UNIVERSITY OF ROCHESTER
ENVIRONMENTAL HEALTH & SAFETY

Policy No.: BS017	**Approved by: UR IBC**
Title: Emergency Procedures for Biological Spills in BSL1, BSL2 and BSL2+ Labs	**Date: December 29, 2021**
Revision No.: 4	**Page 1 of 11**
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I. PURPOSE

The NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules requires each Institutional Biosafety Committee to adopt emergency plans covering accidental spills and personnel contamination resulting from recombinant or synthetic nucleic acid molecule research.

OSHA's Bloodborne Pathogens standard requires immediate and appropriate cleanup, by trained staff, of spills in HIV and HBV research labs. CDC/NIH's Biosafety in Microbiological and Biomedical Laboratories requires the same for infectious materials in BSL2 labs.

This procedure establishes the minimum requirements for responding to biological spills (recombinant and non-recombinant) in labs working at Biosafety Level 1 (BSL1), BSL2 and/or BSL2+.

II. PERSONNEL AFFECTED

University of Rochester research and clinical lab personnel working at BSL1, BSL2 and/or BSL2+

Environmental Health and Safety Spill Team

University Biosafety Officer

III. DEFINITIONS

BSC: Class II biological safety cabinet

Biosafety Level 1 (BSL1) is assigned to work involving well-characterized agents 1) not known to consistently cause disease in immunocompetent adult humans, and 2) present minimal potential hazard to laboratory personnel and the environment. Examples: Adeno-associated virus (and vectors, if delivering non-toxic or non-oncogenic inserts), plasmid DNA administered in vivo, nonpathogenic *E. coli* strains (e.g. K-12 derived strains like DH5alpha, and some non-K-12 derived *E. coli* strains like BL21).

Biosafety Level 2 (BSL2) is assigned to work involving human pathogens that pose moderate hazards to personnel and the environment. This does include agents that can cause fatal disease, but many times, vaccines or treatments are available.

Biosafety Level 2 +: Biosafety Level 2 plus additional precautions required by the IBC

CDC: Centers for Disease Control and Prevention

EH&S: Environmental Health and Safety

Emergency Flip Chart: Also known as the Emergency 13 flip chart is a quick reference guide to handling emergencies at the UR, including a section on Personal Injury/ Exposure; copies are available from EH&S

Engineering Control: A term used by the Occupational Safety and Health Administration (OSHA) and the National Institute for Occupational Safety and Health (NIOSH); engineering controls protect workers by removing hazardous conditions or by placing a barrier between the worker and the hazard. In OSHA's hierarchy of controls, engineering controls should be used before Administrative (work practice) Controls and Personal Protective Equipment.

Higher Risk Biological Spills (for the purposes of this SOP): BSL2+ outside of primary containment

Incident Report: University of Rochester online Employee Incident Reporting System for work-related employee injuries and illnesses

Lower Risk Biological Spills (for the purposes of this SOP):
- All BSL1 spills
- Human blood, body fluids, tissues, cells/cell lines outside of primary containment

Moderate Risk Biological Spills (for the purposes of this SOP): BSL2 microorganisms outside of primary containment

NIH OSP: National Institutes of Health Office of Science Policy, the branch of NIH that administers the NIH Guidelines

NIH Guidelines: NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules

OSHA: Occupational Safety and Health Administration

Primary Containment: CDC and NIH follow OSHA's hierarchy of controls, and add the "box within a box" concept of Primary and Secondary Barriers. Engineering controls are Primary Barriers that protect lab personnel. Primary barriers/containment equipment generally rely on HEPA filters and secure closures (o-ring), and include biological safety cabinets, centrifuge safety cups, and other aerosol containment.

UR IBC: University of Rochester Institutional Biosafety Committee, composed of faculty, staff, and community members; required by the NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules

IV. RESPONSIBILITIES

It is the responsibility of the Principal Investigator and laboratory supervisor to ensure that:

- An appropriate spill response plan for their lab has been developed using these generic procedures as a basis,
- The spill plan for their lab is adequate for the agents possessed and the type of techniques / experiments conducted with those agents
- Each individual in their lab is familiar with it, and
- An appropriate disinfectant, personal protective equipment, and waste containers are readily available.

The NIH Guidelines require that all obvious exposures to organisms containing recombinant or synthetic nucleic acid molecules that occur in BSL2 labs are immediately reported to the Institutional Biosafety Committee and NIH OSP. Personnel must submit an Incident Report (employees) or call EH&S (non-employees).

The University Biosafety Officer approves spill procedures for BSL2+ labs.

V. PROCEDURES

A. Risk Assessment:

It is important to understand the risks and transmission routes of each biological possessed by the lab. In accordance with federal requirements (NIH Guidelines, CDC/NIH), all labs working at BSL2 and above must have a Biosafety Manual. UR's IBC requires that the manual include a section for biological agent overview, transmission routes, clinical signs, strains resistant to treatment, etc., and reference options include, but are not limited to:

- Research articles
- www.cdc.gov
- Carroll KC et al. editor, Manual of Clinical Microbiology, 12th edition, 2019, American Society for Microbiology, https://www.urmc.rochester.edu/libraries/miner/mdl.aspx?redirect=2000000320 (only UR personnel can obtain full access to this text using this link)
- Public Health Agency of Canada Safety Data Sheets https://www.canada.ca/en/public-health/services/laboratory-biosafety-biosecurity/pathogen-safety-data-sheets-risk-assessment.html

When assessing a spill to determine what type of response is necessary, the following should be considered:

1. What was spilled? [e.g. physical characteristics of the spilled material (e.g. liquid vs. solid), potential hazards of particular organism]

2. How much was spilled? (e.g. volume and concentration of the organism)

3. Where is the spill? (e.g. in a BSC, in a centrifuge, in the lab, outside the lab)

4. What is the potential for release outside the lab?

Lower Risk Biological Spills (for the purposes of this SOP):
- All BSL1 spills
- Human blood, body fluids, tissues, cells/cell lines outside of primary containment

Moderate Risk Biological Spills (for the purposes of this SOP): BSL2 microorganisms outside of primary containment

Higher Risk Biological Spills (for the purposes of this SOP): BSL2+ outside of primary containment

B. General Procedure for Biological Spills

For a template of the General Procedure for Biological Spills, contact EH&S.

1. Try not to breathe (for aerosols) as you step back from the spill. Once you're at a distance you think will have fewer aerosols, alert people in the immediate area of the spill and evacuate. If spill is in a lab, ensure the doors are closed.

- For BSL2 and BSL2+ spills
  - Do not re-enter the area for at least 30 minutes. This allows aerosols to settle and to be exhausted by the building ventilation system.
  - Post a "Biohazard Spill – Do Not Enter" sign on doors to keep all unnecessary people out of the area
    - Hint: Keep one in your spill kit along with a magnetic clip for the door; print on red paper or affix a biohazard sticker, laminate).

2. Call Public Safety by dialing 13 from a campus phone or 275-3333 from a cell phone if needed to:

a. Limit access to the area, if the spill is in a hallway or running under the door.

b. Respond to injuries that require emergency personnel. Public Safety will notify emergency personnel and help direct emergency personnel to your location. This is the fastest way to get help!
- UR's Medical Emergency Response Team (MERT) for the Medical Center
- Paramedics for River Campus

c. Get help for larger spills from EH&S's Spill Team

3. Remove contaminated clothing.

4. For exposures, immediately wash, call, report:

a. Wash
- Intact skin - wash with soap and water.

- Non-intact skin and needlesticks/scalpel cuts - wash with soap and water.
- Intra-oral exposure - spit and rinse the mouth with water.
- Eyes – use the lab's eyewash. (Note: Remove contact lenses first. After rinsing eyes, disinfect contacts per manufacturer's recommendation.)

b. Call

- Blood Exposure Hotline (275-1164) for exposures to human blood, body fluids, tissues, or cells/cell lines (or HIV)
- For all other exposures:
  - During office hours - University Health Service (MC 1-5000, 275-2662)
  - After hours - Emergency Department

c. Report

- Contact his/her supervisor
- Submit an Incident Report (www.safety.rochester.edu/SMH115.html)

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5. Contain the spill by placing an absorbent material such as paper towels over the area involved.

6. A properly trained employee performs clean-up and decontamination

a. Wear appropriate personal protective equipment to prevent human blood, other potentially infectious materials, or microorganisms from reaching work clothes, street clothes, undergarments, skin, eyes, mouth, or other mucous membranes. Appropriate personal protective equipment includes:

- double gloves (BSL2, BSL2+) or single gloves (BSL1)
- lab coat
- face protection
  - safety glasses or goggles (BSL1)
  - chin-length face shield (BSL2, BSL2+)
  - surgical mask
- shoe covers/booties (dependent on size of spill, splatter, etc.)

b. In a circular motion from the edges to the center, gently pour or spray freshly prepared 10% solution of household bleach (1 part bleach, 9 parts water) or the disinfectant listed in the lab's IBC LAB form to completely cover the spill. Avoid splashing or splattering of biological material.

c. Allow the disinfectant to be in contact with the biological for at least 30 minutes.

d. Pick up any broken glass or sharps by mechanical means, such as tongs or a broom and dustpan. This debris can then be deposited into a sharps disposal container. Never pick up sharps directly by hand.

e. Working from the edges to the center, wipe up the spill with absorbent cloth or paper towels. Tongs can be used instead of hands.

1) Re-clean area with fresh paper towels soaked with disinfectant.
2) If the spill was on or in equipment and bleach was used, rinse thoroughly with water to remove the bleach (corrosive to stainless steel).

f. Discard disposable cloths/towels and disposable protective equipment into a biohazard/medical waste bin (red bag).

g. Remove non-disposable PPE.

h. Wash hands with soap and water.

i. If the spill was on the floor, contact Environmental Services to have the area cleaned with regular detergent-disinfectant. Eastman Dental Center employees are to call the Maintenance Department at 275-5070 to have their area cleaned with regular detergent disinfectant.

<u>Visual demonstrations for how to clean a biohazard spill:</u>

Biological Spill Cleanup (Rice University)
https://www.youtube.com/watch?v=Dkp_hFEq_1c (3 minutes)
- Notes: recommend wearing a face shield, and use a red waste bin

Safe Biological Spill Response (Texas Tech)
https://www.youtube.com/watch?v=VKIBNz4fHoU (5 minutes)
- Notes: video recommends wearing a face shield, use a red waste bin, and do not autoclave bleached material in a lab-size autoclave.
- For UR - Routine concentrations of bleach (1 part bleach:9 parts water or spilled material) in cleanup materials from small spills can be sent to the Rotoclave™ using the Medical Center's red toters. If more than 5 gallons of spill cleanup materials are produced, contact EH&S for the best disposal method.

Biological Spill Cleanup, University of California at Berkeley EH&S
https://www.youtube.com/watch?v=l6uJvEQ-J9A (2 minutes)

C. Spills inside a biological safety cabinet (BSC):

1. Stop work, remove gloves and plan cleanup

2. Keep the BSC blower turned on

3. Use General Procedure for Biological Spills

4. Ensure that cabinet walls, work surface, and equipment inside the cabinet have been disinfected. Do not place your head in the cabinet. If your arm is not long enough to reach the back wall, then use an assist device such as a 'Swiffer' with a shortened handle to hold the disinfectant soaked towel.

5. If the spilled material has gotten under the work surface into the catch basin below, also disinfect the catch basin.
   a. Refer to the BSC's Operator Manual to locate the drain valve. Ensure the drain valve is closed.
   b. Perform spill cleanup for the catch basin
   c. If you need to flood the catch basin with disinfectant, you can use the drain valve to empty the liquid into a collection vessel. Attach a hose barb and flexible tube to the drain valve. To minimize aerosol generation, put a small amount of disinfectant in the collection vessel, and then submerge the tube end. Flush the catch basin/drain pan with water. Remove the drain tube.
   d. Reassemble the work surface.
6. After cleanup is completed, allow the cabinet to run for ten minutes before resuming work.

Visual demonstrations for spill cleanup in a BSC:

Cleaning Up a Spill (Biosafety Cabinet): Biosafety Level 3, Iowa State University Environmental Health & Safety https://www.youtube.com/watch?v=4St9yxL-crw (2 minutes)

Biosafety Cabinet Training at Arizona State University (includes removing the work surface to clean the catch basin underneath) https://www.youtube.com/watch?v=q_C6xq7j-kg (spill response starts at 13:40)

Fundamentals of Working Safely in a Biological Safety Cabinet (BSC): Cleaning Up a Spill in a BSC, Centers for Disease Control and Prevention https://www.youtube.com/watch?v=xaDny8vGkyg (3 minutes)

D. Spills inside a centrifuge:

1. Most BSL2 labs use centrifuge safety cups or biosafety-lidded rotors that allow the rotor to be removed from the centrifuge without opening the lid. These are opened in the biological safety cabinet. Perform spill cleanup in the BSC following the general spill procedures. Also clean the inside of the centrifuge.
2. For all other spills:
   a. Close the lid, turn off, and follow the General Procedure for Biological Spills.
   b. If possible, move the centrifuge or at least the rotors/buckets to a BSC.
   c. Open the rotor/bucket and using a squeeze bottle, apply disinfectant to inside rotor/bucket.
   d. Carefully remove any broken glass from inside rotor/bucket using forceps and place in a sharps disposal container.

e. Drain/suction the disinfectant from the rotor/bucket. Thoroughly wipe down the inside of the rotor/bucket including the lid with paper towels soaked in disinfectant. Rinse with water or ethanol and dry rotor/bucket and lid.

f. Rinse the disinfectant from the centrifuge chamber with water or ethanol. Absorb the liquid with paper towels and wipe down thoroughly.

3. Avoid using bleach on rotor / buckets. Use a previously-approved alternate disinfectant.

VI. REFERENCES

CDC Laboratory Training, Fundamentals of Centrifuge Safety, https://www.cdc.gov/labtraining/

CDC/NIH Biosafety in Microbiological and Biomedical Laboratories, 6th edition, 2020, https://www.cdc.gov/labs/BMBL.html

NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules https://osp.od.nih.gov/biotechnology/nih-guidelines/

Public Health Agency of Canada Safety Data Sheets https://www.canada.ca/en/public-health/services/laboratory-biosafety-biosecurity/pathogen-safety-data-sheets-risk-assessment.html

VII. APPENDICES/FORMS

Centrifuge Spill Plan

"Biohazard Spill – Do Not Enter" door sign template (hint: print on red paper or affix a biohazard sticker, laminate, and have in spill kit with a magnetic clip for the door).

VIII. REVISION HISTORY

Date	Revision No.	Description
	New	
2/1/2012	1	Included UHS for exposure reporting
6/12/2013	2	Revisions following NIH OBA site visit
6/27/2018	3	Update title and purpose (labs), update definitions, harmonize with Lab Safety Training, add video web links, update references, replace appendix
12/29/2021	4	Triennial review, update web links

Centrifuge Spill Plan

(for centrifuges without safety cups/buckets/rotors)

1. Close the lid. Turn centrifuge off.

2. Try not to breathe (for aerosols) as you step back from the spill. Once you're at a distance you think will have fewer aerosols, alert people in the immediate area of the spill and evacuate. If spill is in a lab, ensure the doors are closed.

- For BSL2 and BSL2+ spills
  - Do not re-enter the area for at least 30 minutes. This allows aerosols to settle and to be exhausted by the building ventilation system.
  - Post a "Biohazard Spill – Do Not Enter" sign on doors to keep all unnecessary people out of the area.
    - Hint: Keep one in your spill kit along with a magnetic clip for the door; print on red paper or affix a biohazard sticker, laminate).

3. Call Public Safety by dialing 13 from a campus phone or 275-3333 from a cell phone if needed to:

a. Limit access to the area, if the spill is in a hallway or running under the door.
b. Respond to injuries that require emergency personnel. Public Safety will notify emergency personnel and help direct emergency personnel to your location. This is the fastest way to get help!
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4. Remove contaminated clothing.

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- double gloves (BSL2, BSL2+) or single gloves (BSL1)
- lab coat
- face protection
  - safety glasses or goggles (BSL1)
  - chin-length face shield (BSL2, BSL2+)
  - surgical mask
- shoe covers/booties (dependent on size of spill, splatter, etc.)

2. If possible, move the centrifuge or at least the rotors/buckets to a BSC.

3. Open the rotor/bucket and using a squeeze bottle, apply disinfectant to inside rotor/bucket. Avoid using bleach on rotor / buckets. Use a previously-approved alternate disinfectant.

4. Allow the disinfectant to be in contact with the biological for at least 30 minutes.

5. Carefully remove any broken glass from inside rotor/bucket using forceps and place in a sharps disposal container.

6. Drain/suction the disinfectant from the rotor/bucket. Thoroughly wipe down the inside of the rotor/bucket including the lid with paper towels soaked in disinfectant. Rinse with water or ethanol and dry rotor/bucket and lid.

7. Rinse the disinfectant from the centrifuge chamber with water or ethanol. Absorb the liquid with paper towels and wipe down thoroughly.

8. Re-clean area with fresh paper towels soaked with disinfectant.

9. Discard disposable cloths/towels and disposable protective equipment into a Discard disposable cloths/towels and disposable protective equipment into a biohazard/ medical waste bin (red bag).

10. Remove non-disposable PPE.

11. Wash hands with soap and water.

(hint: print on red paper or affix a biohazard sticker, laminate, and have in spill kit along with magnetic clip for the door)

Biohazard Spill

Do Not Enter