VIII. Biosafety Laboratory Practices and Equipment

All laboratory personnel shall engage in good microbiological laboratory practices at all times. The following practices incorporate minimal practices and provide guidance for ensuring the protection of personnel, research, and the environment for the level of containment used in university academic and research laboratories.

Hands should be washed frequently during the day. Wash hands after removing gloves, before leaving the laboratory, before and after contact with patients or animals, and before eating, smoking, handling contacts or applying cosmetics.

Hands must also be washed immediately after accidental contact with blood, body fluids, and contaminated materials. Refrigerators, freezers, water baths, and centrifuges should be cleaned and disinfected periodically (the frequency to be established by the PI/laboratory director) and when gross contamination occurs. Wear gloves, gown, and appropriate PPE during cleaning.

Exits and aisles must not be obstructed in any way. No trash, supplies, equipment, or furniture should be permitted in exit routes or aisles.

Exit doors must not be obstructed, bolted, or blocked in any way. Smoke doors must not be obstructed in any way that prevents automatic closing in case of fire.

Do not cover or block access to fire extinguishers, fire alarm boxes, emergency blankets, safety showers, eyewashes or exits at any time, for any reason.

All hazardous materials, including biological, chemical and radioactive materials, should be secured when unattended, to protect from unauthorized access, misuse or removal.
VIII.1. Biosafety Level 1 (BSL-1)

**Biosafety Level 1** is suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment. The laboratory is not necessarily separated from the general traffic patterns in the building. Work is generally conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is neither required nor generally used. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by a scientist with general training in microbiology or a related science.

The following standard and special practices, safety equipment and facilities apply to agents assigned to Biosafety Level 1:

**VIII.1.1. BSL-1 Standard Microbiological Practices**

1. Access to the laboratory is limited or restricted at the discretion of the laboratory director when experiments or work with cultures and specimens are in progress.

2. Persons wash their hands after they handle viable materials, after removing gloves, and before leaving the laboratory.

3. Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human use are not permitted in the laboratory. Food is stored outside the laboratory in cabinets or refrigerators designated and used for this purpose only.

4. Mouth pipetting is prohibited; mechanical pipetting devices are used.

5. Policies for the safe handling of sharps are instituted.

6. All procedures are performed carefully to minimize the
creation of splashes or aerosols.

7. Work surfaces are decontaminated at least once a day and after any spill of viable material.

8. All cultures, stocks, and other regulated wastes are decontaminated before disposal by an approved decontamination method such as autoclaving. Materials to be decontaminated outside of the immediate laboratory are to be placed in a durable, leak-proof container and closed for transport from the laboratory. Materials to be decontaminated outside of the immediate laboratory are packaged in accordance with applicable local, state, and federal regulations before removal from the facility.

9. A biohazard sign must be posted at the entrance to the laboratory whenever infectious agents are present. The sign may include the name of the agent(s) in use and the name and phone number of the investigator.

10. An insect and rodent control program is in effect.

**VIII.1.2. BSL-1 Special Practices**

None

**VIII.1.3. BSL-1 Safety Equipment (Primary Barriers)**

1. Special containment devices or equipment such as a biological safety cabinet, are generally not required for manipulations of agents assigned to Biosafety Level 1. However, if BSCs are used (i.e. tissue culture) they must be tested and certified annually.

2. It is recommended that laboratory coats, gowns, or uniforms be worn to prevent contamination or soiling of street clothes.

3. Gloves should be worn if the skin on the hands is broken or if a rash is present. Alternatives to latex gloves should be
available (i.e. nitrile gloves).

4. Protective eyewear should be worn to conduct procedures in which splashes of microorganisms or other hazardous materials is anticipated. Persons who wear contact lenses in laboratories should also eye protection.

VIII.1.4. BSL-1 Laboratory Facilities (Secondary Barriers)

1. Laboratories should have doors for access control.

2. Each laboratory contains a sink for hand washing.

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

4. Bench tops are impervious to water and are resistant to moderate heat and the organic solvents, acids, alkalis, and chemicals used to decontaminate the work surface and equipment.

5. Laboratory furniture is capable of supporting anticipated loading and uses. Spaces between benches, cabinets, and equipment are accessible for cleaning.

6. If the laboratory has windows that open to the exterior, they are fitted with fly screens.

VIII.2. Biosafety Level 2 (BSL-2)

Biosafety Level 2 is similar to Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It differs from BSL-1 in that (1) laboratory personnel have specific training in handling pathogenic agents and are directed by competent scientists; (2) access to the laboratory is limited when work is being conducted; (3) extreme precautions are taken with contaminated sharp items; and (4) certain procedures in
which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment.

The following standard and special practices, safety equipment, and facilities apply to agents assigned to Biosafety Level 2:

**VIII.2.1. BSL-2 Standard Microbiological Practices**

1. The laboratory supervisor must enforce the institutional policies that control access to the laboratory. Laboratory doors should remain closed when working with BSL2 materials.

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 in cabinets or refrigerators designated and used for this purpose only.

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.

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 an appropriate disinfectant.

8. An effective integrated pest management program is required.
VIII.2.2. BSL-2 Special Practices

1. 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.

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

3. 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), emergency telephone number, and required procedures for entering and exiting the laboratory.

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

5. Institutional policies and procedures describing the collection and storage of serum samples from at-risk personnel must be followed.

6. A laboratory-specific biosafety manual/research protocol must be prepared and adopted as policy. The manual must be available and accessible.

7. The laboratory director ensures that laboratory and support 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.

8. 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 the risk of sharps injuries.

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

a. Careful management of needles and other sharps are of primary importance.

b. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes or otherwise manipulated by hand before disposal. If recapping is deemed necessary, Standard Operating Procedures shall be followed for using recapping sheaths or the one-handed method only.

c. Used disposable needles and syringes must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal. Non-disposable sharps must be placed in a hard-walled container for transport to a processing area for decontamination, preferably by autoclaving.

e. Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs or forceps.

f. Plasticware should be substituted for glassware whenever possible.

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

10. Laboratory equipment should be routinely decontaminated, as well as, after spills, splashes or other
potential contamination. Spills involving infectious materials must be contained, decontaminated and cleaned up by staff properly trained and equipped to work with infectious material.

11. 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.

12. Animals and plants not associated with the work being performed are not permitted in the laboratory.

VIII.2.3. BSL-2 Safety Equipment (Primary Barriers)

1. Properly maintained biological safety cabinets (preferably Class II), other appropriate personal protective equipment or physical containment devices are used whenever:

   a. 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.

   b. 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. 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 manipulated outside the BSC or containment device.

3. Protective laboratory coats, gowns, smocks, or uniforms designated for lab 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. PPE should not be taken home by personnel for laundering. If sent off-site for laundering, PPE should be properly bagged and the laundry facility notified of any potential hazards.

4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Wear two pairs of gloves, when appropriate. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory. Alternatives to latex gloves should be available.

VIII.2.4. BSL-2 Laboratory Facilities (Secondary Barriers)

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

2. Each laboratory contains a sink for hand washing. The sink may be manually, hands-free or automatically operated and located near the exit door.

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

4. Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.

5. Laboratory furniture must be capable of supporting anticipated loads and uses. Spaces between benches, cabinets, and equipment should be accessible for cleaning. Chairs used in laboratory work must be covered with a non-porous material that can easily be cleaned and decontaminated with an appropriate disinfectant.

6. BSCs must be installed so that fluctuations of the room air
supply and exhaust air 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 (i.e. supply air diffusers).

7. An eyewash station must be readily available.

8. Vacuum lines should be protected with an in-line filter. Liquid disinfectant traps may be required, and if glass traps are used, must be stored in appropriate secondary containment.

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 the manufacturer’s recommendations. BSCs can also be connected to the laboratory exhaust system by either a thimble (canopy) connection or a direct (hard) connection, depending on the type of BSC.

11. A method for decontaminating all laboratory wastes should be available in the facility. At OSU, all infectious waste, including liquid infectious waste, must be collected and disposed of in a biohazard box and sent off-site for incineration.

VIII.3. Work with Human or Animal Tissues

Human blood, blood products, cells and cell lines, body fluids and tissues are listed as potentially hazardous biological materials. Animal tissues may also be contaminated with biohazardous materials. Biosafety Level 2 practices and procedures must be followed when handling human blood, blood products, cells and cell lines (including established cell lines), body fluids and tissues and animal tissues because of
the infectious agents that they may contain. Biosafety Level 2 practices and procedures are consistent with the concept of “Universal Precautions” that require all specimens of human blood, blood products, body fluids and tissues to be treated as if they are infectious. The federal regulation, the Occupational Exposure to Bloodborne Pathogens (29 CFR 1910.1030), adopted by the Ohio Public Employment Risk Reduction Program, mandates a series of engineering and work practice controls, training, and Hepatitis B vaccination to control the health risk to employees resulting from occupational exposure to human blood and other potentially infectious materials that may contain human pathogens.

VIII.4. Biosafety Level 3 (BSL-3)

A complete description of Biosafety Level 3 can be found in the BMBL 5th edition. Specific BSL3 practices and procedures for work at the university are described in the BSL3 facility safety manuals.