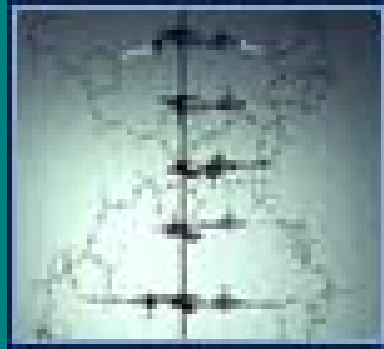


Biological Safety



Temple University

Environmental Health and Radiation Safety

<http://www.temple.edu/ovpr/ehrs/index.html>

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What is Biosafety?

Biosafety is simply the use of procedures, equipment and building design to ensure that workers and the community are protected from Biohazards.

Reference Materials

Biosafety in Microbiological and Biomedical Laboratories, (a.k.a., BMBL) current edition

– The BMBL on-line:

<http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm>

– For an explanation of BioSafety Cabinets,

<http://www4.od.nih.gov/oba/rac/guidelines/guidelines.html>

Goals

- Understand the differences between the 4 BioSafety levels
- Understand the difference between Primary and Secondary Barriers
- Apply your knowledge to a Biosafety Lab.

Primary Barriers

Primary Barriers refers to safety equipment

- Biological safety cabinets (BSCs), enclosed containers, and other engineering controls designed to remove or minimize exposures to hazardous biological materials.
- BSC is the principal device used to provide containment of infectious splashes or aerosols generated by many microbiological procedures
- There are three types of biological safety cabinets (Class I, II, III) used in microbiological laboratories

Secondary Barriers

Secondary Barriers relate to the physical facility design and construction.

- The design and construction of the facility contributes to the laboratory workers' protection, provides a barrier to protect persons outside the laboratory, and protects persons or animals in the community from infectious agents which may be accidentally released from the laboratory.
- The recommended secondary barrier(s) will depend on the risk of transmission of specific agents.
- When the risk of infection by exposure is present, higher levels of primary containment and multiple secondary barriers (specialized ventilation systems) may become necessary

Laboratory Practice and Technique

The most important element of containment is strict adherence to standard microbiological practices and techniques. The term "containment" is used in describing safe methods for managing infectious materials in the laboratory environment where they are being handled or maintained.

Laboratory Practice and Technique

Persons working with infectious agents or potentially infected materials must be aware of potential hazards, and must be trained and proficient in the practices and techniques required to handle such material safely. The director or person in charge of the laboratory is responsible for providing or arranging the appropriate training of personnel.

Laboratory Practice and Technique

Each laboratory should develop or adopt a **Biosafety Manual** that identifies the hazards that will or may be encountered, and that specifies practices and procedures designed to minimize or eliminate exposures to these hazards. Personnel should be advised of special hazards and should be required to read and follow the required practices and procedures.

Laboratory Practice and Technique

A scientist trained and knowledgeable in appropriate laboratory techniques, safety procedures, and hazards associated with handling infectious agents must be responsible for the conduct of work with any infectious agents or material. This individual should consult with Biosafety or other health and safety professionals with regard to risk assessment.

Biosafety Manual

The **Biosafety Manual** provides university and health system-wide safety guidelines, policies and procedures for the use and manipulation of biohazards. Although the implementation of these procedures is the responsibility of the Principal Investigator (PI), its success depends largely on the combined efforts of laboratory supervisors and employees. Planning for and implementation of biological safety must be part of every laboratory activity in which Biohazardous materials are used.

Biosafety Manual

In general, the handling and manipulation of biological agents and toxins, as well as recombinant DNA molecules, requires the use of various precautionary measures depending on the material(s) involved. This manual will provide assistance in the evaluation, containment and control of biohazards. However, it is imperative that all parties involved or working with these materials seek additional advice and training when necessary.

Standard Microbiological Practices

- Access to the laboratory is limited or restricted at the discretion of the PI when experiments are in progress.
- Persons wash their hands after handling infectious materials, after removing gloves, and when they leave the laboratory.
- Eating, drinking, smoking, handling contact lenses, and applying cosmetics are not permitted in the laboratory. Persons who wear contact lenses in laboratories should also wear goggles or a face shield. Food is stored outside the work area in cabinets or refrigerators designated for this purpose only.
- Mouth pipetting is prohibited; mechanical pipetting devices are used.
- Shorts, sandals, and open toe shoes are prohibited.

Standard Microbiological Practices

- Policies for the safe handling of sharps are instituted.
- All procedures are performed carefully to minimize the creation of aerosols.
- Work surfaces are decontaminated at least once a day and after any spill of viable material.
- 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 placed in a durable, leak proof container and closed for transport from the laboratory. Infectious waste from BSL-3 laboratories should be decontaminated before removal for off-site disposal.
- An insect and rodent control program is in effect (can be found in appendix G of the BMBL).

Biosafety Levels

Four Biosafety levels (BSL) are described in Section III, which consist of combinations of laboratory practices and techniques, safety equipment, and laboratory facilities. Each combination is specifically appropriate for the operations performed, the documented or suspected routes of transmission of the infectious agents, and the laboratory function or activity.

Biosafety Levels

- **BSL 1: Not known to consistently cause disease in health adults**
- **BSL 2: Associated with human disease, primarily via percutaneous injury, ingestion, mucous membrane exposure**
- **BSL 3: Indigenous or exotic agents with potential for aerosol transmission: disease has serious or lethal consequences.**
- **BSL 4: Dangerous/exotic agents which pose high risk of life-threatening diseases, aerosol-transmitted lab infections; or related agents with unknown risk of transmission.**

BSL 1 (We have lots!)

Biosafety Level 1 represents a basic level of containment that relies on standard microbiological practices with no special primary or secondary barriers recommended, other than a sink for hand washing.

BSL 1

Typical agents: E. coli,
Saccharomyces pombe

Procedures (Standard microbial practices)

Special Practices

- None

BSL 1

Primary Barriers

- Personal Protective Clothing (Lab coat and gloves)
- Additional maybe needed: Face/Eye Protection

Secondary Barriers

- Sink for hand washing.
- Work surface easily cleaned.
- Bench tops
- Sturdy lab furniture

BSL 2 (We have quite a few)

Biosafety Level 2 is appropriate when work is done with any human-derived blood, body fluids, tissues, or primary human cell lines where the presence of an infectious agent may be unknown. (Laboratory personnel working with human-derived materials should refer to the OSHA *Bloodborne Pathogen Standard* for specific required precautions.)

BSL 2

Agents: HIV, Hepatitis B

Procedures (Standard microbial practices)

BSL 1 with emphasis on....

- Mechanical pipetting
- Limited Access
- Biohazard Warning Signs
- Safe handling of sharps
- Biosafety Manual specific to the lab

BSL 2

Special Practices

- Access to the laboratory is limited or restricted by the PI when work with infectious agents is in progress.
- PI establishes policies and procedures whereby only persons who have been advised of the potential hazards and meet specific entry requirements (e.g., immunization) may enter the laboratory.
- Biohazard sign must be posted on the entrance to the laboratory when etiologic agents are in use

BSL 2

Special Practices (cont.)

- Laboratory personnel receive appropriate immunizations or tests for the agents handled or potentially present in the laboratory (e.g., hepatitis B vaccine or TB skin testing).
- Biosafety manual adopted or prepared specifically for the laboratory by the laboratory director
- PI ensures that laboratory and support personnel receive appropriate training on the potential hazards associated with the work involved

BSL 2

Special Practices (cont.)

- A high degree of precaution must always be taken with any contaminated sharp items, including needles and syringes, slides, pipettes, capillary tubes, and scalpels.
- Syringes which re-sheathe the needle, Needleless systems, and other safety devices are used when appropriate
- Broken glassware must not be handled directly by hand, but must be removed by mechanical means such as a brush and dustpan, tongs, or forceps.

BSL 2

Special Practices (cont.)

- Skin contamination need to be immediately reported to the **PI and a Medical Physician:** Occupational and Employee Health Services (OEHS) during working hours 8:30 am to 4:30 pm. After hours, weekends or holidays consult Temple University Hospital Emergency Department (ED).

BSL 2

Primary Barriers

- Properly maintained biological safety cabinets, preferably Class II
- Face protection (goggles, mask, face shield or other splatter guard)
- Protective laboratory coats, gowns
- Gloves

Secondary Barriers

- Provide lockable doors for facilities that house restricted agents (as defined in 42 CFR 72.6).
- Sink for hand washing
- An eyewash station is readily available
- Illumination is adequate for all activities, avoiding reflections and glare that could impede vision.

BSL 3 (We have a few)

Biosafety Level 3, more emphasis is placed on primary and secondary barriers to protect personnel in contiguous areas, the community, and the environment from exposure to potentially infectious aerosols.

BSL 3

Typical agents: Mycobacterium tuberculosis,
Listeria monocytogenes

Procedures (Standard microbial practices)
BSL 1 and 2 with emphasis on....

- Baseline serum samples
- Warning Signs
- Decon of all wastes
- Decon of lab clothing before laundering

BSL 3

Special Practices

BSL 1 and 2 with emphasis on....

- When infectious materials or infected animals are present in the laboratory or containment module, a hazard warning sign, incorporating the universal biohazard symbol, is posted on all laboratory and animal room access doors. The hazard warning sign identifies the agent, lists the name and telephone number of the laboratory director or other responsible person(s), and indicates any special requirements for entering the laboratory, such as the need for immunizations, respirators, or other personal protective measures.

BSL 3

Special Practices (cont.)

- Baseline serum samples are collected as appropriate and stored for all laboratory and other at-risk personnel. Additional serum specimens may be periodically collected, depending on the agents handled or the function of the laboratory.
- All open manipulations involving infectious materials are conducted in biological safety cabinets or other physical containment devices within the containment module. No work in open vessels is conducted on the open bench.

BSL 3

Primary Barriers

- Class II or Class III biological safety cabinet.
- When a procedure or process cannot be conducted within a biological safety cabinet, then appropriate combinations of personal protective equipment (e.g., respirators, face shields) and physical containment devices (e.g., centrifuge safety cups or sealed rotors) are used.

Note: Contact EHRS about the Respiratory Protection Program

- Protective laboratory clothing such as solid-front or wrap-around gowns, scrub suits, or coveralls are worn by workers when in the laboratory.

BSL 3

Primary Barriers (cont.)

- Protective clothing is not worn outside the laboratory.
- Reusable clothing is decontaminated before being laundered. Clothing is changed when overtly contaminated.
- Gloves {Frequent changing of gloves accompanied by hand washing is recommended. Disposable gloves are not reused}

BSL 3

Secondary Barriers

- The laboratory is separated from areas that are open to unrestricted traffic flow within the building, and access to the laboratory is restricted.
- Passage through a series of two self-closing doors.
- Doors are lockable (Outlined in Appendix F of the BMBL)
- All windows in the laboratory are closed and sealed.
- A method for decontaminating all laboratory wastes is available in the facility and utilized, preferably within the laboratory (i.e., autoclave, chemical disinfection, incineration, or other approved decontamination method).

BSL 3

Secondary Barriers (cont.)

- A ducted exhaust air ventilation system is provided (The exhaust air is not recirculated to any other area of the building)
- An eyewash station is readily available inside the laboratory.
- Illumination is adequate for all activities, avoiding reflections and glare that could impede vision.
- The Biosafety Level 3 facility design and operational procedures must be documented. The facility must be tested for verification that the design and operational parameters have been met prior to operation

BSL 4 (Relax, you wont see this one at Temple!)

Biosafety Level 4 agents are respiratory exposure to infectious aerosols, mucous membrane or broken skin exposure to infectious droplets, and autoinoculation. All manipulations of potentially infectious diagnostic materials, isolates, and naturally or experimentally infected animals, pose a high risk of exposure and infection to laboratory personnel, the community, and the environment.

BSL 4

Typical agents: Marburg virus, Congo-Crimean hemorrhagic fever virus

Procedures (Standard microbial practices)

BSL 3 with emphasis on....

- All procedures are performed carefully to minimize the creation of aerosols
- Work surfaces are decontaminated at least once a day and after any spill of viable material
- All waste is decontaminated before disposal by an approved method such as autoclaving.

BSL-4

Special Practices

BSL 3 with emphasis on.....

- Only persons whose presence in the facility or individual laboratory rooms is required for program or support purposes are authorized to enter.
- Children or pregnant women, are not allowed in the laboratory or animal rooms
- When infectious materials or infected animals are present in the laboratory or animal rooms, hazard warning signs, incorporating the universal biohazard symbol, are posted on all access doors.

BSL-4

Special Practices (cont.)

- The PI is responsible for ensuring that, before working with organisms at Biosafety Level 4, all personnel demonstrate a high proficiency in standard microbiological practices and techniques, and in the special practices and operations specific to the laboratory facility.
- Laboratory personnel receive available immunizations for the agents handled or potentially present in the laboratory.
- Personnel enter and leave the laboratory only through the clothing change and shower rooms.

BSL-4

Special Practices (cont.)

- Personal clothing is removed in the outer clothing change room and kept there
- Decontaminating shower each time they leave the laboratory. Personnel use the airlocks to enter or leave the laboratory only in an emergency.
- Supplies and materials needed in the facility are brought in by way of the double-door autoclave, fumigation chamber, or airlock, which is appropriately decontaminated between each use.
- Other requirements as outlined in BMBL

BSL-4

Primary Barriers

- Class III Biological Safety Cabinet
- Class II biological safety cabinets used in conjunction with one-piece positive pressure personnel suits ventilated by a life support system.

Secondary Barriers

There are two models for Biosafety Level 4 laboratories: *Cabinet Laboratory* and *Suit Laboratory* (Outlined in BMBL)

Next Steps

- Go forth and inspect your labs to make sure they are compliant!
- Document as already discussed.

Current Issues

- Assurance on Hazardous Procedures
- Biosafety Registration Form
- Select Biological Agents
- Shipping of Infectious Substances

Assurance on Hazardous Procedures

- For each new or renewed project, you must tell us what specific hazards you are working with.
- We will check to ensure that you have received the proper training to work safely.
- Approval will not be granted unless safety is assured.

Biosafety Registration

- Users of biohazards or persons doing recombinant DNA work must have prior approval from the Institutional Biosafety Committee.
- Information required includes training, immunizations, persons at risk and handling procedures.

Select Biological Agents

- The Centers for Disease Control and the US Dept. of Agriculture have established a list of select biological agents.
- These are agents of potential use as weapons of terror.
- All users must be registered with the US government and Temple University.

Shipping of Infectious Substances

- Anyone wishing to ship a dangerous good from Temple must first have been trained to do so.
- Materials considered Category A is considered Infectious Substances.
- Contact EHRS for more information.