Emergency Response and Biohazard Exposure Control Plan

I. PURPOSE

To establish emergency response and cleanup procedures for laboratory spills involving biohazards in Biosafety Level 1 (BSL-1) and Biosafety Level 2/2+ (BSL-2/2+) laboratories at Dartmouth College in accordance with:

- CDC/NIH Biosafety in Microbiological and Biomedical Laboratories (BMBL)
- NIH Guidelines for Research Involving Recombinant or Synthetic DNA Molecules (NIH Guidelines)

This document also outlines incident reporting procedures in order to comply with federal reporting requirements and to ensure timely and appropriate follow-up. Principal Investigators shall immediately report exposures and releases involving recombinant or synthetic nucleic acid (r/sNA) molecules as well as violations of the NIH Guidelines to the Dartmouth College Biological Safety Officer (BSO).

II. DEFINITIONS

For the purposes of this Emergency Response and Exposure Control Plan, biohazards are defined as any material or agent that may contain infectious or potentially infectious substances, or any agents or substances that are an environmental release risk (i.e., recombinant DNA).

Examples:
- Microbiological cultures or stocks (including bacterial, viral, parasitic, fungal, etc.)
- Recombinant or synthetic nucleic acid molecules (including viral vectors)
- Organisms or cells that contain recombinant or synthetic nucleic acid molecules (including transgenic organisms and those transiently containing exogenous nucleic acids)
- Human or animal cell or tissue cultures
- Anatomical or pathological waste (human or animal tissue or organs)
- Human clinical specimens (feces, blood, urine or any other bodily fluid)

According to the NIH Guidelines (Section I-B), recombinant or synthetic nucleic acids are defined as:

i. molecules that a) are constructed by joining nucleic acid molecules and b) that can replicate in a living cell, i.e., recombinant nucleic acids;

ii. nucleic acid molecules that are chemically or by other means synthesized or amplified, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules, i.e., synthetic nucleic acids, or

iii. molecules that result from the replication of those described in (i) or (ii) above.
III. RESPONSIBILITIES

i. Principal Investigators (PIs) and research laboratory staff who conduct biological or biomedical research or who work in biological or biomedical teaching laboratories must abide by the guidelines outlined in this Plan.

ii. It is the responsibility of the PI and laboratory supervisor to ensure that:

- An appropriate spill response plan has been developed for their lab using these generic guidelines as a basis,
- A copy of this Plan and/or lab specific plans are available to lab staff,
- Compliance is maintained,
- Appropriate disinfectants, personal protective equipment, and waste containers are readily available, and
- Reporting and follow-up procedures as described herein are conducted for all incidents.

iii. The Dartmouth Environmental Health & Safety (EHS) office provides OSHA-mandated bloodborne pathogen training and biosafety training to all laboratory staff with potential exposures. It is the responsibility of each supervisor to ensure all personnel under his/her supervision complete this required training.

iv. The PI will provide EHS with updated emergency contact info when applicable to ensure laboratory door signage is up to date.

IV. BIOHAZARDOUS SPILL EMERGENCY PREPAREDNESS

A. Risk Assessment

When assessing a spill to determine the appropriate response, the following should be considered:

i. What was spilled or released?
   - Liquid, solid, animal, etc.
   - What is the pathogenicity or route of transmission? Consult the BMBL and/or the Canadian Pathogen Safety Data Sheets for guidance.

ii. How much was spilled?
   - What is the volume and concentration of the organism?

iii. Where is the spill?
   - Is it in a biosafety cabinet, in the lab, outside the lab, etc.?

iv. What is the potential for release outside of the lab?
B. Biohazard Spill Kit

The following items should be assembled in a single container for easy transport to the spill area. Spill kits must be available at all times. A large bucket is a practical container as it may double as the secondary container for waste removal following cleanup.

- An appropriate chemical decontaminant: In most cases a 1:10 dilution of freshly prepared household bleach is appropriate.
- Materials to absorb liquids after decontamination: This may include paper towels, absorbent pads, or other materials designed to absorb large volumes of liquid.
- Appropriate PPE to wear during cleanup: nitrile or heavy duty gloves, a long-sleeved laboratory coat or gown, and goggles are always necessary. Mucous membrane protection should be considered for large spills.
- A mechanical means for handling broken glass: This may include tongs, forceps, small disposable scoops and sponges, autoclavable dustpans, or any other method that prevents direct contact with the broken glass.
- Biohazard bags, sharps containers, and/or other containers: The containers are used to hold the material for further treatment and disposal.

V. BIOHAZARDOUS SPILL EMERGENCY RESPONSE

A. Biohazardous Spill Inside a Biological Safety Cabinet (BSC)

This section provides spill cleanup procedures for biohazardous agents, including recombinant or synthetic nucleic acids, inside a BSC.

Spill inside a BSC that stays contained on the work surface

i. Operate the BSC to prevent escape of contaminants from the cabinet. Spill cleanup procedures should be initiated immediately while the cabinet continues to run.

ii. Remove contaminated sharps from the spill area using mechanical means (e.g. tongs or forceps). Never remove contaminated sharps by hand. Discard contaminated sharps in a sturdy, leak-proof, biohazard labeled sharps container.

iii. Cover the spill with paper towels or other absorbent material. Slowly pour an agent-appropriate disinfectant such that the solution flows into the spill. Paper towels soaked with the disinfectant may also be used to cover the area. A freshly prepared 1:10 dilution of household bleach used with a 20 minute contact time, and followed by a water rinse to prevent corrosion is sufficient in most cases. However, refer to the laboratory specific disinfection protocol for the appropriate disinfectant to use.

iv. A minimum of 20 minutes is generally considered an appropriate contact time for thorough decontamination, but the length of time depends on disinfectant and biohazard. Follow manufacturer’s recommendations and laboratory specific disinfection protocol for the given biohazard.

v. Wipe up the spill, work surfaces, walls, and any equipment in the cabinet with paper towels dampened with decontaminant. Do not place your head in the cabinet to clean the spill; keep your face behind the sash.
vi. Place contaminated paper towels and other spill cleanup materials in orange autoclave biohazard bags.

vii. Decontaminate the spill area a second time, placing all used spill materials into a biohazard bag.

viii. Remove any contaminated PPE in a manner to avoid cross-contamination; dispose of per standard lab practices.

ix. Wash hands thoroughly after removing gloves and other PPE.

**Spill inside a BSC that flows past the work surface through the front or rear grills**

A large spill inside a BSC that flows past the work surface through the front or rear grills requires more extensive decontamination. To prevent escape of contaminants from the cabinet, spill cleanup procedures should be initiated immediately while the cabinet continues to run.

i. Ensure the drain valve under the BSC is closed.

ii. Remove contaminated sharps from the spill area using mechanical means (e.g. tongs or forceps). Never remove contaminated sharps by hand. Discard contaminated sharps in a sturdy, leak-proof, biohazard labeled sharps container.

iii. Flood the top work surface tray and, if a Class II BSC, the drain pans and catch basins below the work surface with an agent appropriate disinfectant solution. A freshly prepared 1:10 dilution of household bleach used with a 20-minute contact time, and followed by a water rinse to prevent corrosion is sufficient in most cases. However, refer to the laboratory specific disinfection protocol for the appropriate disinfectant to use.

iv. A minimum of 20 minutes is generally considered an appropriate contact time for thorough decontamination, but the length of time depends on disinfectant and biohazard. Follow manufacturer’s recommendations and laboratory specific disinfection protocol for the given biohazard.

v. Remove excess decontaminant from the work surface tray by wiping with a sponge or cloth. For Class II BSCs, drain the tray into the catch basin below the work surface, lift the tray and take out the removable front intake grille. Wipe the top and bottom (underside) surfaces of the grille with a sponge or cloth soaked in the decontaminant. Then place the tray in position, drain the decontaminant from the cabinet base into an appropriate container, and dispose of the decontaminant in the sewer.

vi. Place spill cleanup materials (e.g. contaminated gloves, cloth and/or sponge) in autoclavable pans with lids for autoclaving.

vii. Decontaminate the spill area again. Place all used spill materials into a biohazard bag.

viii. Remove any contaminated PPE in a manner to avoid cross-contamination and dispose of per standard lab practices.

ix. Wash hands thoroughly after removing gloves and other PPE.
B. **Biohazardous Spill Outside a Biological Safety Cabinet (BSC)**

This section provides spill cleanup procedures for biohazardous agents, including recombinant or synthetic nucleic acids, outside a BSC.

**Small spills of agents NOT transmitted by inhalation** (can be absorbed with one paper towel)

i. Assemble spill cleanup materials.

ii. Put on appropriate PPE (e.g. long-sleeved lab coat, goggles, and nitrile gloves).

iii. Remove contaminated sharps from the spill area using mechanical means (e.g. tongs or forceps). Discard contaminated sharps in a sturdy, leak-proof, biohazard labeled sharps container.

iv. Pour a disinfectant solution (e.g. freshly prepared 1:10 dilution of household bleach) around the spill and allow it to flow into the spill. Paper towels soaked with the decontaminant may also be used to cover the area. To avoid aerosolization, never pour decontaminant solution directly onto the spill. Refer to the laboratory specific disinfection protocol for the appropriate disinfectant to use.

v. Let stand for 20 minutes to allow an adequate contact time and place all used spill materials in a biohazard bag. The length of time depends on disinfectant and biohazard. Follow manufacturer’s recommendations and laboratory specific disinfection protocol for the given biohazard.

vi. Decontaminate the spill area a second time. Place all used spill materials into a biohazard bag.

vii. Remove any contaminated PPE in a manner to avoid cross contamination and dispose of per standard lab practices.

viii. Wash hands thoroughly after removing gloves and other PPE.

**Larger spills or ANY spill of agents transmitted by inhalation**

i. Evacuate the room immediately and warn other lab occupants of the spill. Close the door and wait 30 minutes to allow aerosols to dissipate.

ii. Post a sign on the door and prevent others from entering the contaminated area.

iii. Remove contaminated garments and put into a container for autoclaving. Thoroughly wash any exposed areas of the body.

iv. Assemble spill cleanup materials.

v. Put on appropriate PPE (e.g. long-sleeved gown, goggles, and nitrile or heavy duty gloves) before re-entering the room.

vi. Remove contaminated sharps from the spill area using mechanical means (e.g. tongs or forceps). Discard contaminated sharps in a sturdy, leak-proof, biohazard labeled sharps container.
vii. Pour a disinfectant solution (e.g. a freshly prepared 1:10 dilution of household bleach) around the spill and allow it to flow into the spill. Paper towels soaked with the decontaminant may be used to cover the area. To avoid aerosolization, never pour decontaminant solution directly onto the spill. Refer to the laboratory specific disinfection protocol for the appropriate disinfectant to use.

viii. A minimum of 20 minutes is generally considered an appropriate contact time for thorough decontamination, but the length of time depends on disinfectant and biohazard. Follow manufacturer’s recommendations and laboratory specific disinfection protocol for the given biohazard.

ix. Using an autoclavable dustpan and squeegee, transfer contaminated materials (e.g. paper towels, glass, gloves, etc.) to an autoclave bag.

x. Decontaminate the spill area a second time, placing all used spill materials into autoclave pan.

xi. Separate reusable items from non-autoclavable plastic as the plastic will melt. Cover the pan with a lid and autoclave according to standard directions.

xii. Remove any contaminated PPE in a manner to avoid cross-contamination and dispose of per standard lab practices.

xiii. Wash hands thoroughly after removing gloves and other PPE.

**Spills inside of a centrifuge**

Spills or breakage inside of an operating centrifuge pose a serious potential for exposure due to the creation of aerosols. If a primary container has broken in a centrifuge without a closed rotor or bucket, immediately suspend use and notify lab staff, PI, and request assistance from the Biosafety Officer (603-646-9790). For large spills or failures, contact EHS (603-646-1762).

i. For suspected or confirmed spills/breakage in any centrifuge, wait at least 30 minutes after the centrifuge has stopped operating to initiate cleanup.

ii. Put on lab coat, gloves and a face shield prior to opening centrifuge. Open carefully to assess the damage.

iii. If the spill is contained within a closed cup, bucket or rotor, spray the exterior with disinfectant and allow 20 minutes of contact time. Transfer the carrier to the nearest biosafety cabinet (BSC). If a biosafety cabinet is not available, close the centrifuge and post a sign warning of the spill. Notify the PI and BSO for assistance.

iv. If a BSC is available, gather supplies needed, such as a sharps container for broken glass and bins filled with disinfectant and place into the BSC. Use forceps to remove broken glass and place directly into a sharps container. Carefully remove any unbroken tubes and place into a bin filled with disinfectant for 20 minutes. Wipe carrier with disinfectant.

v. After disinfection, the carrier, bucket or rotor should be washed with a mild soap and water.

vi. Spray the interior of the centrifuge chamber with a disinfectant, let sit for 20 minutes and then wipe down.
vii. Dispose of all cleanup materials (except sharps) in orange autoclave bags. Dispose of sharps in biohazard sharps containers.

viii. Remove protective clothing and thoroughly wash hands.

C. Spills Outside the Laboratory in Public Spaces

Samples must be transported in secondary, leak-proof containers to minimize the potential for spills. However, if a spill does occur in a common hallway or public space and cannot be immediately decontaminated, cordon off the area, restrict access, and contact EHS immediately at 603-646-1762.

VI. EXPOSURE RESPONSE AND REPORTING PROCEDURES

Examples of biohazard exposures that require response and reporting include:

- Needlesticks or other percutaneous injuries from a contaminated sharp
- Splashes to mucous membranes (e.g. eyes, nose, mouth)
- Bites/scratches from animals that have been exposed to any recombinant or synthetic nucleic acid material, whether or not the exposure leads to illness

A. Immediate Response

i. For **SKIN** exposure: Immediately remove contaminated personal protective equipment or clothing and wash the contaminated area with an iodine solution or antibacterial soap and copious water for 10 minutes.

ii. For **EYE** exposure: Flush the eye with water for at least 15 minutes at an eyewash station.

iii. Notify PI or supervisor. If PI/supervisor is not available, immediately proceed to next step.

B. Medical Treatment

i. During work hours, report to Dick’s House on the Hanover campus (603-646-9400) between 7:30 am-4:30 pm if you are a student **-OR-** report to Occupational Medicine at DHMC Faulkner Building, Level 4 near parking garage (603-653-3850) between 7:30 am-4:30 pm if you are an employee.

ii. Bring along Safety Data Sheets or other literature pertaining to any chemical or biohazard exposure.

iii. After hours and weekends: report to DHMC Emergency Room.

iv. If transport assistance is needed, contact Safety and Security on Hanover campus at 603-646-3333, or 5555 if at DHMC.

v. If exposure requires emergency treatment, call 911 (or 5555 at DHMC).
C. Reporting Exposures, Incidents, Accidental Releases

i. Notify the Biological Safety Officer (BSO) immediately.

ii. The BSO will investigate the incident and notify the IBC Chair and EHS Director.

iii. The PI will complete an internal Incident Report Form and submit it to the BSO and Risk Management within 24 hours.

iv. If the IBC Chair and BSO determine that the incident involves non-exempt r/sNA molecules, the BSO will submit an NIH incident report to the NIH Office of Biotechnology Activities within 30 days. Incidents occurring in BSL-2 laboratories resulting in an overt exposure will be immediately reported to NIH OBA.

v. The Office of Risk Management and the Vice Provost for Research will also receive a copy of the incident report.

VII. BSL-2/BSL-2+ RESEARCH

A. Overview

i. Improved engineering and regulation of work practices are the primary means of elimination or minimization of exposure for personnel. Where the possibility of occupational exposure remains, personal protective equipment is to be used.

ii. Universal Precautions are used to prevent exposure to bodily fluids/tissues or other materials containing biological pathogens. These precautions are to be taken at all times when working with all RG2 agents or when the risk of exposure/contamination is present.

iii. All clinical specimens of blood, human tissue and bodily fluids are to be handled utilizing BSL-2 work practices and procedures. These practices, procedures and facility requirements are described in detail in Biosafety in Microbiological and Biomedical Laboratories and are to be followed by all Dartmouth College laboratories working with biological materials.

B. Engineering Hazard Controls

Engineering controls are to be utilized in circumstances in which an occupational exposure exists.

i. **Primary Barriers.** Class II Type A Biological Safety Cabinets (BSC) or other physical containment devices must be used when aerosol-creating procedures are performed. Such procedures include centrifuging, grinding, vortexing, blending, sonication and opening containers of infectious materials. Intranasal inoculations or other animal procedures that have the potential for producing splashes and aerosols must be performed in a biological safety cabinet. In special cases, procedures (such as an animal necropsy) may be performed on an open bench if it is determined by the Biological Safety Officer that the employee is at a significant increased risk of percutaneous exposure to an infectious agent transmissible by the bloodborne route when working in a biological safety cabinet. In these cases, strict adherence to mucous membrane protective practices is required.
ii. **Biological Safety Cabinets.** Annual inspection and certification of biological safety cabinets is the responsibility of the primary investigator. EHS maintains a database of biological safety cabinets and will aid in the scheduling of inspections and certifications. If you need to move your BSC, it must first be professionally decontaminated and recertified once in place. Please contact EHS for assistance (603) 646-1762.

iii. **Mechanical Pipetting Devices.** Mechanical pipetting devices are to be used for all pipetting activities. **Mouth pipetting is strictly prohibited.** Aerosol-resistant pipette tips are recommended to control carry-over contamination caused by aerosol formation. The tips are ideal for a wide variety of applications including PCR, tissue culture, forensic studies, gel loading, and serological assays as well as pipetting radioactive samples.

iv. **Sharps Containers.** Puncture resistant sharps containers are to be used at all work sites where needles and syringes, Pasteur pipettes, scalpel blades and other sharps are used. Sharps containers should be closed and prepared for disposal when ¾ full. Please refer to the [Biohazardous Waste Disposal Guide](#) for the correct disposal option for your building. Puncture resistant sharps containers are available through the scientific stockrooms.

v. **Safer Needle Devices.** On April 18, 2001, the Occupational Safety and Health Administration (OSHA) issued the Needlestick Safety and Prevention Act. Congress directed OSHA to make changes to its Bloodborne Pathogens Standard (CFR 1910.1030) to require the use of safer needle devices when drawing blood. For further information, please contact EHS. Blood drawing in your laboratory must be approved by EHS.

vi. **Safety Devices for Centrifuges.** For low speed centrifugation of infectious materials, safety centrifuge cups must be used. If used, the cups are to be loaded and unloaded only within a biological safety cabinet. High-speed centrifugation of infectious material must be performed using a suitable gasketed rotor that is loaded and unloaded within a biological safety cabinet. Inside a biological safety cabinet, wipe off the exterior of the rotor with a suitable disinfectant before loading and unloading.

vii. **Vacuum Line Protection.** All vacuum lines must be protected by using in-line vacuum HEPA filters and vacuum traps. These disposable filters are available through the stockrooms. In labs where the vacuum lines are used routinely, it is recommended that the filters be changed every six months. In other areas, it is recommended that filters be changed on an annual basis. Immediately change the filter if the system no longer works effectively or becomes overtly contaminated.

viii. **Vacuum Trap Decontamination.** Vacuum traps used for liquid biohazardous waste collection must contain appropriate disinfectants prior to use. Wescodyne is recommended since it is a stable disinfectant. The waste must be removed and the flask cleaned whenever ½ volume of flask volume is reached, or every 3 months (which ever comes first). Vacuum trap decontamination schedule stickers must be placed on all vacuum trap flasks to serve as a record of proper disinfection and disposal. These stickers are readily available from EHS as seen here.
C. Work Practice Controls

i. **Hand Washing.** Hand washing is the single most effective means of preventing exposure to, or spread of, infectious agents. Hand washing must be performed after removing gloves, upon completion of a task within a biological safety cabinet, before leaving the lab, when hands are known or suspected to be contaminated and before contact with face or mouth. Hands should be lathered thoroughly with an antimicrobial soap, vigorously washed for approximately 15-30 seconds then rinsed with copious amounts of water. Use paper towels to turn off the water faucet to avoid immediate recontamination.

ii. **Food and Consumables.** Eating, drinking, smoking, applying cosmetics or lip balm, and handling contact lenses are prohibited in all laboratory settings and other work areas where there is a possibility of occupational exposure. Food and drink shall not be kept in laboratories or refrigerators, freezers, shelves, cabinets or on countertops or bench tops.

iii. **Sharps Safety.** Needles and other sharps must not be bent, recapped, or removed from syringes except when the lab supervisor determines that no alternative is feasible or that such action is required by a specific procedure. Recapping should only be performed using a mechanical recapping device. Shearing or breaking of contaminated needles is prohibited.

iv. **Aerosols.** All procedures involving potentially infectious material shall be performed in such a manner as to minimize splashing, spraying, spattering, and the generation of droplets of these substances. Work shall be conducted in a biosafety cabinet whenever possible to minimize exposures to aerosols.

v. **Transport.** Transport of biohazardous material outside of the laboratory must be done in secondary containment. If the specimen could puncture the primary container, the primary container shall be placed within a secondary container that is puncture-resistant in addition to the above characteristics. For transport or shipping of the material outside of the facility, additional US Department of Transportation (DOT) training requirements apply. Contact the Biosafety Officer at biosafety@dartmouth.edu for more information.

D. Personal Protective Equipment

i. **Laboratory Coats.** Lab coats are to be worn at all times while working on, or adjacent to, bench top procedures utilizing hazardous chemicals, biological or unsealed radiological materials. Laboratory coats must be appropriately sized for the individual and must be fully buttoned when worn. Sleeves must be of appropriate length as to not expose skin while wearing gloves. Lab coats are not to be taken from the laboratory and brought to non-laboratory areas, especially where food and drink is being prepared or served. **It is the responsibility of the PI to ensure all laboratory personnel adhere to Dartmouth’s standard of wearing laboratory coats.**

ii. **Gloves.** Gloves are to be worn by all employees directly handling biological material or contaminated surfaces. Vinyl examination gloves, surgical synthetic, or nitrile gloves may be chosen by the employee based on individual need and preference. Gloves are to be inspected before use and changed routinely. Gloves must be replaced when visibly soiled, torn, or punctured. All gloves must be discarded into an autoclave bag for proper disposal. Information on glove selection and chemical resistance is found on a poster displayed in each laboratory and from EHS. **Gloves are never a substitute for thorough hand washing. Hands should be thoroughly washed when entering the lab, after removing gloves and before leaving the lab.**
iii. **Mucous Membrane Protection.** Mucous membranes must be protected by wearing a surgical-type mask with safety glasses or a surgical-type mask with attached acetate eye shield when working with infectious or potentially infectious materials outside of a biological safety cabinet.

iv. **Safety Glasses/Shields.** Safety glasses are recommended at all times in the laboratory.

v. **Personal Clothes.** Any unprotected skin is forbidden. Sandals and other open-toed footwear as well as shorts or skirts that leave the skin unprotected are prohibited in all Dartmouth laboratories.

vi. **Respirators.** Respirators must not be used in the laboratory without prior approval from EHS. Contact EHS if you feel respiratory protection is required. Surgical facemasks used for mucous membrane protection are not respirators and are not to be used in situations where respiratory protection is required. All respirator users must be enrolled in the Dartmouth College Respiratory Protection Program. EHS supplies and maintains the recommended respiratory protective devices.

**E. Posting Hazard Requirements**

All labs working with biohazardous agents must post the EHS provided biohazard sign on the laboratory door. This sign must indicate the types of hazards, PI, and emergency contact information, and should be updated as necessary.

Any piece of equipment in common room areas (such as cold room, incubator room, equipment room, etc.) that contains or is used with RG-2 agents must have a biohazard warning label attached. The label must state the biological hazard, date and person of contact. These labels can be obtained from the stockrooms or from EHS.

**VIII. RESOURCES**

- **Biohazardous Waste Disposal Guide.** Dartmouth IBC 2022
  [http://www.dartmouth.edu/~ehs/biological/policies_sops.html](http://www.dartmouth.edu/~ehs/biological/policies_sops.html)

- **Biosafety in Microbiological and Biomedical Laboratories, 6th ed.** Centers for Disease Control and Prevention and National Institutes of Health, 2020 June
  [https://www.cdc.gov/labs/BMBL.html](https://www.cdc.gov/labs/BMBL.html)

- **Bloodborne Pathogens.** Occupational Safety and Health Administration, 29 CFR 1910.1030

- **NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules (NIH Guidelines).** National Institutes of Health Office of Science Policy, 84 FR 17858, 2019 April