NIPISSING UNIVERSITY

ENVIRONMENTAL HEALTH AND SAFETY

Standard Operating Procedure for Biosafety Cabinet Use

Revised January 2022

STANDARD OPERATING PROCEDURE FOR BIOSAFETY CABINET USE

INTRODUCTION

When properly maintained and used in conjunction with good laboratory techniques, Biosafety Cabinets (BSCs) provide effective primary containment for work with infectious material or toxins. In containment level 2 facilities, BSCs are used for procedures with the potential to produce infectious aerosols and for high concentrations or large volumes of infectious material. All manipulations of biohazardous agents that require level 2 containment with level 3 operational practices (commonly called 2+) must be performed in a BSC.

This SOP outlines the safety policies and work practices to provide optimum contamination control and safety when working in a Biosafety Cabinet (BSC). Please review the following videos prior to proceeding: (https://youtu.be/4DoHJS8JL4U for set-up and https://youtu.be/18QEJUA9XBs for Best Practices for use.

MATERIALS REQUIRED

- Bottle of disinfectant suitable for the pathogens being used. If using 10 % bleach, a bottle of type II water or 70% ethanol will also be required to rinse the bleach.
  ****NOTE** Refer to the PSDS for the appropriate disinfectant and contact time.**

Paper towel and tissues such as Kimwipes.

Biohazard bags.

Tongs or large forceps for spill clean-up.

Autoclave tape.

Discarded? pipette tray or box.

Bacticinerator for loop sterilization.

Hot/MicroBead Sterilizer for spreader sterilization or ideally, use disposable spreaders.

****NOTE** Open Flames Are Not Allowed Inside The BSC**.

- PPE appropriate for the task, i.e. non-flammable gloves, lab coat, eye protection.
- Any other materials required for your work or task.

PROTOCOL

1. START-UP

- Check the certification date expiry. If expired, do not use and contact the Human Resources Generalist: Health, Safety & Wellness at extension 4811.
- Turn on the cabinet fluorescent lighting and turn off the UV lighting if on. UV lighting is not effective for disinfecting surfaces.
- Close the drain valve.
- Turn on the blower fan and allow to run for at least 10 minutes.
- Check the magnehelic or electronic gauge (will vary by BSC) to ensure proper HEPA filter operation. It should read within 0.2 of the recorded reading on the certification sticker. If lower, do not use the cabinet. Call the Laboratory Safety Coordinator for instructions.
- Check the alarms for proper function by pressing the test button if available, or by raising the sash 10.5 inches.
- Surface decontaminate the work surface, side walls and inner back walls (usually by spraying with 70% ethanol or bleach and then wiping down after an appropriate contact time). Wear gloves for hand protection and it's important to re-sterilize your gloves each and every time the users hands come out of the cabinet. Please watch this video for proper technique: https://youtu.be/b0PtPEnNakc.
- Surface decontaminate all items required and place into the cabinet (same method as above).
- Set up the workflow from clean to dirty with the work area in the middle (see Figure 1). Keep bulky items such as biohazard bags and pipette trays and boxes towards the back and side of the cabinet. Place aerosol generating equipment towards the back of the cabinet.

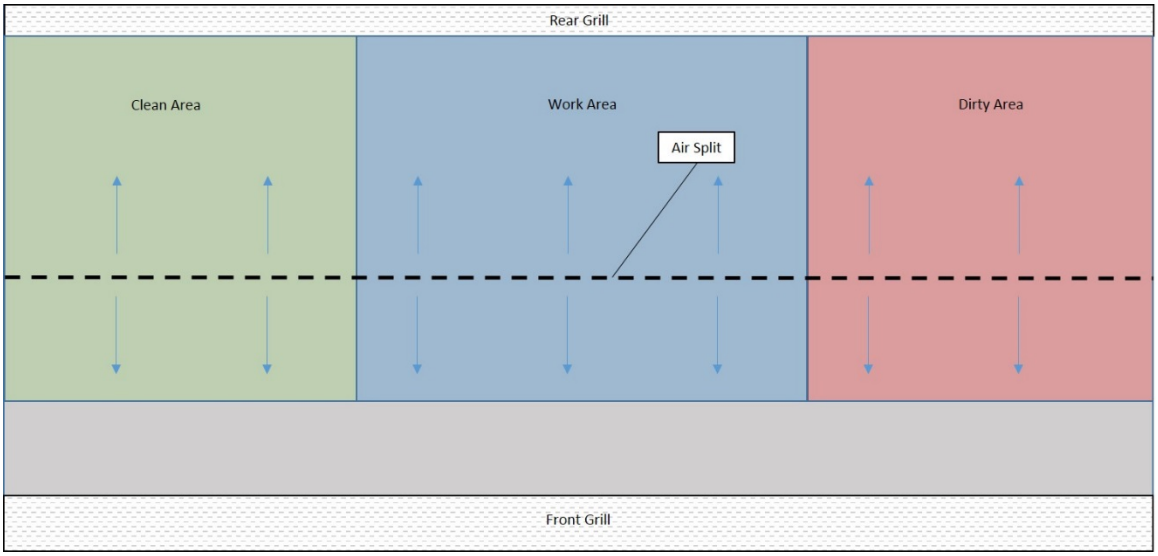

Figure 1. Biosafety cabinet workspace set-up (top view). Work should be performed at or behind the air split from clean to dirty.

- Check that air grills are not blocked.
- Do not overcrowd the work zone.
- Re-check the magnehelic or electronic gauge and check the front air flow at the grill using a tissue.
- Arrange stool height so that your armpits are at the level of the bottom of the sash.
- Allow the work zone air to purge for at least 5 minutes before commencing work.

2. WORKING INSIDE THE BSC

- Movement of arms into and out of the cabinet can disrupt airflow. This can adversely affect cabinet performance. Whenever possible, place all materials needed for a procedure inside the cabinet before starting. Move arms slowly and move straight out of the cabinet; do not sweep arms across the front of the cabinet. Do not walk quickly in front of a cabinet when someone else is working Only one individual should be working in the cabinet at a time unless absolutely necessary.
- Never put anything on the grill at the front opening of the cabinet.
- Do not block the air openings/grill at the back of the cabinet.
- Place supplies, equipment, and papers well back from the front of the cabinet, positioned so that air intake or exhaust grills are not obstructed. Avoid bringing non-essential equipment and supplies into the cabinet.

- Work towards the middle of the cabinet, at the air split or at least 6 inches from the front grill.
- Discard contaminated waste including contaminated gloves INSIDE the cabinet. Do not allow contaminated gloves outside of the cabinet.
- **Clean up spills as soon as they occur**. For procedures on spill clean-up see "Spill Response Procedure" below.

3. UPON COMPLETION OF WORK

- Close or cover open containers and leave the blower on for at least five minutes with no activity to purge the cabinet.
- Surface-disinfect objects before removal from the cabinet. Remember that aerosols generated during operations in the cabinet, such as pipetting, might have contaminated objects in the cabinet. As a result, there does not need to have been a spill for this step to be necessary.
- Remove contaminated gloves and dispose of them appropriately (inside the cabinet). Wash hands thoroughly with soap and running water.
- Don clean gloves and ensure that all materials are placed into biohazard bags within the cabinet.
- Disinfect cabinet surfaces with an appropriate disinfectant. Periodically remove the work surface and disinfect the area beneath it (including the catch pan) and wipe the surface of the UV light with disinfectant.
- Allow the blower fan to run for at least 5 minutes
- Turn off the blower and fluorescent lamp. **Do not turn on the UV lamp.**
- Disinfect or dispose of personal protective equipment appropriately and wash hands.

4. PRECAUTIONS

- **An open flame should not be used in a BSC**. Bunsen burners are fire and explosion hazards, they contribute to the heat load, generate convection currents that interfere with airflow and have been known to burn holes in the HEPA filters. If necessary, discuss alternatives with the Laboratory Safety Coordinator (e.g. micro-incinerator for bacterial loops).
- The HEPA filters in the BSCs remove particulates from air, but they are **not effective at collecting chemical gases or vapours**. If you need to use such material in a BSC, contact the Laboratory Safety Coordinator for advice.
- **DO NOT use the BSC if the ALARM sounds** or if there are other indications of cabinet malfunction, such as no airflow, reduced pressure on gauges (drop > 0.2), or unusual noises.
- If alarm or other indication of failure happens while using the cabinet:
    - Seal, surface decontaminate and remove any biohazardous material.
    - Decontaminate the interior of the BSC.
    - Switch off the alarm or the power if the motor is making noise.

- Place a sign on the cabinet to indicate that it is broken and must not be used.
- Contact HR Generalist: Health, Safety and Wellness for advice and servicing (ext. 4811).
- **If personnel may have been exposed to infectious material due to cabinet failure, then the supervisor must be promptly notified, and an incident report completed and the appropriate first aid and medical follow-up action taken.**

5. SPILL RESPONSE PROCEDURE

A. SPILL IN CONTAINMENT

Remove and disinfect the grill if contaminated and remember to clean under the grill. If the spill was relatively large or contained concentrated infective material, then allow the cabinet to sit undisturbed for at least 5 minutes for aerosols to clear before beginning cleanup. While you wait, remove and disinfect or dispose appropriately of contaminated personal protective equipment (PPE) and wash thoroughly. Then don clean PPE and proceed with spill cleanup as outlined in the Biosafety Manual. *Remember to allow appropriate contact time for the disinfectant*.

B. SPILL OUTSIDE OF CONTAINMENT

Review the spill response procedures as outlined in the Biosafety Manual. The response will depend on the nature of the spilled material and how much is spilled.