the area thoroughly with soap and water. Notify your PI or supervisor immediately. Students may report to the Norris Health Center, faculty/ staff must report to their hospital of choice. Please work with the Department of University Safety and Assurances to complete the appropriate forms within 24 hours of injury.

Although recapping needles is not recommended in the lab, there are times in which it must be done. In the event that needles must be filled in advance of their use, there are safe methods that can be used to "recap" them using one hand.  Here are several suggestions for doing this in a safe manner:

1. “One-handed scoop” method:

Place the cap on the benchtop and hold the syringe in one hand. Keep the other hand by your side. Slide the needle into the cap, then lift it up and snap it on securely using only one hand.





1. Using a sterile 50 mL centrifuge tube or Styrofoam rack:

Place the uncapped needle inside a conical tube temporarily instead of recapping. Alternatively, put the cap inside an open centrifuge tube or rack so that the needle can be inserted into it and the cap and secured by firmly pushing the needle downward into it. There are also commercial [needle recapping devices](http://www.isips.org/safety_needle_recapping_devices.php) available for this purpose.





Remember to keep a designated sharps container nearby for disposal of sharps, and don’t recap unless absolutely necessary!

## Autoclave Use

Include the specific instructions for use of your autoclave, including PPE and recordkeeping.

# Waste Decontamination and Disposal

All personnel are responsible for maintaining a clean work area. Only trained individuals should operate the autoclave.

1. Solid infectious materials

Insert procedures here.

1. Liquid infectious waste

Insert procedures here.

1. Uncontaminated waste

Uncontaminated non-sharp waste should be disposed of in the general lab waste stream.

Uncontaminated broken glass is disposed of in a sturdy cardboard box, preferably lined with a plastic bag. When full, the box should be taped closed and disposed of in the dumpster. Housekeeping will not dispose of broken glass.

1. Sharps disposal

Sharps are items which pose a puncture or cutting hazard, such as glass, needles, and razors. Sharps should be disposed of in approved autoclave-resistant puncture-proof containers. Please refer to section IV of this manual for more information.

1. Animal Carcasses

Place animal carcasses/tissues into a plastic bag. Double bag all carcasses when zoonotic agents are present. Store bag in freezer until removal. Disposal of animal carcasses/tissues is coordinated through the Animal Resource Center and Waste Management. Non-transgenic carcasses will go to MERI for radiation. Transgenic carcasses will go to Veolia for incineration.

1. Disposal of waste into dumpsters

Lab staff is responsible for transporting autoclaved waste to the dumpsters in a timely manner. Waste bags should not be left sitting in the laboratory or autoclave room for more than a few hours. If the dumpster is full, trash bags may NOT be discarded outside the dumpster. Bags must be returned to the lab and disposed of when the dumpster has been emptied. Although environmental services does a sweep of the labs, it is the lab personnel’s responsibility for ensuring their lab is tidy and organized.

# Spill Response Protocol

*The following sample protocols are provided to facilitate emergency planning and should be modified to include lab-specific, potentially biohazardous materials. These instructions should be displayed in the laboratory and periodically reviewed with personnel. In the event of an emergency do not hesitate to call 911 if necessary. University Safety and Assurances Biological Safety Program is available for additional assistance and information at 414-588-4261.*

## Spills INSIDE Containment

|  |  |
| --- | --- |
| **INITIAL RESPONSE** | * Immediately stop all work, but leave BSC or hood blower fan on during clean-up**.** |
| **CLEAN UP RESPONSE** | * Wear PPE (gloves, lab coat, eye protection). * Cover spill with disinfectant soaked paper towels and pour an appropriate disinfectant solution around spill. * Using paper towels and appropriate disinfectant detergent, wipe down walls, work surfaces and equipment. * Flood work surface and drain pan (Type II BSC) with disinfectant and let stand for at least 20 minutes. * Wipe up all excess disinfectant and empty drain pain into a collection vessel with disinfectant. * Flush drain pan with water and remove drain tube. * Transfer all contaminated materials into an autoclave bag. * Wipe down autoclave bag, disinfectant container and other contact surfaces with disinfectant. |
| **WRAP UP** | * Remove and discard PPE (autoclave prior to disposal). * Wash hands with soap/antimicrobial agent and water. * Autoclave all contaminated materials. * Report incident to the PI (if not already contacted). |

Use the guidelines below for response to spills of biological materials outside of the biosafety cabinet.

Your laboratory should be equipped with a spill kit containing necessary materials for cleaning up a spill. Know where it is stored so that you can retrieve it quickly. Maintain the spill kit. Replace spill kit components as they are used to prepare for the next incident.

## Spills OUTSIDE Containment

|  |  |
| --- | --- |
| **IMMEDIATE RESPONSE** | **EVACUATE** if necessary   * Alert co-workers and leave lab area immediately * Determine if medical attention is needed (injury, direct or potential exposure). In an emergency please call the Police Department at **9911** if on a campus phone or **(414) 229-9911** if using cell or off campus phone. * Close door and post lab with **Do Not Enter** sign. * Remove and put contaminated garments into a container for autoclaving. * Wash hands/face with soap/antimicrobial agent. |
| **CLEAN UP** | * Wait at least 30 minutes before re-entry to allow aerosols to dissipate. * Wear PPE upon re-entry (disposable gown, mask/eye protection, double gloves). * Cover spill with disinfectant soaked paper towels. * Pour an appropriate disinfectant solution around spill (1:10 dilution of bleach). Take care not to create aerosols while pouring. Let stand for at least 20 minutes. * Use tools (tongs, broom & dustpan) rather than hands to pick up sharps and broken glass, and contaminated materials whenever possible. * Transfer all contaminated materials into an autoclave bag. * Wipe down entire area with disinfectant (including autoclave bags, disinfectant container and other contact surfaces). |
| **WRAP UP** | * Remove and discard PPE (autoclave prior to disposal). * Shower or wash hands with soap/antimicrobial agent and water. * Autoclave all contaminated materials. * Report incident to the PI (if not already contacted) and the BSO (Biological Safety Officer) using the online [First Report of Biological Exposure or Release](http://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/)form (on UWM Biosafety site) within 24 hours. |

*Biohazardous spills involve: pathogenic, infectious and recombinant materials; biological toxins; human blood, tissues and other potentially infectious material (PIM) such as semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, pericardial fluid, peritoneal fluid, amniotic fluid, saliva in dental procedures and any bodily fluid that is visibly contaminated with blood.*

**Report all spills at:** [**https://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/**](https://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/)

# EMERGENCY PROCEDURES

1. Fire evacuation procedures

During a fire emergency, lab staff should prioritize life safety. Cultures and animals may be put away if time allows; if not, walk to the nearest exit. Pull the fire alarm if necessary, and call 911 once outside the building.

1. Power outage

In the event of a power outage, put away cultures and animals. Remove PPE and exit the lab normally. Emergency lighting within the buildings should provide adequate visibility to exit the building. Notify the PI immediately.

1. Medical emergency

In the event of a medical emergency in the lab, follow appropriate procedures depending on the hazards present. If the emergency involves a spill of hazardous agent onto the clothing or body, assist the victim to the shower or eyewash station. If the victim requires medical attention, call 911.

1. Accidental exposure or needlestick
   1. **Care for Personnel**
      1. If there has been a needlestick/puncture, wash the affected area thoroughly with soap and water for 15 minutes.
      2. For splashes to the eyes/mucous membrane, rinse the affected area under the eyewash for 15 minutes.
      3. **[Students]** If medical attention is needed, contact the Norris Health Center (during business hours) or an emergency medical care center (after hours, <https://uwm.edu/norris/health-services/emergencies/>).
      4. **[Employees]** If medical attention is needed, contact your primary care physician or an emergency medical care center.
   2. If a spill has occurred, contain the spill and initiate cleanup (see spill cleanup procedures below).
   3. **Notify** your PI, manager, or supervisor.
   4. **Notify** the Biological Safety Officer at [mcclary@uwm.edu](mailto:mcclary@uwm.edu) or 414-588-4261 to initiate incident reporting.

For all exposure/ spill incidents involving biological materials, report the incident online to the biological safety program by completing the form at: <http://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/>

# Biological Agent/ Toxin Exposure Control Plans

All risk group 2 agents are required to have an exposure control plan, which is to be submitted as part of their IBC registration and is a requirement for approval for work with risk group 2 agents. Each agent used must have its own fact sheet. The following is the template to be used, and may be copied and pasted to make additional fact sheets.

## Exposure Control Plan for: *XXXX*

Agent Risk Group: Risk Group 2

Containment Level: BSL 2

### Characteristics

Describe the pathogen here- i.e. bacteria (type), virus, fungi, etc., structural elements, virulence factors

### Incubation Period

Identify the length of time after infection before the prodromal phase of infection will appear

### Hazards

[Include general description of hazards posed by infectious agent, which may include characteristics of the agent, diseases or symptoms it may cause, and the major risk factors for infection within the laboratory. Much of this information may be pulled from a Material Safety Data Sheet(MSDS) or from the pathogen safety data sheets.

### Modes of Transmission

[Include likely modes of transmission in the laboratory, such as direct skin, eye, or mucosal membrane exposure, parenteral inoculation by needle or other contaminated sharp, ingestion of liquid suspension or contaminated hand to mouth exposure, or inhalation of aerosols. This information may also be found in the MSDS or PSDS.

### Signs/ Symptoms of Disease

Include the description of all signs/ symptoms associated with infection by this pathogenic agent.

### Engineering Controls

[Describe engineering controls used to reduce exposure in the lab, some examples are listed below- you need to include safety precautions taken during injection and also during transport of loaded syringes.]

* **A Certified Biosafety Cabinet** must be used for **all manipulations** of the agent (i.e., pipetting, harvesting, infecting cells, filling tubes/containers, opening sealed centrifuge tubes/rotors, shaking, mixing, etc.) and for handling infected cells.
* **Safety Engineered Sharps, such as those with retracting needles,** shall be used for injections. In addition, the use of other sharps (i.e., glass Pasteur pipettes) must be **eliminated** wherever possible.
* For animal injections, the animal must be **restrained or anesthetized.**
* **Biohazard Sharps Containers** shall be available to dispose of sharps waste, including broken glass, needles, blades, etc.
* When centrifuging, use aerosol containment devices such as **safety cups** that fit in the centrifuge bucket, covers for the centrifuge bucket, heat **sealed tubes**, or **sealed centrifuge rotors**. Rotors should be removed and opened inside a BSC. Centrifuge tubes should be filled and opened in BSC.
* **An in-line HEPA filter** must be used for vacuum aspiration of spent media.

### Administrative/ Work Practice Controls

[Describe various work practices used to control exposures, some examples are listed below]

* Access to the lab shall be restricted while work is in progress, doors shall remain closed during experimentation
* A sign incorporating the universal biohazard symbol shall be posted at the entrance of the laboratory or tissue culture room where agent is used (see last page)
* All lab personnel must be informed of the hazards of agent
* All lab personnel must be trained in proper handling, use, and disposal prior to working agent
* All lab personnel are advised to avoid rubbing eyes as a precautionary measure against eye infections
* All lab personnel will remove lab coat, discard gloves, and wash hands before exiting the lab

### Personal Protective Equipment (PPE)

[Describe personal protective equipment required to be worn when working with infectious agent, some examples are listed below]

Lab coat shall be worn while working in the lab

Safety glasses or goggles shall be worn when handling agent

Disposable gloves shall be worn while working in the lab

Respirators are required for aerosol-producing procedures performed outside of a biosafety cabinet. Contact the US&A office at X6339 for fit-testing prior to use of respirators.

### Disinfection

Describe the methods of disinfecting the agent.

### Disposal

[Describe how the agent will be disposed]

### Accidental Spill

[Describe spill procedures; standard methods are listed below]

In case of spill inside of biosafety cabinet:

* Lower sash and let biosafety cabinet continue to run (at least 5 minutes) in order to contain aerosols
* Immediately notify others around you
* Contaminated personal protective equipment(PPE), such as gloves, labcoat, and safety glasses, should be removed and disposed of as biohazardous waste or set aside for disinfection

For exposures/contamination, see “Personnel Contamination/Exposure Response” guidelines below

* Don appropriate PPE if not already wearing
* Use forceps to remove any broken glass or other sharp items; sharps should be placed into biohazard sharps containers
* Cover the spill with paper towels or other absorbent materials
* Apply 10% bleach directly around and onto the paper towels covering the spill
* Allow 15 minute contact time before cleaning, starting at the perimeter and working inwards towards the center
* Dispose of materials into biohazard bins
* Disinfect all surfaces of the biosafety cabinet with freshly prepared 10% bleach with a 15 minute contact time, followed by a wipedown with 70% ethanol to reduce corrosion
* Allow biosafety cabinet to run for at least 10 minutes before resuming work or turning off
* For large spills, you may contact the the Biosafety Office at 414-588-4261 for additional assistance.

In case of spill in lab (outside of biosafety cabinet):

* Immediately notify others around you
* Contaminated personal protective equipment(PPE), such as gloves, labcoat, and safety glasses, should be removed and disposed of as biohazardous waste or set aside for disinfection
* For exposures/contamination, see “Personnel Contamination/Exposure Response” guidelines below
* Leave the room and restrict access for 30 minutes to allow aerosols to settle
* Enter room wearing appropriate PPE
* Use forceps to remove any broken glass or other sharp items; sharps should be placed into biohazard sharps containers
* Cover the spill with paper towels or other absorbent materials
* Apply 10% bleach directly around and onto the paper towels covering the spill
* Allow 15 minute contact time before cleaning, starting at the perimeter and working inwards towards the center
* Dispose of materials into biohazard bins
* For large spills, you may contact the the Biosafety Office at 414-588-4261 for additional assistance.

### Exposure Response

[Describe the steps to take in the event of an exposure; standard procedures are listed below]

In the event of an exposure, take the following precautions:

* Remove any contaminated clothing
* Wash all affected areas; for eye exposures, rinse for 15 minutes in eyewash or flush area with water, for needle-stick or other sharps exposure, wash wound area with soap and water for 15 minutes
* Report the exposure to your supervisor immediately

### Special Practices for Animal Injections

[Describe special practices for handling animals exposed to infectious agent; standard procedures are described below]

* **Facility:** When animals are infected with agent, the Animal Biosafety Level of the project will generally be assigned to ABSL-2. This requires Biosafety Level-2 practice and facilities for procedures involving agent.
* **Signage:** Attached door sign must be posted on the door leading into the housing or procedure room. Cages must be labeled with the biohazard cage card label with agent identification and injection date upon injection of agent. Signage/labels must remain in place for a minimum of 1 week after the date of injection/exposure.
* **Animal Excretion:** Infected animals my excrete agent. Precaution must be taken not to create aerosols when emptying animal waste material and when washing down cages, or cleaning the room with pressure hoses. Surfaces that may be contaminated will be decontaminated ASAP with bleach solution. This practice must be followed for one full week after infection.
* **Cage Change:** Use a certified Class II biosafety cabinet when moving animal from dirty to clean cages. If multiple cages are being changed in the same biosafety cabinet, cages with animals infected with the agent should be changed last. Spray the inside surfaces of each cage with bleach solution and leave the cages in the biosafety cabinet for 15 minutes before returning them to the cage washing facility. Decontaminate the biosafety cabinet immediate after removing the contaminated cages with bleach solution.
* **Bedding:** A biosafety cabinet or negative airflow cage changing station should be used when disposing bedding into biohazard red bag. The red biohazard bag must be sealed and placed into the biohazard container.
* **Animal Carcass:** Infected carcasses should be placed in red biohazard bag and stored in designated for biohazardous carcass disposal.

**[General Door sign to be placed on animal room where infected animals are housed; fill in applicable information and coordinate with ARC Manager and Campus Veterinarian]**

### Post-Exposure Treatment

Identify the treatment plan for post-exposure here. Include reporting online.

#### If symptoms appear with no known incidence of exposure…

Seek medical attention and inform the health care provider of the microorganisms used in the workplace.

**Reporting of any exposure events**

Make note of the date and time of the incident and any relevant details. Inform principal investigator, fill out the appropriate paperwork, and contact the Biological Safety Program at: [First Report of Biological Exposure or Release Event](http://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/). If recombinant, the incident must be reported to NIH Office of Biotechnology Activities, which can be coordinated with the biological safety officer through using the [First Report of Biological Exposure or Release Event](http://uwm.edu/safety-health/first-report-of-biological-exposure-or-release-event/) page.

# Working with Animals

Animals must be housed, handled, and used in accordance with the federal Animal Welfare Act (P.L. 89-544, *et seq*) and the NIH *Guide for the Care and Use of Laboratory Animals*. All research involving animals must be done under a protocol approved by the University of Wisconsin-Milwaukee ACUC. The Associate Director of the Animal Care Program and the ARC Manager are responsible for assuring the safety and wellbeing of the research animals. Please insert all specific practices conducted for cleaning, PPE, training, etc. that have been coordinated with the animal care program.

Safety procedures for working with animals at ABSL2 containment

• Access to the animal facilities is restricted to personnel who have been advised of the potential hazard & who have a need to enter the room for program or service purposes.

• Staff will be advised of increased risks for persons who are immunocompromised, pregnant, or for whom infection might be unusually hazardous

• Personnel must wash their hands after handling cultures &/or animals, and before leaving the animal facility.

• Eating, drinking, handling contact lenses & applying cosmetics are not permitted in the animal rooms.

1. • Storing of food for human use is not permitted in animal rooms.
2. • Doors to animal rooms within the buildings are kept closed when animals are present. The building access doors are kept locked.
3. • Work surfaces are decontaminated after use or a spill of a viable material.
4. • An insect & rodent control program is in effect.
5. • Bedding & waste materials from animal cages are removed in such a manner as to minimize the creation of aerosols & disposed of by autoclaving.
6. • Cages are washed & decontaminated after use.
7. • All waste & animal carcasses from the animal rooms are double-bagged before removal from the building for incineration.
8. • Sharps shall be handled properly according to the relevant section of this manual.
9. • Broken glassware is not to be handled by hand, but should be removed by mechanical means such as a broom & dustpan, tongs or forceps.
10. • Spills, which result in exposure to infectious materials, should be reported to the immediate supervisor.
11. • All personnel entering animal rooms shall wear appropriate protective equipment.

# References for More Biosafety Information

*University Biosafety Manual:*  <http://uwm.edu/safety-health/biosafety-manual/>

*Biosafety in Microbiological and Biomedical Laboratories*, 5th edition

<http://www.cdc.gov/biosafety/publications/bmbl5/index.htm>

*Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines)*

<http://oba.od.nih.gov/rdna/nih_guidelines_oba.html>

# Biosafety Manual Review

The Biosafety Manual should be read and reviewed by each laboratory member before initiating work and re-reviewed annually thereafter. By signing below you are stating that you have read this document in its entirety and feel confident about the information that is presented.

|  |  |  |
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| **Name** | **Signature** | **Date** |
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# Appendix A: Approved/ Registered IBC Protocol(s) and any supplemental protocols (IACUC, IRB)

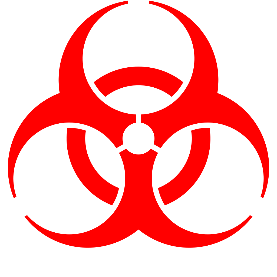
Include all approved IBC protocols (and IACUC if necessary).

# Appendix B: Biological Inventory

Attach inventory here.

# Appendix C: BSL-2 facility signage

Instructions: Complete the sign below, print, and cut paper down to size. Attach to all doors that allow for entry to the facility. BSL-2 facilities should keep their doors closed at all times.

**BIOSAFETY LEVEL 2**

**RESTRICTED ACCESS**

|  |  |
| --- | --- |
| Procedure Required for Entry/ Exit: |  |
| Special Practices (immunizations, etc.) |  |

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Notice** | **Call or See** | **Building** | **Room** | **Phone** | **E-mail** |
| PI |  |  |  |  |  |
| Emergency/ Co-PI |  |  |  |  |  |
| Biological Safety Officer | **Jill McClary-Gutierrez** | **Engelmann Hall** | **270** | **(414) 588-4261** | [mcclary@uwm.edu](mailto:mcclary@uwm.edu) |

|  |  |  |
| --- | --- | --- |
| **Building** | **Room** | **Date Posted:** |

# Appendix D: BBP Plans and SOPs