



# ECKERD COLLEGE

## Safe Operating Procedure

(1/04)

### USING BIOSAFETY CABINETS

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**There is a class IIB3 cabinet in SHB 114.  
It is not a fume hood.**

The following is a modification of information available at the following site:  
<http://www.cdc.gov/od/ohs/biosfty/bsc/bsc.htm>

#### Tips for Using Biological Safety Cabinets

#### **Read the operator's manual and follow all manufacturer's recommendations.**

Our BCS is located in a small room with an exterior and interior door. When using the cabinet be sure to close both doors and indicate that neither door should be opened while the cabinet is in use. The cabinet should be located in an area where it will not be adversely affected by air currents:

- Away from pedestrian traffic.
- Away from other room ventilation devices (e.g., supply diffusers, fans, fume hoods, etc.).

You will need to control the situation to obtain maximum efficacy of the BCS.

Stool height should be adjusted so that the investigator's face is above the front opening.

Movement of arms into and out of the cabinet can disrupt airflow and affect cabinet performance.

- Use a checklist to ensure that all necessary items for a specific activity have been placed in the cabinet prior to starting experimentation.
- Only materials needed for the particular procedure should be in the hood since many items in a hood can cause turbulence and disruption of the airflow.

If it is necessary to move arms in and out of the cabinet, this should be done slowly and with movement perpendicular to the face of the cabinet.

- Do not block front grille by resting arms on them or by putting notes or any other materials on them.
- Manipulation of materials should be delayed for one minute after putting arms in hood to allow the hood environment to stabilize.

Work at least 4-6" from the opening of the cabinet. Work as far back in the cabinet as is feasible since exhaust air is primarily drawn from the back of the cabinet.

**Do not use a bunsen burner in a biosafety cabinet.** The flame can produce eddy currents, which can push infectious agents out of the cabinet towards the worker. In addition, build-up of flammable gases can result in a fire or explosion.

**Personal protective clothing such as a laboratory coat and gloves must be worn when working at the hood.** A solid front, back closing gown provides the best protection. Gloves should be pulled over the knitted wrists of the gown. If two pairs of gloves are worn, put the first pair under and the second pair over the wrist of the gown. Elasticized sleeves can also be worn to protect the investigator's wrists.

**Carry out work on an absorbent pad to contain small spills. Ensure that the pad does not cover the front grille opening.**

- Clean up spills as soon as they occur.
- Remove and disinfect the grill if contaminated.
- When working in a Class II cabinet, ensure that contaminated items or spill clean-up components are immediately removed to the decontamination module or dunk tank as soon as possible.
- Work from "clean" to "dirty" areas.
- Place used pipettes in horizontal trays containing appropriate disinfectant. Dropping pipettes into vertical receptacles creates aerosols.

Place contaminated materials toward the rear of the cabinet.

**Disinfect the cabinet surfaces after use with a disinfectant that is appropriate for the agent in use. Never work inside the cabinet when the UV (germicidal) lamp is on. Be sure to wear UV protective eyewear.**

BSCs are designed to be operated 24 hours per day, and some investigators find that continuous operation helps to control the laboratory's level of dust and other airborne particulates. Although energy conservation may suggest BSC operation only when needed, especially if the cabinet is not used routinely, room air balance is an overriding consideration. In some instances, room exhaust is balanced to include air discharged through ducted BSCs.

Cabinet blowers should be operated at least three to five minutes before beginning work to allow the cabinet to "purge". This purge will remove any particulates in the cabinet. The work surface, the interior walls (not including the supply filter diffuser), and the interior surface of the window should be wiped with a 1:100 dilution of household bleach (i.e., 0.05% sodium hypochlorite); if this is unavailable use a 70% ethanol. When bleach is used, a second wiping with sterile water is needed to remove the residual chlorine, which may eventually corrode stainless steel surfaces. Wiping with non-sterile water may recontaminate cabinet surfaces, a critical issue when sterility is essential (e.g., maintenance of cell cultures).

**Never attempt to remove or change the HEPA filters.**

Leave the fan blower on in the cabinet for a short period of time (5-10 minutes) after finishing work to allow the system to purge. Some safety cabinets should be left on at all times. Check with your supervisor for operation procedures.

Ensure that the cabinet is certified upon installation, modification, movement, and annually.

## Background

Biological Safety Cabinets (BSCs) decrease the risk of airborne infection by reducing the escape of aerosolized infectious agents into the laboratory environment. Some biological safety cabinets also protect the products/cultures within from environmental contamination. These protective features are achieved with High Efficiency Particulate Air (HEPA) filters. Product protection is achieved by passing the supply air for the BSC through a HEPA filter. Only Class II and III BSCs are equipped with supply HEPA filters. Environmental and personnel protection is achieved by passing the exhaust air through one or more HEPA filters. All classes of BSCs are equipped with exhaust HEPA filters. The exhaust design and other factors are used to classify BSCs. The following table summarizes various classes of BSCs and their approved uses.

BSC Class pre-2002	BSC Class post-2002	Risk Group	Design	Product protection
Class I	Class I	2, 3	Minimum face velocity of 75 lfm. Exhaust is HEPA-filtered and may be recirculated to the room or exhausted from the building. Open-front - no sash.	No (no HEPA filtration of supply air)
Class II, Type A	Class II, Type A1	2, 3	Minimum face velocity of 75 lfm. 70% of the air is re-circulated in the cabinet. Exhaust is HEPA-filtered and may be re-circulated to the room or exhausted from the building. Front sash.	Yes (HEPA filtration of supply air)
Class II, Type B1	Class II, Type B1	2, 3	Minimum face velocity of 100 lfm. 30% of the air is re-circulated in the cabinet. Exhaust is HEPA-filtered and discharged from the building. Front sash.	Yes (HEPA filtration of supply air)
Class II, Type B2	Class II, Type B2	2, 3	Minimum face velocity of 100 lfm. No air is recirculated in the cabinet. Exhaust is HEPA-filtered and discharged from the building. Front sash.	Yes (HEPA filtration of supply air)
Class II, Type B3	Class II, Type A2	2, 3	Minimum face velocity of 100 lfm. No air is recirculated in the cabinet. Exhaust flows through two HEPA-filters and is discharged from the building. Front sash.	Yes (HEPA filtration of supply air)
Class III	Class III	3, 4	Totally enclosed, gas-tight, glove ports for manipulation of pathogens. Supply air and exhaust flows through two HEPA filters. No air recirculation within the cabinet and exhaust is ducted to the outside, independent of the building exhaust.	Yes (HEPA filtration of supply air)

Horizontal and vertical laminar flow clean benches are not BSCs. These devices are intended for “clean operations” and do not provide personnel or environmental protection. They are not suitable for handling potentially infectious materials, radionuclides, or chemicals.

**In general, BSCs are not designed for use with chemicals or radionuclides.** HEPA filters are not designed to “trap” chemical vapors. Therefore, cabinets that exhaust into the laboratory can discharge chemical vapors resulting in exposures to laboratory workers and others in the building when the vapors are picked up by the general building ventilation system. HEPA filters, gaskets, and their housing assemblies can also be damaged by some chemicals thereby compromising worker and environmental

protection against infectious agents. BSCs are not designed with intrinsically-safe electrical components (i.e., switches, lights, etc.). Flammable chemical vapors in the cabinet can result in a fire or explosion in contact with these ignition sources. *The low face velocity of Class I and IIA BSCs is insufficient to capture chemical vapors.* Minute quantities of typical biomedical/research chemicals and non-volatile radionuclides can be used in Class IIB1 BSCs. Small quantities of typical biomedical/research chemicals and non-volatile radionuclides can be used in a Class IIB2 and IIB3/IIA2 BSC if the cabinet is exhausted outside of the building, the ducts are under negative pressure or leak tight, and with assurance that the exhaust system is operating properly.