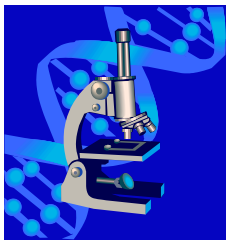
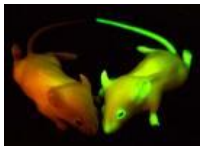
Standard Microbiological Practices for Biosafety Level 1 Laboratories at the University of Tennessee-Knoxville, Institute of Agriculture and Graduate School of Medicine





Overview and Definitions

Standard microbiological practices (SMPs) are generally defined as the basic "hygiene" practices that apply to all labs that manipulate microorganisms or any biological materials that contain microorganisms. SMPs serve to minimize the spread of contamination generated through lab processes and to protect both personnel and the environment. As such, they are often cited by regulatory and granting agencies such as NIH, CDC, OSHA, and USDA APHIS as minimum standards to be followed in biological research laboratories. Therefore, SMPs apply to a broad spectrum of lab activities including:

- Manipulation of any microbes including bacteria, viruses, fungi, and protozoa.
- Manipulation of materials that may contain microbes including animal and plant tissues, soil samples, and water samples.
- Receiving, processing, and testing of diagnostic samples.
- Research involving recombinant DNA molecules, transgenic animals, or genetically modified plants.
- Manipulation of animals or plants that are experimentally infected with microbes.
- Work with biological toxins and other bioactive molecules.

Biosafety Level 1 is suitable for work involving well-characterized agents not known to consistently cause disease in immunocompetent adult humans, and otherwise present minimal potential hazard to laboratory personnel and the environment. BSL-1 laboratories are not necessarily separated from the general traffic patterns in the building. Work is typically conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is not required, but may be used as determined by appropriate risk assessment. Laboratory personnel must have specific training in the procedures conducted in the laboratory and must be supervised by a scientist with training in microbiology or a related science.

The following standard practices, safety equipment, and facility requirements apply to BSL-1:

A. *Standard Microbiological Practices*

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

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

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

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

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

    a. 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.

    b. Used disposable needles/syringes and other disposable sharps must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal (i.e., "sharps containers").

    c. Non-disposable sharps must be placed in a hard walled, puncture proof container for transport to a processing area for decontamination, preferably by autoclaving.

    d. Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs, or forceps. Plasticware should be substituted for glassware whenever possible.

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

7. Decontaminate work surfaces after completion of work and after any spill or splash of biological material with an appropriate disinfectant. Follow the manufacturer's instructions regarding dilution ratio, contact time, and shelf life. Finally, wear the proper personal protective equipment for dispensing the disinfectant as recommended by the manufacturer.

8. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method. Generally accepted methods are summarized below (refer to UT's *Biohazardous Waste Basics* for details):

    a. Disposal guidelines:

        - Liquid wastes — All biohazardous liquid waste must be decontaminated by either autoclaving or chemical disinfection. If autoclaving, follow all recommended safety guidelines for loading and unloading liquid wastes. Allow liquids to cool prior to disposal. Alternatively, liquid wastes may be decontaminated using an appropriate disinfectant. Ensure that proper dilution requirements and contact times are met. Do not autoclave liquids containing disinfectant. All decontaminated liquids may be disposed via the lab sink.
        - Solid biohazardous waste (non-sharps) — All solid biohazardous waste must be collected in a solid-walled container equipped with a lid, labeled with the universal biohazard symbol, and lined with a bag. The bag must contain the universal biohazard symbol if the collected wastes include human-derived materials or agents that are an infectious disease risk for humans/animals.

          Full waste bags must be autoclaved using validated autoclave settings and procedures. Place all autoclaved waste bags in a non-see-through bag for disposal to the dumpster (the biohazard symbol must not be visible). Alternatively, waste bags may be removed to a central collection facility for pick-up by a contracted waste hauler (only applicable to some facilities, contact the Biosafety Office for further details).

- Biohazardous sharps — All biologically contaminated objects that are inherently sharp enough to puncture the skin are biohazardous sharps (e.g., needles, scalpel blades, pasteur pipettes, etc.). These must be collected in a solid, puncture-proof biohazardous sharps container marked with the universal biohazard symbol and fitted with a lid that restricts access into the container. Be sure to use the correct sharps container size for the biohazardous sharps being generated. Dispose of all sharps into a designated sharps container immediately after use (no recapping, bending, or breaking).

  Sharps containers are full when they are ¾ full or objects no longer freely fall into the container. All sharps containers must be permanently closed, wiped down with disinfectant, and disposed of through Environmental Health and Safety, the Biosafety Office, or the contracted waste hauler collection facility (contact the Biosafety Office for sharps disposal guidance).

b. Materials to be decontaminated outside of the immediate laboratory must be placed in a durable, leak proof container and secured for transport.

c. Materials to be removed from the facility for decontamination must be packed in accordance with applicable local, state, and federal regulations.

d. Decontamination and disposal of mixed hazardous wastes (i.e., biological waste combined with hazardous chemical and/or radiological waste) can be challenging as these may pose additional risks to personnel and the environment. Furthermore, these wastes have to be managed in accordance with various local, state, and federal disposal regulations. Researchers should avoid generating mixed hazardous waste if at all possible. If this situation is unavoidable, please contact the appropriate Safety Office for guidance on proper collection, decontamination, and disposal:

Radiation Safety Office	974-5580
Environmental Health and Safety Office	974-5084 (UTK/GSM) 974-4904 (UTIA)
Biological Safety Office	974-1938 or 974-9836

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.

10. 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. Please contact the UT Occupational Health Nurse at 974-5728 or aknowles@utk.edu for further information.

B. *Special Practices*

None required.

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

1. Special containment devices or equipment, such as BSCs, are not generally required.

2. Protective laboratory coats, gowns, or uniforms are recommended to prevent contamination of personal clothing.

3. Wear protective eyewear when conducting procedures that have the potential to create splashes of microorganisms or other hazardous materials. Persons who wear contact lenses in laboratories should also wear eye protection.

4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to latex gloves should be available. Wash hands prior to leaving the laboratory. In addition, BSL-1 workers should:

    a. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary.

    b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.

    c. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.

D. *Laboratory Facilities (Secondary Barriers)*

1. Laboratories should have doors for access control.

2. Laboratories must have a sink for hand washing.

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

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

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

    b. Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.

5. Laboratories with windows that open to the exterior should be fitted with screens.

E. *References*

1. *Biosafety in Microbiological and Biomedical Laboratories* (5th ed.). 2007. U.S. Government Printing Office, Washington, D.C. Link: http://www.cdc.gov/OD/ohs/biosfty/bmbl5/BMBL_5th_Edition.pdf.
2. *NIH Guidelines for Research Involving Recombinant DNA Molecules*. 2002. Link: http://www4.od.nih.gov/oba/rac/guidelines_02/NIH_Gdlnes_lnk_2002z.pdf.
3. *OSHA Bloodborne Pathogens Standard 29CFR 1910.1030*. 2001. U.S. Department of Labor, Occupational Safety and Health Administration. Link: http://www.osha.gov/pls/oshaweb/owadisp.show_document?p_table=STANDARDS&p_id=10051.
4. Tennessee Department of Environment and Conservation. Link: TDEC Rule 0400-11-01-.04(2)(k)(4).

Standard Microbiologial Practices Training Record

This document is intended to serve as a written record acknowledging that standard microbiological practices (SMPs) training has been given to the indicated laboratory personnel (including students) by a qualified site-specific trainer (i.e., principal investigator, laboratory manager, or other senior personnel).

SMP training included: general definitions; laboratory hygiene; laboratory access policies; disinfection and decontamination; biohazardous sharps management and safety; biohazardous waste segregation, treatment, and disposal; personal protective equipment; and facility standards. This training is intended to comply with the guidelines issued by NIH, CDC, USDA APHIS, and other applicable regulatory and/or funding agencies.

Trainee Name (Print)	Trainee Signature	Trainer Name (Print)	Trainer Signature	Date of Training