

MICROBIOLOGICAL LABORATORY PRACTICES

Introduction

In accordance with the following referenced sources, certain microbiological practices must be observed in laboratories working with biological agents, including recombinant or synthetic nucleic acids (r/sNA). This SOP summarizes these practices and can be used as a training and information tool and should be included in the laboratory biosafety manual.

This SOP applies to all work at UNL and affiliated campuses that is subject to the UNL Biosafety Guidelines.

References

The information in this SOP is extracted from the following sources:

NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines), National Institutes of Health (current revision)

Biosafety in Microbiological and Biomedical Laboratories, (current ed.) Centers for Disease Control and National Institutes of Health

Bloodborne Pathogens Standard, 29 CFR 1910.1030, Occupational Safety and Health Administration



Note: Work with most biological materials requires submission and subsequent review and approval of an Institutional Biosafety Committee (IBC) protocol. Please see the **UNL Biosafety Guidelines**, available on the EHS website, for additional information.

Laboratory Practices

In general, the following practices must be observed in all microbiological laboratories. Some practices, as indicated, are dependent on the IBC approved containment level for the laboratory.

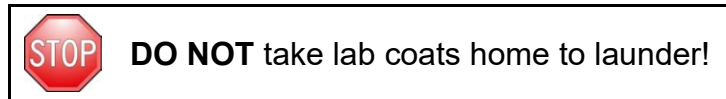
1. A laboratory-specific biosafety manual is available and accessible to laboratory personnel. See EHS SOP, ***Preparing a Laboratory Biosafety Manual***.
2. Laboratory personnel must be appropriately trained. This includes:
 - a. training on the laboratory specific manual;

- b. demonstrating proficiency in microbiological practices (BSL-2 or higher);
- c. appropriate training regarding their duties;
- d. necessary precautions to prevent exposures, including medical conditions that may increase personal susceptibility;
- e. exposure evaluation procedures, and
- f. annual refresher training or additional training when procedural or policy changes occur.

See EHS SOP, ***Biosafety Training*** and the ***Training Needs Assessment*** for more information on available training from EHS.

3. Laboratory access is restricted to authorized personnel. In accordance with UNL policy:
 - a. Laboratory entrances have current and accurate hazard warning placards posted. See the following EHS SOPs:
 - ***Door Postings for Potentially Hazardous Locations***
 - ***Biohazard Door Postings***
 - b. Laboratory doors should be closed while work is being conducted, and locked when not occupied. Biological agent storage areas are secured to prevent unauthorized access.
 - c. If the containment level is BSL-2 or higher:
 - only persons who have been adequately trained, advised of potential hazards, meet specific entry requirements (e.g., vaccination, other medical surveillance or qualification, etc.) as specified in the project specific protocol, and who comply with all entry and exit procedures are allowed to work in the laboratory.
 - access for minors may be restricted or prohibited in accordance with UNL Human Resources policies.
4. Good personal hygiene practices are observed:
 - a. Eating, drinking, handling contact lenses, applying cosmetics, and storing food for human consumption are prohibited in the laboratory.
 - b. Persons wash their hands thoroughly:
 - after handling potentially infectious materials, r/sNA molecules, and/or research organisms;
 - before exiting the laboratory.
 - c. Mechanical pipetting devices are used; mouth pipetting is prohibited.
5. Appropriate Personal Protective Equipment (PPE) is available and used.
 - a. Laboratory-specific PPE selection considers and provides protection against all potential hazards (chemical, biological, radiation, etc.). For additional information, refer to the following EHS SOPs:

- **Personal Protective Equipment – Body Protection**
 - **Personal Protective Equipment – Eyes and Face Protection**
 - **Personal Protective Equipment – Hand Protection**
- b. Laboratory coats, gowns, smocks, etc., are worn while in the laboratory, and removed before exiting the laboratory. It is recommended that lab coats with knit cuffs be used for work in a biological safety cabinet with microorganisms to prevent contaminated air from being blown up the sleeves of loose-cuff lab coats. Dispose of protective clothing appropriately. Re-usable lab coats/gowns should be laundered on a regular basis, some Departments have laundry facilities on campus or a commercial laundry service can be utilized to launder the coats.



- c. Appropriate protective gloves are used when contact with r/sNA, infectious materials, and/or research organisms occur. Gloves are changed when contaminated, integrity has been compromised, or when otherwise necessary. Disposable gloves are not reused and are managed as potentially contaminated laboratory waste.
- d. For HIV and HBV laboratories, solid front or wrap-around gowns, scrub suits, or coveralls are worn in the laboratory.
- e. Eye protection is used. Goggles may be required if there is substantial risk of splashes and/or aerosols. Additional PPE may be required for special tasks or additional hazards (e.g., chemical, radiation, etc.). Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse, as necessary.
- f. PPE is never worn outside laboratory areas, and it is decontaminated prior to laundering and/or disposal, as necessary.
- g. Observe good personal hygiene habits and wash hands thoroughly after removing PPE and before leaving the lab.
6. Disinfection and decontamination.
- a. Using an approved chemical disinfectant, work surfaces are decontaminated at the completion of each day's work session and after any spill of potentially infectious materials. See EHS SOP, **Chemical Disinfectant for Biohazardous Materials**.
- b. Laboratory equipment is routinely decontaminated, as well as after spills, splashes, or other potential contamination. Equipment is decontaminated before repair, maintenance, or removal from the laboratory. See EHS SOP, **Biological Decontamination of Laboratory Equipment**.
- c. An emergency plan must be developed and include response to spills involving biological agents. Such spills must be promptly contained, decontaminated, and cleaned up. Laboratory staff must be trained on the plan, competent to clean-up

- spills, and have appropriate spill clean-up materials readily available. See EHS SOP, ***Spill and Exposure Response for Biohazardous Materials (including Recombinant or Synthetic Nucleic Acids)***.
- d. All contaminated 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 a durable leak-proof, sealable container that is closed before being removed from the laboratory. See EHS SOP, ***Autoclave Operation and Use***.
7. All procedures are performed to minimize the creation of splashes and aerosols. Certified biosafety cabinets are an effective engineering control to minimize exposure and contamination from aerosols. For HIV and HBV production or research labs, **all** work with other potentially infectious material will be conducted in a Class II biosafety cabinet or other primary containment device (not on the open bench).
 8. Use of sharps (e.g., needles, scalpels, pipettes, glassware) is minimized when practical. Plastic-ware is substituted for glassware whenever possible. See the following EHS SOPs: ***Sharps - Handling and Disposing*** and ***Glass Disposal – Intact or Broken*** for further guidance.
 - a. When absolutely necessary, the following general precautions are taken.
 - Needles are not bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
 - Used sharps are carefully placed in puncture-resistant containers.
 - Broken glassware is not handled directly. Instead, it is handled using mechanical assistance (e.g., brush and dustpan, tongs, forceps).
 - Effective hand protection is used (e.g., molded guards, use of forceps when discarding scalpels, etc.).
 - b. If the containment level is BSL-2 or higher, hypodermic needles and syringes are used only for parenteral injection and aspiration of fluids from laboratory animals and diaphragm bottles. Extreme caution is used when handling needles and syringes to avoid autoinoculation and the generation of aerosols during use and disposal.
 9. For all containment levels, emergency action procedures are developed that include immediate reporting of spills and accidents that result in overt exposures to recombinant nucleic acid-containing materials or pathogenic agents to the Principal Investigator (PI) and EHS. Medical evaluation is sought immediately for exposures to pathogenic agents. See EHS SOP, ***On the Job and Student Injuries***. In some cases, reporting to regulatory authorities may be required. See EHS SOP, ***Incident Reporting – National Institutes of Health (NIH) Guidance***.
 10. An effective rodent and insect control program is observed. Signs of infestation are immediately reported to the Principal Investigator (PI) and action taken to impede incursion. If the containment level is BSL-2, animals and plants unrelated to the work are not permitted in the work area.



11. For HIV and HBV research and/or production laboratories, vacuum lines are protected with high efficiency particulate air (HEPA) filters **and** liquid disinfection traps, which are checked routinely and maintained or replaced as necessary.
12. Personnel are advised that personal health status may impact an individual's susceptibility to infection, and their ability to receive immunizations or prophylactic interventions. Personnel are encouraged to self-report conditions that may impact their immune competence or predispose them to infection so that they may seek counseling and guidance from UNL's healthcare provider