**Directions for Completing the Biosafety Manual (BSM) Template**

**NOTE:** The Principle Investigator (PI) is responsible for providing a Biosafety Manual and written risk assessment for all research utilizing potentially biohazardous materials performed at the BSL-2 level. In order to assist WSU investigators in these endeavors this Biosafety Manual Template has been developed.

**BACKGROUND:** The information provided in this template has been obtained from the following sources: “Biosafety in Microbiological and Biomedical Laboratories” ([BMBL](https://www.cdc.gov/labs/BMBL.html) 6th edition) CDC/NIH and “The NIH Guideline for Research Involving Recombinant DNA Molecules” ([NIH Guidelines](https://osp.od.nih.gov/wp-content/uploads/NIH_Guidelines.pdf)) As well as WSU’s Safety Policies and Procedures Manual (SPPM) and other pertinent regulations and guidelines.

1. Fill in all sections of the manual in blue print**.**
2. Please utilize the [Potentially Biohazardous Guideline](https://biosafety.wsu.edu/potentially-biohazardous-materials/) to verify that you have included all appropriate agents in Section one page 1. “Potentially biohazardous materials”. Underlined text in the body of this document indicates a link that when engaged will take you to more information on this topic. (In some cases you may need to select the “control” key on the keyboard and click with the mouse to engage the link)
3. Sections in red apply to recombinant DNA laboratory research. Sections in **lavender** apply to animal laboratory research. Sections in **green** constitute facility and operations requirements for greenhouse biosafety manuals at BSL-2P research. Sections in **Brown** constitute facility and operations requirements for animal ABSL-2 work. Within this section are items in red that are applicable if the ABSL-2 work included rDNA.
4. For topics where lab specific requirements or training are required; information has been developed and is located in the supplemental resources at the back of this document. If upon review of the provided information you determine that it fits your lab situation then simply incorporate it into the body of the document at the designated location by copying and pasting. Unused supplemental resource information may be deleted from the manual. For questions or assistance please contact Levi O’Loughlin, WSU Biosafety Officer, email: [levi.oloughlin@wsu.edu](mailto:levi.oloughlin@wsu.edu), phone: 335-1585.
5. Review all sections of the template to ensure that they accurately reflect lab standards and practices.
6. As part of the BAF (Biosafety Approval Form) review process the WSU Biosafety Officer will review the Lab Biosafety Manual and discuss lab practices with you and members of your lab staff. Please contact the Biosafety Officer to schedule a convenient time to meet.
7. A biosafety risk assessment guide is available. This guide is provided as a means for the PI to provide a written risk assessment for those hazards which are not sufficiently addressed by this manual. When this assessment is needed it should be included as part of the Biosafety Manual. The link to the biosafety risk assessment guide is below:

https://biosafety.wsu.edu/forms-templates-inspection-checklists/

1. For work with Human, and non-human primate tissues, cells or body fluids an Exposure Control Plan will be needed. In general work with these agents only will not require a BAF. Contact Levi O’Loughlin @ 509-335-1585 for more information..
   1. For cell lines or tissues with known or characterized agents a BAF will generally be required. If you have questions regarding this matter please contact the WSU Biosafety Officer.



**BIOSAFETY MANUAL**

**BIOSAFETY LEVEL 2 (BSL-2) LABORATORY FACILITY**

[Building(s)/Room Number(s)]

BAF # XXXX

WASHINGTON STATE UNIVERSITY

Pullman, WA

(Month/Year Originally) Written

(Month/Year Last) Revised

BSO Review: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Date: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Expires: 3 years from end of month of IBC approval.

**Biosafety Manual**

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| **Section 1** | Emergency Response Plan includes: contact information and list of all potentially biohazardous materiel in use. |
| **Section 2** | Standard Operating Procedures for each IBC-approved protocol and Exposure Control Plan as appropriate. |
| **Section 3** | Copy of BAF (Biosafety Approval Form) and Approval letter. Completed Facility Review Checklist. |
| **Section 4** | Training Documentation. |
| **Section 5** | Incident log and Supplemental Resources. |

**NOTE:** IBC approval is needed for activities working with potentially biohazardous agents. Additional approvals may be required e.g.; IRB approval for research activities where human subjects are utilized, IACUC approval for research activities utilizing vertebrate animals.

**SECTION 1**

|  |  |  |
| --- | --- | --- |
| **CONTACT INFORMATION** | | |
| Office/Situation | Name | Phone/email |

|  |  |  |  |
| --- | --- | --- | --- |
| Department | Principal Investigator | Office: Bldg/Room | XXX-XXXX |
|  |  | Lab: Bldg/Room | XXX-XXXX |
|  |  | Home: | XXX-XXXX |
|  |  |  |  |
| Department | Alternate Investigator or Technician | Lab/Office: Bldg/Room | XXX-XXXX |
|  |  | Home: | XXX-XXXX |
| Office of Research Assurances | Levi O’Loughlin –Biosafety Officer | Office: | 335-1585  [levi.oloughlin@wsu.edu](mailto:levi.oloughlin@wsu.edu) |
|  |  |  |  |
| Emergency |  | FIRE | 911 |
|  |  | POLICE | 911 |
|  |  | AMBULANCE | 911 |
|  |  | BIOLOGICAL SPILL | 911 |
|  |  | CHEMICAL SPILL | 911 |
|  |  | MEDICAL EMERGENCY | 911 |
|  |  | RADIOACTIVE SPILL | 911 |
|  |  | SECURITY | 911 |
|  |  |  |  |
| Non-Emergency |  | Police | 334-0802 |
| EH&S |  | Biological Spill | 335-3041 |
| EH&S |  | Body Fluid Exposure | 335-3041 |

Waste Management (Brandy Dean) Bio-Hazardous Waste 335-4530

brandy.dean@wsu.edu Pick-up/disposal

Bio-Hazardous waste disposal SPPM:

[https://policies.wsu.edu/prf/index/manuals/4-00-contents/4-24-disposal-biohazard-wastes/osal-biohazard-wastes/](https://policies.wsu.edu/prf/index/manuals/4-00-contents/4-24-disposal-biohazard-wastes/)

**SECTION 1**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **CONTACT INFORMATION** | | | | |
| Office/Situation | Name | | Phone/email | |
| Radiation |  | Radioactive Spills | | 335-8916 |
| EH&S |  | Chemical Spills | | 335-3041 |
| EH&S |  | Pest Control | | 335-3041 |
| Facilities Operations |  | Maintenance | | 335-9000 |
| List of all [potentially biohazardous materials](https://biosafety.wsu.edu/potentially-biohazardous-materials/) utilized at this facility: | | | | |

**EMERGENCY RESPONSE PROCEDURES**

(Suggested practices are below, modify as appropriate)

**Personal Contamination**

1. Alert people in the spill area.
2. Remove contaminated articles.
3. Vigorously wash exposed area with soap and water for at least 1 minute.
4. If eye exposure occurs, use eye wash per instructions.
5. Obtain medical attention as appropriate.
6. Report spill to supervisor and Biosafety Officer

**Surface Spill in the Lab Outside the Biosafety Cabinet**

1. Clear the room of all personnel.
2. Call WSU Biosafety Officer if you feel that you will need assistance during work hours. During off work hours contact EH&S for assistance 335-3041.
3. Wait at least 30 minutes for aerosols to settle before reentry.
4. Wearing appropriate PPE for size and nature of the spill to include at a minimum: lab coat, gloves and safety glasses.
5. Place dry paper towels to establish a physical barrier between the spill and yourself. Then layer a second set of disinfectant-soaked towels over the spill.
6. Starting from the outside and working in, carefully soak the spill with disinfectant being careful to minimize aerosolization.
7. Decontaminate all items within the spill area. Wait at least the amount of time that follows disinfectant manufacturer’s recommendations for disinfectant contact time to allow for adequate inactivation.
8. Wipe equipment and reusable items with the disinfectant.
9. Wipe up spill and discard contaminated disposables in biohazardous waste stream.
10. If sharps are present, us a mechanical device such as a dustpan and brush to pick up the sharps and place in an approved sharps container.

**Spill Inside a Centrifuge**

1. Clear area of personnel
2. Wait at least 30 minute for aerosols to settle before starting clean-up.
3. Wearing appropriate PPE (lab coat, gloves and safety glasses.)
4. Wipe rotors and buckets with disinfectant then remove to nearest BSC for more extensive decontamination
5. Thoroughly disinfect inside of centrifuge with a disinfectant that is effective for the spilled agent and follow the manufacturer’s recommendations for contact time.
6. Dispose of contaminated materials in the biohazardous waste stream.

**Spill Outside the Lab in Transit**

1. To prevent or minimize a spill, all potentially biohazardous material is to be transported in a primary unbreakable, leak-proof, sealed primary container placed inside a secondary unbreakable, leak-proof, and break proof container. A biohazard symbol should be used on these containers.
2. Should a spill occur in a public area, do not attempt to clean up without the appropriate PPE.
3. Secure the area around the spill.
4. Call EH&S for spills of potential Bloodborne Pathogen containing material. For all other such spills call the WSU Biosafety Officer or EH&S 509-335-3041for assistance.
5. Stand by for further assistance if required.

**Spill Inside the Biosafety Cabinet**

1. Move the glass shield down and wait at least 5 minutes to allow the BSC to filter and clear aerosols.
2. Wearing appropriate PPE at a minimum lab coat, safety glasses and gloves. You may want to double glove in the event the outer pair becomes contaminated.
3. Move the glass shield back up and allow cabinet to equilibrate for an additional 5 minutes.
4. With the BSC operational apply a disinfectant that follows manufacturer’s recommendations and is effective for the spilled agent directly on the spill and on all the potentially exposed surfaces of the cabinet.
5. Wipe up spill with disinfectant soaked towels or other appropriate absorbent material.
6. Wipe the walls, work surfaces, inside of sash and any potentially contaminated equipment with disinfectant soaked towels before removing it from the BSC.
7. Lift exhaust grill and tray and wipe all surfaces.
8. Discard contaminated disposable material using appropriate biohazardous waste disposal procedures.
9. Wipe down contaminated reusable items with disinfectant then place in autoclave bag or autoclave pans with lids for autoclaving.
10. Those items that are non-autoclavable should be wiped down with a disinfectant for an amount of time that follows manufacturer’s recommendations and is effective for spilled agent before removal from BSC.
11. Remove protective clothing, when done and place in biohazard bag for disposal or autoclaving for reusable items.
12. Run the BSC for 10 minutes after clean-up before reusing
13. WASH HANDS!

**Reporting Requirements:** Spills of BSL-2 material resulting in personal contamination and any spill of a BSL-2 agent with a potential exposure such as the release of a BSL-2 organism with excessive splashing and agitation, e.g., performing an aerosolizing activity such as vortexing a capped vial and the tube breaks. The release of a large volume of BSL-2 organisms (as a general rule anything over 10 ml or opening a petri- plate with subsequent spore release outside of a Biosafety Cabinet) etc. or a spill of material containing a high concentration of organisms is reported to the Lab Director and the WSU Biosafety Officer immediately. The Biosafety Officer will assist the PI with bio-risk assessment and emergency response as requested/appropriate. An Incident Reporting Form and an Accident Investigation Form is filled out by the PI and submitted. The Biosafety Officer reviews all incident and accident investigation reports and works collaboratively with the PI to determine what if any measures may be appropriate to mitigate exposure risks in the future.

**ORA (Office of Research Assurances) appreciates receiving incident reports for events that do not result in an exposure “near misses” by filling out an Incident Report and submitting to EH&S. Evaluation of “near misses” can lead to alternative work practices and implementation of engineering controls to minimize future incidents**.

An accident log is available in this manual for use at the discretion of the lab director.

**SECTION 2**

Standard Operating Procedures

The information provided in this template has been obtained from the following sources: “Biosafety in Microbiological and Biomedical Laboratories” ([BMBL](https://www.cdc.gov/labs/BMBL.html) 6th edition). CDC/NIH and “The NIH Guideline for Research Involving Recombinant DNA Molecules” ([NIH Guidelines](https://osp.od.nih.gov/wp-content/uploads/NIH_Guidelines.pdf)), . As well as WSU’s Safety Policies and Procedures Manual (SPPM) and other pertinent regulations and guidelines.

&

Exposure Control Plan (ECP)

As required for work with human and non-human primate tissues, cells or body fluids including blood. An ECP for established or characterized tissue cultures that compliments this BSL-2 biosafety manual is included. All other ECPs must be approved by the BSO or EH&S and then attached to this manual.

# 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 1) Laboratory personnel have specific training in handling pathogenic agents and are supervised by scientists competent in handling infectious agents and associated procedures; 2) Access to the laboratory is restricted when work is being conducted; and 3) All procedures in which infectious aerosols or splashes may be created are conducted in BSC’s or other physical containment equipment.

# Standard Microbiological Practices

* 1. Access to the laboratory is limited or restricted by the laboratory director when work with infectious agents or recombinant DNA is in progress. Additionally, the PI must enforce institutional policies that control access to the laboratory. The door should be kept closed whenever work is being performed with Risk Group 2 agents in the laboratory or animal facility. Insert any specific guidelines for this lab facility here.
  2. Persons must wash their hands after working with potentially hazardous materials, animals**, recombinant DNA material,** removing gloves and prior to leaving the laboratory.
  3. Eating, drinking, smoking, handling contact lenses, applying cosmetics and storing food for human consumption is not permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.
  4. Contact lens users should wear safety glasses, goggles or face shields.
  5. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
  6. 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. Refer to the WSU Safety Policies and Procedures Manual section [S4.25](https://policies.wsu.edu/prf/index/manuals/4-00-contents/4-25-disposal-sharps/)
    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. Per current WSU policy all contaminated sharps are incinerated.
    4. Extreme caution is used when handling needles and syringes to avoid autoinoculation and the generation of aerosol during use and disposal.
    5. Syringes which re-sheath the needle, needle-less systems, and other safety devices should be used. If these are not practical please consult with EH&S to ensure that you have the appropriate documentation required for your current needle use practices.
    6. Broken glass 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. Refer to the WSU Safety Policies and Procedures Manual section [S4.26](https://policies.wsu.edu/prf/index/manuals/4-00-contents/4-26-disposal-laboratory-glass-waste/).
  1. Perform all procedures to minimize the creation of splashes and/or aerosols.
  2. Decontaminate work surfaces daily after completion of work and after any spill or splash of potentially infectious material and **recombinant DNA** with appropriate disinfectant. List disinfectants used to decontaminate work surfaces on a daily basis. See emergency response plan for decontamination of contaminated surfaces.
  3. Decontaminate all cultures, stocks, **recombinant DNA,** and other potentially infectious materials before disposal using an effective method. STATE THE METHOD OF DECONTAMINATION (autoclaving, incineration, bleaching, etc.) OF INFECTIOUS OR REGULATED INCLUDES rDNA. ALSO STATE THE LOCATION OF THE EQUIPMENT NEEDED, SUCH AS AN AUTOCLAVE.
     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.
  4. All infectious liquid or solid wastes are decontaminated before disposal. Contaminated materials that are to be decontaminated at a site away from the laboratory are placed in durable, labeled, leak-proof containers which are sealed and labeled in accordance with SPPM 4.24 before being removed from the laboratory.
     1. Autoclaved waste: Many labs collect potentially biohazardous waste and inactivate it by autoclave, thereby allowing the waste to be disposed in landfill. The waste container should be marked with the biohazard symbol; however the bag should be black, white, or clear and free of the biohazard symbol (once the bagged waste is autoclaved, it is no longer biohazardous and can be sent to the landfill). Any contents or bags marked with the biohazard symbol, labeled “infectious” or “pathogenic,” or other similar markings discovered at any point in the waste stream cannot be disposed of in the landfill and will initiate an emergency biohazard response. Remember that opaque bags may rupture during waste processing to reveal inner biohazard labeling.
     2. Incineration or STI Treatment: Can be bagged (any color or markings) and boxed waste marked and labeled appropriately for STI or incineration. This requires a WSU waste Management Form: [WSU Pullman STI/Incineration Waste Management Request](https://s3.wp.wsu.edu/uploads/sites/2905/2022/05/WSU-Pullman-STI_Incineration-Waste-Management-Request-Form.pdf). Contact waste.management@wsu.edu or call 509-335‐3089 for more information. STI treated waste is state funded and does not cost individual research investigators.
     3. Sharps: All biohazardous sharps (e.g., needles, razor blades) in the state of Washington are disposed by incineration only. Sharps must be collected in a hard-walled container, preferably designed for sharps, and then placed in a box for incinerator processing. Sharps not used or associated with biohazardous materials must be collected in a hard-walled container, preferably designed for sharps, and disposed in the landfill (Pullman campus). Any sharps container marked with the biohazard symbol, labeled “infectious” or “pathogenic,” or other similar markings discovered at any point in the waste stream cannot be disposed of in the landfill and will initiate an emergency biohazard response.
  5. **All Recombinant DNA wastes from labs and animal rooms are appropriately decontaminated before disposal.** Describe decontamination procedures for **rDNA wastes** and **Wastes from Animal Rooms.**
  6. 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 for BSL-2 labs is posed on this sign also in accordance with institutional policy. **Signage on the green house must indicate that a restricted experiment is in progress. This sign must include plant names, person responsible, and any other special requirements for entry access etc.**
  7. An insect and rodent and weed, and arthropod pests and pathogens control program is in effect. The laboratory is routinely inspected for evidence or signs of infestation. If found the Laboratory Director is immediately notified. Additionally, EH&S is contacted at 335-3041 for pest control assistance.
  8. All laboratories are required to supply access to Material Safety Data Sheets (MSDS's) for all chemicals and [infectious biological agents](https://www.canada.ca/en/public-health/services/infectious-diseases.html) in the laboratory. For more information on chemical MSDS's, refer to your Laboratory Safety Manual. For assistance in locating biological MSDS’s refer to [www.absa.org](http://www.absa.org).
  9. 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 an appropriate health care provider for appropriate counseling and guidance.

1. **Special Practices**
   1. All persons entering the laboratory must be advised of the potential hazards and meet specific entry/exit requirements. In general, persons who are at increased risk of acquiring infection or for whom infection may be unusually hazardous are not allowed in the laboratory. The director has the final responsibility for assessing each circumstance and determining who may enter or work in the laboratory.
      1. Personnel (including greenhouse personnel) **(including animal personnel)** are advised of special hazards and are required to read instructions on practices and procedures, and to follow them.
      2. **Include any entry policies here. This includes any entry policies for animal and plant facilities. If there are none state that there are no special requirements for entry.**
   2. Laboratory personnel must be provided medical surveillance and offered appropriate immunizations for agents handled or potentially present in the laboratory.
      1. When appropriate, considering the agent(s) handled baseline serum samples for laboratory and other at-risk personnel are collected and stored. Additional serum specimens may be collected periodically, depending on the agents handled or the function of the facility. Refer to Supplemental Resources A for laboratory Medical Surveillance procedures. Medical Surveillance records shall be kept for the duration of employment plus 30 years.
   3. A laboratory-specific biosafety manual must be prepared and adopted as policy. The biosafety manual must be available and accessible. A greenhouse practices manual is available that advises personnel of consequences of not following protocol. It also outlines contingency plans to be implemented in the event of the unintentional release of organisms.
   4. The laboratory supervisor must ensure that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-2 agents.
   5. Potentially infectious materials must be placed in a durable, leak proof container during collection, handling, processing, storage, or transport within a facility.
   6. 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.
      3. **PROTOCOL FOR DAILY SURFACE DECONTAMINATION: Must indicate the rationale for disinfectant used in regard to the following factors. (You needn’t address each area just outline the pertinent factors used for disinfectant selection.)**
         1. **Spectrum of activity, efficacy, susceptibility to inactivation by organic matter, compatibility with soaps and detergents used, toxicity to personnel/animals, contact time required, optimal temperature, residual activity, corrosiveness, environmental effects, and cost:**
   7. Incidents that may result in exposure to infectious materials must be immediately evaluated and treated according to procedures described in the laboratory biosafety manual. All such incidents must be reported to the laboratory supervisor. Medical evaluation, surveillance, and treatment should be provided, and appropriate records maintained.
      1. Spill cleanup procedures are listed in the Emergency Response section 1 of this manual**.** Medical evaluation, surveillance, and treatment are provided as appropriate and written records are maintained for at least 30 years. **List all Medical Surveillance in place for this protocol here.**
      2. **All accidents resulting in overt or potential exposure are immediately reported to the Biosafety Officer and /or Institutional Biosafety Committee.**
      3. PI will report any greenhouse accident involving the inadvertent release or spill of recombinant microorganisms to the Greenhouse Director, Biosafety Officer and/or the IBC.
      4. See the emergency response plans in section one for more specific information on bio-agent response requirements.
   8. Animals and plants not associated with the work being performed must not be permitted in the laboratory.
   9. All procedures involving the manipulation of infectious materials that may generate an aerosol should be conducted within a BSC or other physical containment devices.
   10. Biohazard signage is used when special provisions for entry are required e.g. PPE or immunization. If your lab utilizes recombinant DNA and has no special provisions for entry you may delete this item as well as 4 above and 6 below. If your lab utilizes recombinant DNA and there are special entry requirements you must stipulate those requirements here and can delete number 4 above and 6 below**.**
   11. Access to greenhouse is limited to individuals directly involved with the experiments. **List the requirements here.**
   12. Refer to Supplemental Resources D for examples of specific safe lab practices, risks associated with agents encountered in the lab, and special precautions needed to work with the agents present. Records are kept of experiments and movement in/out of the greenhouse. Containment is required for containment in/out of the greenhouse. Experimental organisms are biologically inactivated at the end of the experiment. Gravel in greenhouse (if applicable) is decontaminated periodically. Appropriate caging and precautions for escape of motile organisms. **Include these practices here.**
   13. An area of the lab needs to be designated and SOPs need to be described for particularly hazardous chemicals in Section IV of the Lab Safety Manual.
   14. Laboratory personnel receive appropriate training on the potential hazards associated with the work involved, the necessary precautions to prevent exposures, and the exposure evaluation procedures. Personnel receive annual updates, or additional training as necessary for procedural or policy changes. Refer to Section 4 for list of training options and to Supplemental Resources B for an optional training example. **Include all appropriate training requirements here. Be sure that this training is documented on the training log in section 4 of this manual**.
   15. Special care is taken to avoid skin contamination with infectious materials and with organisms containing recombinant DNA molecules; gloves should be worn when handling experimental animals and when skin contact with the agent is unavoidable.
2. **Safety Equipment (Primary Barriers)**
   1. Properly maintained BSC’s (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 concentration 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. Alternative 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 material 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) &* Personal Protective Equipment

* 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 disinfectants.
  5. Laboratory windows that open to the exterior are not recommended. However, if a laboratory does have windows that open to the exterior, the must be fitted with fly screens.
  6. BSC’s must be installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSC’s 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. BSC’s can also be connected to the laboratory exhaust system by either a thimble (canopy) connection of 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).
  12. A written assessment of laboratory risks must be performed to determine the appropriate personal protective equipment (PPE) required for a particular task. The appropriate PPE will be assigned for each potentially hazardous task performed in the lab. Include decontamination and waste disposal processes. \***Fill in a row on the PPE assessment form on the next page for each laboratory task**.
  13. This is different than the Biosafety Risk Assessment. Biosafety Risk Assessment is a process used to identify the hazardous characteristics of a known infectious or potentially infectious agent or material, the activities that can result in a person’s exposure to an agent, the likelihood that such exposure will cause a LAI, and the probable consequences of such an infection. The information identified by risk assessment will provide a guide for the selection of appropriate biosafety levels and microbiological practices, safety equipment, and facility safeguards that can prevent LAIs.

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| **WORKPLACE HAZARD ASSESSMENT CERTIFICATION FORM** | | | | |
| ***Instructions: Complete form using Personal Protective Equipment Hazard Assessment Guidelines. Completed form is to be retained for departmental records.*** | | | | |
| **Supervisor conducting the hazard assessment:** | | | **Date of hazard assessment:** | |
| **Work Activity Assessed** | **Location of Assessment (Blg/Rm)** | **Hazard(s) Identified** | | **PPE Selected (Make & Model #)** |
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| I, , certify that the assessment of the identified work activities has been performed.  **Date:**  ***Signature*** | | | | |

**Use the following examples as an assessment guide:**

* + - * Face protection (goggles, mask, or face shield) is used for anticipated splashes or sprays of potentially biohazardous materials, when manipulated outside of a biosafety cabinet.
      * Laboratory coats, gowns, smocks, or uniforms are worn while in the laboratory (greenhouse). Before leaving the (greenhouse) laboratory for non-laboratory (non-greenhouse) areas, this protective clothing is removed and left in the laboratory (greenhouse). All protective clothing is either disposed of in the laboratory (greenhouse) or laundered by the institution. Taking home protective equipment is prohibited. At the discretion of the Greenhouse director disposable clothing e.g. solid front or wrap-around gowns etc. shall be worn if deemed necessary due to potential dissemination of experimental microorganisms.
      * Protective clothing is changed whenever overtly contaminated. It is either disposed of in the lab or laundered on-site by the institution.
      * Gloves are worn when handling infected animals and when hands may contact infectious or hazardous materials, surfaces, or equipment. Wearing two pairs of gloves may be appropriate; if a spill or splatter occurs; the hand will be protected after the contaminated glove is removed. Gloves are disposed of when contaminated or integrity is compromised and removed when work is completed. Gloves are not worn outside of the laboratory. Disposable gloves are not washed or reused. Non-latex gloves are recommended by WSU Environmental Health and Safety.
      * If respiratory protection is required, contact WSU Environmental Health and Safety for a medical evaluation, hazard assessment, and fit testing before doing any work requiring the use of a respirator.
      * Gloves are worn if skin on hands is broken or has a rash.
      * Gloves are not worn outside the lab or when touching “clean” surfaces (e.g. telephone, keyboards, doorknobs, and drawer handles, etc.)
      * Gloves are disposed of when overtly contaminated, work with infectious materials is completed, or integrity is compromised. Disposable gloves are not reused.
      * Lab coats or gowns are worn when working with biological agents.

1. **Plant Facilities & Practices**
   1. **Access to the greenhouse shall be limited or restricted, at the discretion of the Greenhouse director, to individuals directly involved with the experiments when they are in progress.**
   2. **Personnel shall be required to read and follow instructions on BL1&2-P practices and procedures. All procedures shall be conducted in accordance with accepted greenhouse practices that are appropriate to the experimental organism(s).**
   3. **For BL1-P & BL2-P, a record shall be kept of experiments currently in progress in the greenhouse facility.**
   4. **For BL2-P, a record shall be kept of experimental plants, microorganisms, or small animals that are brought into or removed from the greenhouse facility**
   5. **The Principal Investigator shall report any greenhouse accident involving the inadvertent release or spill of microorganisms to the Greenhouse Director, and the Institutional Biosafety Committee.**
   6. **Experimental organisms shall be rendered biological inactive by appropriate methods before disposal outside of the greenhouse facility**
   7. **Decontamination of run-off water is not necessarily required. If part of the greenhouse is composed of gravel or similar material, appropriate treatments should be made periodically to eliminate, or render inactive any organisms potentially entrapped by the gravel**
   8. **A program shall be implemented to control undesired species (e.g., weed, rodent, or arthropod pest and pathogens) by methods appropriate to the organisms and in accordance with applicable state and Federal laws.**
   9. **Arthropods and other motile macroorganisms shall be housed in appropriate cages. If macroorganisms (e.g. flying arthropods or nematodes) are released with the greenhouse, precautions shall be taken to minimize escape from the greenhouse facility.**
   10. **Experiments involving other organisms that require a containment level lower than BL1-P may be conducted in the greenhouse concurrently with experiments that require BL1-P containment provided that all work is conducted in accordance with BL1-P greenhouse practices**
   11. **Experiments involving other organisms that require a containment level lover than BL2-P may be conducted in the greenhouse concurrently with experiments that require BL2-P containment provided that all work is conducted in accordance with BL2-P greenhouse practices.**
   12. **A sign shall be posted indicating that either a BL-1 or a BL-2 experiment is in progress. The sign shall indicate the following (i) the name of the responsible individual (ii) the plants in use, and (iii) any special requirements for using the area.**
   13. **If organisms are used that have a recognized potential for causing serious detrimental impacts on managed or natural ecosystems, their presence shall be indicated on a sign posted on the greenhouse access doors.**
   14. **If there is a risk to human health, a sign shall be posted incorporating the universal biosafety symbol**
   15. **Materials containing experimental microorganism, which are brought into or removed from the greenhouse facility in a viable or intact state, shall be transferred in a closed non-breakable container**
   16. **In a BL2-P, a greenhouse practices manual shall be prepared or adopted. This manual shall: 9i) advise personnel of the potential consequences if such practices are not followed and outline contingency plans to be implemented in the event of unintentional organism release.**
   17. **In BL1-P, the green house floor may be composed of gravel or other porous material. At a minimum, impervious (e.g., concrete) walkways are recommended.**
   18. **In BL2-P, a greenhouse floor composed of an impervious material. Concrete is recommended, but gravel or other porous material under benches is acceptable unless propagules of experimental organisms are readily disseminated through soil. Soil beds are acceptable unless propagules or experimental organisms are readily disseminated through soil.**
   19. **In BL2-p, Windows and other opening in the walls and roof or the greenhouse facility may be open for ventilation as need for proper operation and do not require any special barrier to exclude pollen or microorganisms; however, screens are required to exclude small flying animals (e.g., arthropods and birds).**
   20. **An autoclave shall be available for the treatment of contaminated greenhouse materials.**
   21. **If intake fans are used, measures shall be taken to minimize the ingress of arthropods. Louvers or fans shall be constructed such that they can only be opened when the fan is in operation**
   22. **For BL1-P & BL2-P greenhouse containment requirements may be satisfied by using a growth chamber or growth room within a building provided that the external physical structure limits access and escape of microorganisms and macroorganisms in a manner that satisfied the intent of the foregoing clauses.**

# Animal Facilities & Practices

* 1. **The animal facility director establishes and enforces policies, procedures, and protocols for institutional policies and emergency situations.**

**Each institute must assure that worker safety and health concerns are addressed as part of the animal protocol review.  
Prior to beginning a study animal protocols must also be reviewed and approved by the IACUC and the Institutional Biosafety Committee.**

* 1. **A safety manual specific to the animal facility is prepared or adopted in consultation with the animal facility director and appropriate safety professionals.**

**The safety manual must be available and accessible. Personnel are advised of potential hazards and are required to read and follow instructions on practices and procedures.**

**Consideration should be given to specific biohazards unique to the animal species and protocol in use.**

* 1. **The Supervisor must ensure that animal care, laboratory and support personnel receive appropriate training regarding their duties, animal husbandry procedure, potential hazards, manipulations of infectious agents, necessary precautions to prevent hazard or exposures, and hazard/exposure evaluation procedures (physical hazards, splashes, aerosolization, etc.). Personnel must receive annual updates or additional training when procedures or policies change. Records are maintained for all hazard evaluations, employee training sessions and staff attendance.**
  2. **Appropriate medical surveillance program is in place, as determined by risk assessment. The need for an animal allergy prevention program should be considered.  
     Facility supervisors should ensure that medical staff is informed of potential occupational hazards within the animal facility, to include those associated with research, animal husbandry duties, animal care and manipulations.  
     Personal health status may impact an individual’s susceptibility to infection, ability to receive immunizations or prophylactic interventions. Therefore, all personnel and particularly women of child-bearing age should be provided 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 health care provider for appropriate counseling and guidance.  
     Personnel using respirators must be enrolled in an appropriately constituted respiratory protection program.**
  3. **A sign incorporating the universal biohazard symbol must be posted at the entrance to areas where infectious material and/or animals are housed or are manipulated when infectious agents are present. The sign must include the animal biosafety level, general occupational health requirements, personal protective equipment requirements, the supervisor’s name (or other responsible personnel), telephone number, and required procedures for entering and exiting the animal areas. Identification of specific infectious agent is recommended when more than one agent is being used within an animal room.  
     Security-sensitive agent information and occupational health requirements should be posted in accordance with institutional policy.  
     Advance consideration should be given to emergency and disaster recovery plans, as a contingency for man-made or natural disasters.**
  4. **Access to the animal room is limited. Only those persons required for program or support purposes are authorized to enter the animal facility and the areas where infectious materials and/or animals are housed or are manipulated.  
     All persons including facility personnel, service workers, and visitors are advised of the potential hazards (natural or research pathogens, allergens, etc.) and are instructed on the appropriate safeguards.**
  5. **Protective laboratory coats, gowns, or uniforms are recommended to prevent contamination of personal clothing.  
     Gloves are worn to prevent skin contact with contaminated, infectious and hazardous materials and when handling animals.  
     Gloves and personal protective equipment should be removed in a manner that minimized transfer of infectious materials outside of the areas where infectious materials and/or animals are housed or are manipulated.  
     Persons must wash their hands after removing gloves, and before leaving the area where infectious materials and/or animals are housed or are manipulated.  
     Eye and face and respiratory protection should be used in rooms containing infected animals, as dictated by the risk assessment.**
  6. **Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human use should only be done in designated areas and are not permitted in animal or procedure rooms.**
  7. **All procedures are carefully performed to minimize the creation of aerosols or splatter of infectious materials and waste.**
  8. **Mouth pipetting is prohibited. Mechanical pipetting devices must be used.**
  9. **Policies for the safe handling of sharps, such as needles, scalpels, pipettes, and broken glassware must be developed and implemented. When applicable, animal supervisors should adopt improved engineering and work practice controls that reduce the risk of sharps injuries. Precautions, including those listed below, must always be taken with sharp items. These include:**
     1. **Needles and syringes or other sharp instruments are limited to use in the animal facility when there is not alternative for such procedures as parenteral injection, blood collection, or aspiration of fluids from laboratory animals and diaphragm bottles.**
     2. **Disposable needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal. Used disposable needles must be carefully placed in puncture-resistant containers used for sharps disposal. Sharps containers should be located as close to the work site as possible.**
     3. **Non-disposable sharps must be placed in a hard-walled container for transport to a processing area for decontamination.**
     4. **Broken glassware must not be handled directly; it should be removed using a brush and dustpan, tongs, or forceps. Plastic ware should be substituted for glassware whenever possible.**
     5. **Equipment containing sharp edges and corners should be avoided.**
  10. **Equipment and work surfaces are routinely decontaminated with an appropriate disinfectant after work with an infectious agent, and after any spills, splashes, or other overt contamination.**
  11. **Animal and plants not associated with the work being performed must not be permitted in the areas where infectious materials and/or animals are housed or are manipulated.**
  12. **An effective integrated pest management program is required.**
  13. **All wastes from the animal room (including animal tissues, carcasses, and bedding) are transported from the animal room in leak-proof containers for appropriate disposal in compliance with applicable institutional, local and state requirements.  
      Decontamination of all potentially infectious material before disposal is required using an effective method. Insert decontamination methods here.**
  14. When an animal containing recombinant DNA or a recombinant DNA-derived organism is euthanized or dies, the carcass shall be disposed of to avoid its use as food for human beings or animals.
  15. A permanent record shall be maintained of the experimental use and disposal of each animal or group of animals.
  16. To animal containment area shall be locked.
  17. The animal facility director shall establish policies and procedures whereby only persons who have been advised of the potential hazard and who meet any specific entry requirements (e.g., vaccination) may enter the animal rooms.
  18. Animals of the same or different species, which are not involved in the work being performed, shall not be permitted in the animal area.
  19. Appropriate steps should be taken to prevent horizontal transmission or exposure of animal personnel. If the agent used as a vector is known to be transmitted by a particular route (e.g., arthropods), special attention should be given to preventing spread by that route. In the absence of specific knowledge of a particular route of transmission, all potential means of horizontal transmission (.e.g., arthropods, contaminated bedding or animal waste, etc.) should be prevented.
  20. Eating, drinking, smoking, and applying cosmetics shall not be permitted din the animal work area
  21. Individuals who handle materials and animals containing recombinant DNA molecules shall be required to wash their hands before exiting the containment area
  22. Surfaces of the animal containment area shall be impervious to water and resistant to acids, alkalis, organic solvents and moderate heat.
  23. The animal containment area shall be designed so that it can be easily cleaned
  24. Windows that open shall be fitted with fly screens
  25. If arthropods are used in the experiment or the agent under study can be transmitted by an arthropod, interior work areas shall be appropriately screened (52 mesh). All perimeter joints and opening shall be sealed and additional arthropod control mechanisms used to minimized arthropod entry and propagation, including appropriate screening of access doors or the equivalent.
  26. The containment area shall be patrolled or monitored at frequent intervals.
  27. All genetically engineered neonates shall be permanently marked with 72 hours after birth, if their size permits. If their size does not permit marking, their containers should be marked. In addition, transgenic animals should contain distinct and biochemically assayable DNA sequences that allow identification of transgenic animals from among non-transgenic animals.
  28. A double barrier shall be provided to separate male and female animals unless reproductive studies are part of the experiment or other measures are taken to avoid reproductive transmission. Reproductive incapacitation may be used.
  29. The containment area shall be in accordance with state and Federal laws and animal care requirements.
  30. Animals shall be confined to securely fences areas or be in enclosed structures (animal rooms) to minimize the possibility of theft or unintentional release.
  31. **Animal care staff and routine support personnel must be provided a medical surveillance program as dictated by the risk assessment, and administered appropriate immunization for agents handled or potentially present, before entry into animal rooms.  
      When appropriate, a base line serum sample should be stored.**
  32. **Procedures involving a high potential for generating aerosols should be conducted with a biosafety cabinet or other physical containment device. When a procedure cannot be performed with a biosafety cabinet, a combination of personal protective equipment and other containment devices must be used.  
      Consideration should be given to the use of restrain devices and practices that reduce the risk of exposure during animal manipulations (e.g., physical restraint devices, chemical restrain medications, etc.).**
  33. **Decontamination is recommended for all potentially infectious materials and animal waste before movement outside the areas where infectious materials and/or animals are housed or are manipulated by an appropriate method (e.g. autoclave, chemical disinfection, or other approved decontamination methods). This includes potentially infectious animal tissues, carcasses, contaminated bedding, unused feed, sharps and other refuse.  
      Consideration should be given to means for decontaminating routing husbandry equipment, sensitive electronic and medical equipment.  
      Materials to be decontaminated outside the immediate areas where infectious materials and/or animals are housed or are manipulated must be placed in a durable. Leak proof, covered container and secured for transport. The outer surface of the container is disinfected prior to moving materials. The transport container must contain a universal biohazard label.  
      Develop and implement an appropriate waste disposal program in compliance with applicable institutional, local and state requirements. Autoclaving of content prior to incineration is recommended.**
  34. **Equipment, cages and racks should be handled in a manner that minimized contamination of other areas.  
      Equipment must be decontaminated before repair, maintenance, or removal from the areas where infectious materials and/or animals are housed or are manipulated.  
      Spills involving infectious materials must be contained, decontaminated, and cleaned up by staff properly trained and equipped to work with infectious material.**
  35. **Incidents that may result in exposure to infectious materials must be immediately evaluated and treated according to procedures described in the animal facility safety manual. All such incidents must be reported to the animal facility supervisor and Mike Kluzik at EH&S. Medical evaluation, surveillance, and treatment should be provided as appropriate and records maintained.**
  36. **Properly maintained BSC’s, personal protective equipment (e.g., gloves, lab coats, face shields, respirators, etc.) and/or other physical containment devices or equipment, are used whenever conducting procedures with a potential for creating aerosols or splashes. These include necropsy of infected animals, harvesting of tissues or fluids from infected animals or eggs, and intranasal inoculation of animals.  
      When indicated by risk assessment, animals are housed in primary biosafety containment equipment appropriate for the animal species such as solid wall and bottom cages covered with filter bonnets for rodents, or larger cages placed in inward flow ventilated enclosures or other equivalent primary containment systems for larger animal cages.**
  37. **A risk assessment should determine the appropriate type of personal protective equipment to be utilized.  
      Scrub suits and uniforms are removed before leaving the animal facility. Reusable clothing is appropriately contained and decontaminated before being laundered. Protective clothing should never be taken home.  
      Gowns, uniforms, lab coats and personal protective equipment are worn while in the areas where infectious materials and/or animals are housed or manipulated and removed prior to exiting. Disposable personal protective equipment and other contaminated waste are appropriately contained and decontaminated prior to disposal.**
  38. **Eye and face protection (mask, goggles, face shield or other splatter guard) are used for anticipated splashes/sprays from infectious or other hazardous materials and when the animal or 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 should also wear eye protection when entering areas with potentially high concentrations or airborne particulates.**
  39. **Gloves are worn to protect hands from exposure to hazardous materials. A risk assessment should be performed to identify the appropriate glove for the task and alternative to latex gloves should be available.  
      Gloves are changed when contaminated, integrity has been compromised, or when otherwise necessary.  
      Gloves must not be worn outside the animal rooms.  
      Gloves and personal protective equipment should be removed in a manner that prohibits transfer of infectious materials.  
      Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated waste.  
      Persons must wash their hands after handling animals and before leaving the areas where infectious material and/or animals are housed or are manipulated. Hand washing should occur after the removal of gloves.**
  40. **The animal facility is separated from areas that are open to unrestricted personnel traffic within the building. External facility doors are self-closing and self-locking.  
      Access to the animal facility is restricted.  
      Doors to areas where infectious material and/or animals are housed, open inward, are self-closing, are kept closed when experimental animals are present, and should never be propped open. Doors to cubicles inside an animal rom may open outward or slide horizontally or vertically.**
  41. **A hand washing sink is located at the exit of the areas where infectious materials and/or animals are housed or are manipulated. Additional sinks for hand washing should be located in other appropriate locations within the facility.  
      If the animal facility has segregated areas where infectious materials and/or animals are housed or manipulated, a skink must also be available for hand washing at the exit from each segregated area.  
      Sink traps are filled with water, and/or appropriate liquid to prevent the migration of vermin and gases.**
  42. **The animal facility is designed, constructed, and maintained to facilitate cleaning and housekeeping. The interior surfaces (walls, floors and ceilings) are water resistant.  
      Penetrations in floors, walls and ceiling surfaces are sealed, to include openings around ducts, doors and door frames, to facilitate pest control and proper cleaning.  
      Floors must be slip resistant, impervious to liquids, and resistant to chemicals.**
  43. **Cabinets and bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals. Spaces between benches, cabinets, and equipment should be accessible for cleaning.  
      Furniture should be minimized. Chairs used in animal areas must be covered with a non-porous material that can be easily cleaned and decontaminated. Furniture must be capable of supporting anticipated loads and uses. Sharp edges and corners should be avoided.**
  44. **External windows are not recommended; if present, windows should be sealed and must be resistant to breakage. The presence of windows may impact facility security and therefore should be assessed by security personnel.**
  45. **Ventilation should be provided in accordance with “the Guide for Care and Use of Laboratory Animals”***.* **The direction of airflow into the animal facility is inward**; **animal rooms should maintain inward directional airflow compared to adjoining hallways. A ducted exhaust air ventilation system is provided. Exhaust air is discharged to the outside without being re-circulated to other rooms.  
      Ventilation system design should consider the heat and high moisture load produced during the cleaning of animal rooms and the cage wash process.**
  46. **Internal facility appurtenances, such as light fixtures, air ducts, and utility pipes, are arranged to minimize horizontal surface areas, to facilitate cleaning and minimize the accumulation of debris or fomites. Floor drains must be maintained and filed with water, and/or appropriate disinfectant to prevent the migration of vermin and gases.**
  47. **Cages should be autoclaved or otherwise decontaminated prior to washing. Mechanical cage washer should have a final rinse temperature of at least 180 degrees F. The cage wash area should be designed to accommodate the use of high-pressure spray systems, humidity, strong chemical disinfectants and 180 degree F water temperatures, during the cage/equipment cleaning process.**
  48. **Illumination is adequate for all activities, avoiding reflections and glare that could impede vision.**
  49. **If BSC’s are present, they must be installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSC’s should be located away from doors, heavil8y traveled laboratory areas, and other possible airflow disruptions.  
      HEPA filtered exhaust air from a CLASS II BSD 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. BSC’s 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. Correct performance of the BSD’s should be recertified at least once a year.  
      All BSC’s should be used according to manufacturer’s recommendation, to protect the worker and avoid creating a hazardous environment form volatile chemical and gases.**
  50. **If vacuum service (i.e., central or local) is provided, each service connection should be fitted with liquid disinfectant traps and an in-line HEPA filter, placed as near as practicable to each use point or service cock. Filters are installed to permit in-place decontamination and replacement.**
  51. **An autoclave should be considered in the animal facility to facilitate decontamination of infectious materials and waste.**
  52. **Emergency eyewash and shower are readily available; location is determined by risk assessment.**

**EXPOSURE CONTROL PLAN**

**Bloodborne Pathogen Standard Information - Established Human Cell Lines**

Both OSHA and the State of Washington’s Labor and Industries Bloodborne Pathogen Standard (BPS) are regulations intended to protect employees from exposure to human blood or other potentially infectious materials that may contain bloodborne pathogens. Since it is not feasible to test for the presence and thus ascertain the absence of every human blood-borne pathogen (BBP) that may be present in human cell lines, the BPS must be addressed when these cell lines are used.

Per the BPS, employers must implement an Exposure Control Plan, provide employees specific training, and offer hepatitis B virus vaccinations (if there is potential for employees to be exposed to that virus). Since the biosafety level 2 precautionary measures prescribed in this manual are similar, this manual is considered this lab’s BBP Exposure Control Plan. To address the BPS specific training requirements, the information in this manual in combination with the information presented below must be reviewed and understood by individuals who may have contact with human cell lines. As such, please review this information and if you have any questions during the review, feel free to ask your lab’s supervisor or contact Environmental Health & Safety (EH&S) at 335-3041.

A copy of the State of Washington BPS; Occupational Exposure to Bloodborne Pathogens, Washington Administrative Code (WAC) Chapter 296-823, can be obtained by accessing them on-line at: [Occupational Exposure to Bloodborne Pathogens](https://apps.leg.wa.gov/wac/default.aspx?cite=296-823) or from EH&S.

A copy of this lab’s Exposure Control Plan, which is this biosafety manual and appendix, can be obtained from the lab supervisor. This plan was established in accordance with the WSU Institutional plan, SPPM 2.44 Bloodborne Pathogens, which can be obtained by accessing: <https://apps.leg.wa.gov/wac/default.aspx?cite=296-823> or from EH&S.

Bloodborne Pathogens:

Human immunodeficiency virus (HIV), hepatitis B virus (HBV), hepatitis C virus (HVC) are examples of bloodborne pathogens. Exposure to BBPs includes contact with mucous membranes such as in eyes, mouth, or nose, broken skin, or by injection with a contaminated sharp object. Personnel may develop infections upon exposure to these human body fluids. Infections can be mild with a quick recovery time (e.g., HBV), or severe with life-threatening consequences (e.g., HIV and/or HBV) depending upon the general health condition of each employee.

Human body fluids which could contain and possibly spread HIVare human blood in any body fluids, semen, breast milk, vaginal secretions, amniotic fluids, cerebrospinal fluids, pericardial fluids, alveolar fluids, tears, saliva, and/or urine. Other potentially infectious materials include cell, tissue, or organ cultures containing HIV; HIV- or HBV-containing culture medium or other solutions; and blood, organs, or other tissues from experimental animals infected with HIV or HBV.

Precautionary Measures:

Tasks or activities involving primary or established human cell lines could involve exposure to potentially infectious materials. The equipment and safer medical devices, work practices, and personal protective equipment (PPE) described in this biosafety manual will prevent or reduce this exposure potential. Information about PPE used in this lab for work with these materials can be found in section D of this manual. Procedures for post exposure evaluations and follow-up in the event of an accidental exposure to these materials can be found in Section 3, Supplemental Resource B of this manual.

Medical Surveillance:

BBP regulations include provisions for employers to offer employees hepatitis B virus vaccinations. Any laboratory that is dealing directly with primary human or primate cells or tissues, especially human or primate liver should be concerned with the hepatitis B virus and offer the HBV vaccinations.

Since the discovery of hepatitis B virus (HBV) there has been considerable effort expended to find a cell culture system that will produce infectious hepatitis B virus. It is possible to infect primary (that is, cells derived from a liver) human or humanoid hepatocytes (liver cells) but, this infection last for only a brief period of time. These hepatocytes then become refractory to a HBV viral infection. There are no other cells that are known to be permissive (allow an infection to occur with infectious virus produced) for a hepatitis B virus infection. Since no other cell lines, including those used in this lab, are known to be infected with hepatitis B virus, and based on all available data, cannot be infected with HBV, hepatitis B vaccinations are not being offered to personnel in this lab unless specifically requested.

Please notify your supervisor or contact EH&S if you have any questions to obtain for more information.

**SECTION 3**

Copy of:

1. BAF (Biosafety Approval Form).
2. IBC Approval letter for BAF.
3. Lab Facility Review (Completed Checklist).

**Section 4 - Training**

Training Required for Bio-Hazards Associated with **(Lab Building/Room Number)**

At a minimum this training should include:

1. Basic biosafety training appropriate for the research activities of this protocol. See [WSU Web Training](https://hrs.wsu.edu/training/) for some web training options.
2. Special local practices, equipment, organism pathogenesis and routes of exposure, proper use of protective measures such as personal protective equipment, use of biological safety cabinets, or any other specific training necessary to safely work with the organism or equipment. The WSU SPPM, Lab Safety Manual, or equipment owner/operators manual may be referenced if the material is already addressed.
3. Sharps training: if sharps are to be used in the laboratory. Sharps training can be found on [Percipio Compliance](https://hrs.wsu.edu/training/skillsoft-percipio/). Also see: the [Sharps Injuries Factsheet](file:///C:\Users\calvin.franklin\Downloads\1.%09:%20https:\ehs.wsu.edu\ehs-training\factsheets\factsheets-faqneedlesticks09\) for more information.
4. Biological MSDS for all infectious biological agents.
5. A Biological Safety Cabinet, (BSC) Operation Procedures should be included for all protocols that will use a BSC. Proper use of biological safety cabinets should be referenced to the cabinet owners/operators manual.

**Section 4 4– TRAINING DOCUMENTATION**

Laboratory Personnel Listing for **\*(Lab Building/Room Number)**

The personnel listed below have been trained on the potential hazards of the work involved, the necessary precautions to avoid exposure, the exposure evaluation process, and safe operation of the equipment used.

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| Name | Signature | Position | Date Manual Reviewed | Date Biosafety Training Completed | Annual Retraining Initial and date |
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**SECTION 5**

INCIDENT LOG

&

Supplementary Resources

SECTION 5: SPILL & ACCIDENT RECORD for **[Building/room(s)]**

**Chemical and Biological Spills**:

Trained laboratory workers may clean up relatively small spills. Seek assistance when personnel qualified to clean up the release are not available, proper PPE or spill kit supplies are not available, spill cannot be identified, chemical is highly flammable or highly toxic, or the spill is outside the employee’s immediate work area. For spill assistance call Fire Services/EHS by dialing 911 or call EHS directly at 335-3041.

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| --- | --- | --- | --- | --- |
| **Date of spill** | **Substance spilled** | **Location of spill** | **Name of personnel involved** | **Method of clean-up** |
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**Accidents:**

Call 911 for help when required. Fill out all required accident forms located [HERE](https://hrs.wsu.edu/employees/disability-services/workers-compensation/)

Supplemental Resource A Medical Surveillance\*

To determine if Medical Surveillance is appropriate a risk assessment should be made based on the following risks:

* The probability of infection
  + Implies an estimate of numbers exists
  + Predict an outcome given similar events
* What is the natural host?
* Does the agent cross species barriers?
* Wild-type agent or attenuated?
* Infectious for normal healthy adult?
* What if adult is immunocompromised?
* Mode of Transmission
  + Contact
  + Fomites
  + Mucous membrane exposure
  + Ingestion
  + Inoculation or insect bites
  + Inhalation
  + Sex
* Volume being manipulated?
* Concentration of agent?
* Infectious dose?
* Past history of lab-associated infection?
* Secondary spread in community?
* Prophylaxis
  + Immunizations available?
  + Pharmaceuticals?
  + Effectiveness?
* Post-Exposure
  + Anti-microbial agents?
  + Pharmaceuticals?
  + Effectiveness?
* Dealing with an unknown agent?
  + Epidemiological data
  + Patterns parallel to other agents
  + Data from animal studies
  + Route of infection

\* (At the discretion of the Laboratory Director, if a Medical Surveillance program is established, it is in accordance with the WSU Laboratory Safety Manual Section II-10 and WAC 296-62-40001 for chemical exposures. For microbiological exposures, the program is developed to detect immunological exposures related to the microorganisms used within the laboratory. State whether a Medical Surveillance Program is instituted and if serum samples are being stored and if immunizations are required to work in the lab. Include a procedure for obtaining medical treatment if needed.)

Supplemental Reference B

Biological Safety Cabinet Operating Procedures **(Lab Building/Room Number)**

1. **Prior to beginning work in the Biosafety Cabinet:**

* Air curtain disruptions will compromise the fragile air barrier of the cabinet. Prepare a written checklist of materials necessary for a particular activity and place necessary materials in the BSC before beginning work, in order to minimize the number and extent of air curtain disruptions.
* Laboratory coats should be worn buttoned over street clothing; wear latex, vinyl, nitrile or other suitable gloves to provide hand protection.
* Before beginning work, the investigator should adjust the stool height so that his/her face is above the front opening.
* If the cabinet has been shut down, the blowers should be operated at least four minutes before beginning work to allow the cabinet to “purge.” This purge will remove any particulates suspended in the cabinet.
* If there is a drain valve under the work surface, it should be closed prior to beginning work in the BSC.
* Only the materials and equipment required for the immediate work should be placed in the BSC.
* All materials should be placed as far back in the cabinet as practical, toward the rear edge of the work surface and away from the front grille of the cabinet.
* Aerosol-generating equipment (e.g., vortex mixers, tabletop centrifuges) should be placed toward the rear of the cabinet to take advantage of the air split.
* The correct sash position (usually 8˝ or 10˝ above the base of the opening) should be indicated on the front of the cabinet. On most BSCs, an audible alarm will sound if the sash is in the wrong position while the fan is operating.

1. **While working in Biosafety Cabinet:**

* Good microbiological techniques should always be used when working in a BSC
* The rapid movement of a worker’s arms in a sweeping motion into and out of the cabinet will disrupt the air curtain and compromise the partial containment barrier provided by the BSC. Move arms in and out slowly, perpendicular to the face opening of the cabinet, in order to reduce this risk.
* Manipulation of materials should be delayed for approximately one minute after placing the hands/arms inside the cabinet. This allows the cabinet to stabilize, to “air sweep” the hands and arms, and to allow time for turbulence reduction
* The front grille must not be blocked with toweling, research notes, discarded plastic wrappers, pipetting devices, etc. All operations should be performed on the work surface at least four inches in from the front grille.
* Plastic-backed absorbent toweling can be placed on the work surface but not on the front or rear grille openings.
* Bulky items such as biohazard bags, discard pipette trays and vacuum collection flasks should be placed to one side of the interior of the cabinet
* Only horizontal pipette discard trays containing an appropriate chemical disinfectant should be used within the cabinet
* Potentially contaminated materials should not be brought out of the cabinet until they have been surface decontaminated. Alternatively, contaminated materials can be placed into a closable container for transfer to an incubator, autoclave or another part of the laboratory prior to removal from the BSC.
* The workflow should be from “clean to dirty.” Materials and supplies should be placed in the cabinet in such a way as to limit the movement of “dirty” items over “clean” ones.

1. **After working in biosafety cabinet:**

* The work surface, the interior walls (except the supply filter diffuser), and the interior surface of the window should be wiped with 70% ethanol (EtOH), a 1:100 dilution of household bleach (i.e., 0.05% sodium hypochlorite), or other disinfectant as determined by the investigator to meet the requirements of the particular activity.
* When bleach is used, a second wiping with sterile water is needed to remove the residual chlorine, which may eventually corrode stainless steel surfaces. Wiping with non-sterile water may recontaminate cabinet surfaces, a critical issue when sterility is essential (e.g., maintenance of cell cultures).
* Similarly, the surfaces of all materials and containers placed into the cabinet should be wiped with 70% EtOH to reduce the introduction of contaminants to the cabinet environment.

1. **Additional Techniques:**

* A solid front, back-closing laboratory gown provides better protection of personal clothing than a traditional laboratory coat and may be required at BSL-3.
* When the user’s arms rest flatly across the front grille, the grille opening will be occluded. Room air laden with particles may flow directly into the work area, rather than being drawn down through the front grille. Raising the arms slightly will alleviate this problem.
* BSCs are designed for 24-hour per day operation, and some investigators find that continuous operation helps to control the laboratory’s level of dust and other airborne particulates. Although energy conservation may suggest BSC operation only when needed, especially if the cabinet is not used routinely, room air balance is an overriding consideration. Air discharged through ducted BSCs must be considered in the overall air balance of the laboratory.
* Materials or equipment placed inside the cabinet may cause disruption of the airflow, resulting in turbulence, possible cross-contamination and/or breach of containment. Extra supplies (e.g., additional gloves, culture plates or flasks, culture media) should be stored outside the cabinet.
* The biohazard collection bag should not be taped to the outside of the cabinet The frequent inward/outward movement needed to place objects in these containers is disruptive to the integrity of the cabinet air barrier and can compromise both personnel and product protection.
* Upright, or vertical, pipette collection containers should not be used in BSCs, nor placed on the floor outside the cabinet. The frequent inward/outward movement needed to place objects in these containers is disruptive to the integrity of the cabinet air barrier and can compromise both personnel and product protection.
* Keeping clean materials at least one foot (30.48cm or .3 meters) away from aerosol-generating activities will minimize the potential for cross-contamination.