Class II Biosafety Cabinets

Biohazardous Spills inside Biosafety Cabinet (BSC)

Follow these procedures when cleaning a small biohazardous spill inside a BSC:

- Decontamination steps should be initiated while the BSC continues to operate to prevent the escape of contaminants. Notify the Principal Investigator (PI) before starting the spill cleanup.
- Notify co-workers of the spill.
- Wait at least five minutes to allow the BSC to contain aerosols.
- Wear lab coat, safety glasses and gloves during cleanup.
- Cover small spill with paper towels or other absorbent material. Slowly pour disinfectant solution (70% alcohol or other effective disinfectant) around the spill and allow to flow into the spill. Let the disinfectant work for 15 to 30 minutes.
- Dispose of the spill-disinfectant-soaked paper towels/absorbent in a biohazard bag.
- Wipe the spillage area with disinfectant-soaked paper towels.
- Spray or wipe the walls, work surface and any equipment in the cabinet with disinfectant-soaked paper towels, and remove all equipment inside the BSC. Do not put your head inside of the BSC! Do not spray the HEPA filter!
- Discard contaminated disposable materials using appropriate biohazardous waste disposal procedure (Stericycle).
- Pick up any glass (or other sharps) with tongs or forceps and discard in a sharps container labeled with a biohazard sign.
- Place contaminated reusable items in autoclavable bags and store these closed bags in a secondary container before autoclaving is done.
- Expose non-autoclavable materials to disinfectant (20 minute contact time) before removal from the BSC.
- Remove all PPE and place protective clothing in an autoclavable bag for autoclaving.
- Wash your hands with soap and warm water.
- Run cabinet 10 minutes after cleanup before turning cabinet off.

If the spill in the BSC is quite substantial (spill flows past the work surface through the front or rear grilles), it may be necessary to decontaminate the cabinet’s fans, filters, and airflow plenums. An outside company must do this. Call the EH&S Office at 817-272-2185 for assistance.
Biosafety Cabinet (BSC) Decontamination Guidelines

Decontamination of a BSC is an important function in terms of both containment and sterility. Aseptic technique and daily decontamination will eliminate the majority of contaminants. In addition, periodic and thorough decontamination routines (including disinfecting all removable parts and surfaces) is recommended by the manufacturer and is a good laboratory practice to reduce wear on the equipment and provide greater safety to the worker, samples that are worked with, and the environment.

Extreme caution should be observed when decontaminating BSCs. For example, use caution when wiping BSC surfaces to avoid injury from broken glass that may be present and sharp metal edges. When removing internal parts of the cabinet, it is best to work with a partner as some of the workbenches and grilles may be heavy and have sharp edges. Proper personal protective equipment (PPE) must be used.

The guidelines below address routine periodic decontamination of most BSCs used in BSL-1 and BSL-2 settings. Always refer to the manufacturer’s operation and maintenance manual for instructions on periodic decontamination procedures. These guidelines are intended to be supplemental and are not a replacement for the manufacturer’s recommendations.

The guidelines below do not refer to the decontamination procedure that would be performed by a third party (e.g. using formaldehyde gas) when BSC that has been used for work involving infectious materials must be decontaminated before high efficiency particulate air (HEPA) filters are changed, or when maintenance work is planned.

Follow these procedures when performing a periodic and thorough decontamination of a BSC:

- **Always wear appropriate PPE when performing decontamination procedures.** At minimum, PPE includes a lab coat, gloves, and eye protection. Long pants and sturdy, closed-toe shoes that provide maximum foot protection are recommended. Whenever possible, tuck lab coat cuffs inside gloves.

- **Prepare for the decontamination.** Plan on at least one hour or more of uninterrupted time to complete a thorough disinfecting routine. Schedule this in advance with others in the lab in order to minimize down time. When performing a full decontamination, which includes removing internal parts (such as the grille and work bench), it is best to set up an area adjacent to the BSC to place these items. Ensure that several layers of absorbent materials are placed on the floor to soak up the disinfectant being used. It is also a good idea to have a partner who can help move the heavier removable parts in and out of the BSC.

- Determine the appropriate disinfectant based on the microbiological materials used in the BSC. Twenty (20) to 30 minutes is generally considered an appropriate contact time for decontamination, but this varies with the disinfectant and the microbiological agent.
Manufacturer’s directions must always be followed. The use of chlorine bleach or halogen chemicals may damage the stainless steel surfaces. For example, when using a 1% bleach solution, follow with a sterile distilled water rinse to remove any residue or with 70% alcohol (or similar non-corrosive antimicrobial agent) for final decontamination. Consider using a squeegee type tool or a duster with disposable pads (such as a clean room duster) to wipe the internal surfaces, rather than paper towels (which can cause lint). This is also more sustainable. A less expensive and effective alternative is a sweeper floor mop (e.g., Swiffer) with a shortened handle. The disposable dry cloth can be soaked with disinfectant and then easily applied evenly to all surfaces. Remove and discard the cloth into the biohazard waste after use and repeat the procedure with a fresh cloth for the sterile distilled water and/or the 70% alcohol.

Follow decontamination procedure. For all decontamination steps, consider using a spray bottle with a mist setting to liberally apply disinfectant. Using an up and down, left to right sweeping motion ensures all surface areas are completely covered and no area remains dry. Avoid spraying the ultraviolet (UV) light bulb and HEPA filter. Always wait for the appropriate contact time to elapse, then wipe to remove excess liquid. If bleach was used as a disinfectant, follow with either sterile distilled water and/or 70% ethanol to remove any residue.

- Turn on the BSC in normal operational mode with internal blower and light on.
- Check the pressure on the magnehelic gauge. (The pressure should be the same after cleaning and if not, could indicate the HEPA filter was disrupted and the BSC may require recertification).
- Open the sash for normal operation and allow for the normal purge time of at least five minutes before starting the decontamination of the BSC.
- Spray disinfectant on a biohazard waste bag and place it in the BSC to collect the used absorbent materials.
• Remove and place any small removable parts on the workbench surface.

• Working from left to right and top to bottom, spray disinfectant on all internal surfaces: the left wall, back wall, and right wall. Next, spray the workbench and the entire inside of the glass sash. Allow the disinfectant to soak while conducting the next steps.

• Carefully loosen any thumbscrews (if applicable) to release the removable workbench surface.

• To ensure thorough cleaning of the workbench area, these parts need to be removed and cleaned outside of the BSC. Using a partner, remove the front perforated grille and removable workbench surface. Prop up against a sturdy surface adjacent to the BSC (e.g., lab bench) by positioning perpendicular on the floor on top of several layers of absorbent materials (e.g., lab bench protectors, soakers).

• Clean the front perforated grille and removable workbench surfaces by spraying disinfectant onto the outward facing surfaces. Allow for appropriate contact time to elapse before wiping. Turn the pieces over and repeat the cleaning process.

• Once the grille and workbench have been thoroughly cleaned outside of the BSC, remove gloves and wash your hands with soap and water and put on a new pair of gloves before re-entering the BSC to complete the disinfection process under the workbench surface now that it has been removed.

• Inspect under the workbench surface. Carefully remove any debris such as paper towels, glass beads, stray sharps, tubes, Pasteur pipettes, ampoules, or broken glass. Use tongs or forceps for glass!

• Disinfect the under workbench surfaces including tray supports and the plenum drain area. Rinse by spraying sterile distilled water (if required) and follow with spraying 70% ethanol. Wipe dry.

• Using a partner, carefully replace and reposition the grille and removable workbench surface back into the BSC.

• Tighten any thumbscrews to secure the workbench surface in place (if applicable).

• Disinfect the grille and workbench surface.

• Working from left to right and top to bottom, spray with sterile distilled water (if rinse is required) or spray 70% alcohol on all internal surfaces: the left wall, back wall, and right wall, then the workbench and the entire inside of the glass sash. Wipe dry.

• Transfer all waste materials into the biohazard bag, close the bag inside of the cabinet, and wipe with disinfectant prior to removal.

• Repeat the check of the pressure reading on the magnehelic gauge. (The pressure should be the same after cleaning and if not, could indicate the HEPA filter was disrupted and the BSC may require recertification.)

• Let the BSC blower operate 10 minutes after decontamination before turning it off.

• Remove all PPE and place protective clothing in an autoclavable bag for autoclaving.

• Wash your hands with soap and warm water.
Special Decontamination Considerations

Periodic and thorough disinfection also provides the opportunity to inspect and replace vacuum system parts and tubing, and to disinfect small equipment used in the BSC.

The chairs should also be regularly disinfected, paying particular attention to the areas of the chairs where gloved hands may have touched.