UNIVERSITY OF TEXAS ARLINGTON | ENVIRONMENTAL HEALTH & SAFETY

Ten Tips for Working Safely in Your Biosafety Cabinet (BSC)

Currently 63 certified BSCs are actively used at UTA. There are basic techniques that need to be followed when working with (potentially) biohazardous materials within a BSC to minimize personnel/environmental/sample contamination. If BSCs are not used properly, their protective benefits may be greatly diminished. Please, utilize the 10 tips presented here as the golden rules to assist all laboratory personnel how to work safely with biological agents and materials that may be associated with the dissemination of biohazards.

1. **Know the Airflow of the BSC**

   BSCs create a unidirectional laminar airflow across the work surface following parallel patterns. Particulates are trapped in a high-efficiency particulate air (HEPA) / ultra-low particulate air (ULPA) filter. Class II, Type A2 cabinets maintain a negative pressure inside the cabinet during operation to prevent contaminants from escaping the work area. Incoming air is captured at the opening and passed through a HEPA/ULPA filter. The air flows down to the work surface in a laminar flow pattern where it splits and re-enters the plenum. 70% of the air is recirculated and 30% is passed through another HEPA/ULPA filter to be exhausted to the room. If cabinet's normal airflow pattern is disrupted, airflow alarm will indicate immediate danger to the operator or product. Work in the BSC should cease immediately and the laboratory supervisor should be notified.

   When working in a BSC, items should always be placed at least 4 inches from the front of the cabinet so that they do not block the airflow from the grill. Bulky items should be placed to the side of the cabinet, where they will be less disruptive to the airflow.

2. **Always Work at the Proper Window Sash Height**

   BSCs have a window on the front that protects laboratory workers from the active work area. This is called the window sash. Window sash positioning is among the most vital information about BSCs, and the accuracy of the biosafety process depends on the cabinet maintaining a balance between inflow and down flow velocities. When working within the cabinet and not placing the cabinet's window sash at the certified working level laboratory worker compromises airflow balancing. If the inflow velocity is too strong, contaminated air can enter the sterile work area and contaminate the work. Excessive down flow velocity can push contaminated air from inside the cabinet out into the laboratory without filtering as well as create turbulent air at the work surface causing product contamination.

3. **Never Cover the BSC Air Grill**

The front airflow grill on a BSC performs the important job of separating the clean air inside the cabinet from the contaminated laboratory air outside. The front grill reduces the risk of contaminants entering the sterile work zone and disrupting the experiment inside, and they should never be covered. The front grill, which is located beneath the window sash that separates the laboratory worker from the cabinet contents, can easily be blocked by hands, arms, elbows, consumables, or laboratory equipment. Laboratory workers should avoid resting any parts of their bodies on the grill (elbow rests are advisable to be used to prevent arms from blocking the grill). Blocking the front airflow grill on a BSC raises the risk of the experiment inside being ruined by contaminants from outside and presents the possibility of substances inside the cabinet leaking out into the laboratory. This could present a substantial risk to the safety of laboratory workers and the local environment, particularly if the person using the cabinet is working with infectious materials.

Elbow rests in BSC help to keep airflow steady

4. **Minimize Rapid Movements When Working in a BSC**

Rapid arm movements in a sweeping motion in and out of a BSC can disrupt the air barrier and compromise containment. The best practice is to move hands slowly in and out of the cabinet perpendicular to the front grill, rather than sweeping an arm across the grill, to minimize disruption of the airflow. Operators should organize the workspace inside the cabinet to minimize the movements they have to make with their arms.

Other activities in the laboratory (e.g., walking rapidly past the BSC, opening/closing doors, etc.) may also disrupt the air barrier. A good idea might be to create a working/no walking zone in front of the cabinet e.g., with tape.

5. **Keep First Air Sterile**

   First Air is defined as the undisrupted air coming directly from a HEPA/ULPA filtration source, and it is important to note that many things can disrupt First Air. Laboratory personnel should never place hands above an object on the BSC work surface. Contaminants from a user's hands may compromise the sterility of First Air!

   Maintaining proper airflow within a BSC is critical to the sterility of the worked materials. No items should disrupt the flow of air to the work surface, or to the air grills at the front and rear of the BSC work area.

6. **Reduce Splatter and Formation of Aerosols**

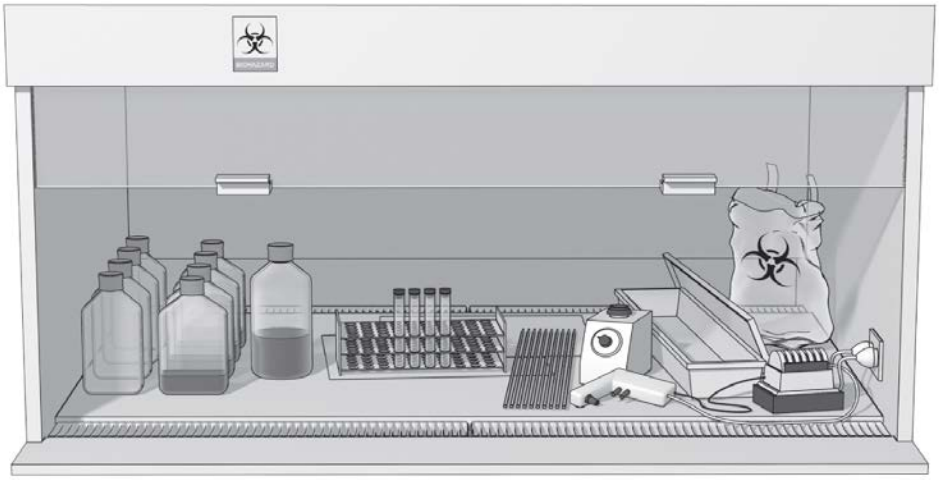
   Many common procedures conducted in BSCs can create splatter or aerosols. Proper aseptic techniques can reduce the generation of splatter and to minimize the potential for personnel exposure to infectious materials manipulated within the cabinet. As a general rule of thumb, keeping clean materials at least twelve inches (31 cm) away from aerosol-generating activities will minimize the potential for cross-contamination.

7. **Know Your Work Area**

   The middle third of the BSC work surface is the ideal area to use. All operations should be performed towards the back of the cabinet away from the user and at least four inches from the front grille. All materials and equipment should be placed inside the cabinet with care to avoid disrupting airflow that can cause turbulence, cross-contamination and/ or breach of containment.

8. **Work from Clean to Contaminated**

   Active work should flow from the clean to contaminated area across the BSC work surface. Materials and supplies should be placed in such a way as to limit the movement of dirty items over clean ones. Maintaining a proper balance of materials from left to right in order to prevent an airflow imbalance within the work zone is important. Bulky items such as biohazard bags, discarded pipette trays, and suction collection flasks should be placed to one side of the interior cabinet. All materials should be placed as far back in the cabinet as practical, toward the rear edge of the work surface and away from the front grille of the cabinet. Similarly, aerosol-generating equipment such as vortex mixers and tabletop centrifuges should be placed toward the rear of the cabinet.



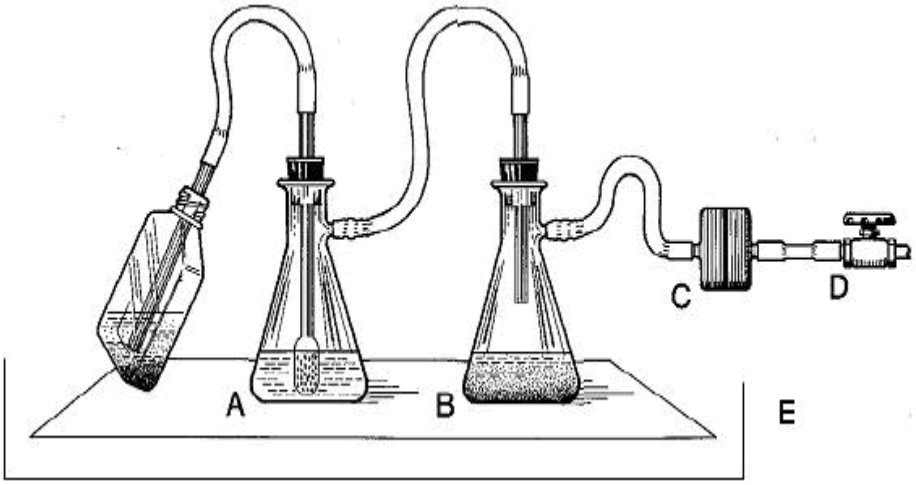
CLEAN ⟶ DIRTY

A typical layout for working “clean to dirty” within a BSCA typical layout for working “clean to dirty” within a Class II BSC. Clean cultures (left) can be inoculated (center); contaminated pipettes can be discarded in the shallow pan and other contaminated materials can be placed in the biohazard bag (right). This arrangement is reversed for left-handed persons.

Source: Adapted from BMBL, fifth edition, Appendix A.

9. **Protect The Central Building Vacuum System and Vacuum Pump**

Aspirator bottles or suction flasks should be connected to an overflow collection flask containing appropriate disinfectant, and to an in-line HEPA or equivalent filter. This combination will provide protection to the central building vacuum system or vacuum pump, as well as to the personnel who service this equipment. Inactivation of aspirated materials can be accomplished by placing sufficient chemical decontamination solution into the flask to kill microorganisms, as they are collected. Once inactivation occurs, liquid materials can be disposed of appropriately as noninfectious waste. Empty the waste from the flask when it reaches no higher than ¾ full. Replace the flask with fresh disinfectant.



The left suction flask (A) is used to collect contaminated fluids into a suitable decontamination solution; the right flask (B) serves as a fluid overflow collection vessel. Flask B is used to minimize splatter. An in-line HEPA filter (C) is used to protect the vacuum system (D) from aerosolized microorganisms. A spill tray (E) should be used when the flasks are outside the BSC.
Source: Adapted from BMBL, fifth edition, Appendix A.

10. **Keep BSC Clean and Disinfected**

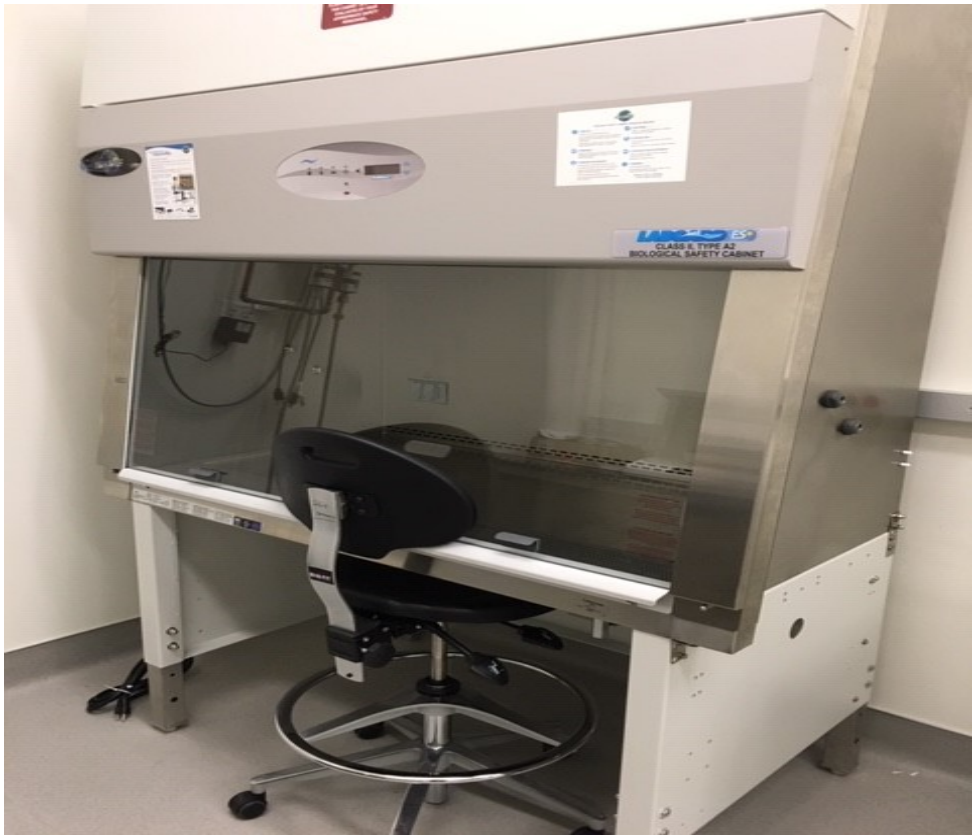
Do not bring potentially contaminated materials out of the BSC until they have been surface decontaminated. Alternatively, contaminated materials can be placed into a closable container while inside the BSC for transfer to an incubator or autoclave area.

For BSC clean up, use disposable lint-free wipes and purified water and for disinfection apply 70% alcohol in the same manner versus using spray bottles to minimize solvent vapor concentrations being re-circulated in the cabinet. NOTE: When bleach (a 1:10 dilution of household bleach, i.e., 0.05% sodium hypochlorite) is used, a second wiping with sterile water is needed to remove the residual chlorine, which may eventually corrode stainless steel surfaces. Other disinfectants can be used as determined by the investigator to meet the requirements of the particular activity.

Wipe down the following BSC surfaces with the appropriate disinfectant for the materials in use:

- Work surface
- Interior walls (except for the supply filter diffuser)
- Interior surface of the window

Record cleaning and disinfection of the BSC in a quality control log. Avoid storing items in BSC when not in use.



BCSs offer added layers of protection for both the product/process on the work surface, the user and the environment. Following above tips when working with a BSC enables everyone in the laboratory to safely carry out experiments and avoid compromising results. To maintain a good safety record within the laboratory, it is advisable to make sure every laboratory worker is briefed on how to work safely in a BSC. Keeping these tips in mind can reduce the potential for contamination and keep laboratory personnel and samples worked with safe!