

SAFETY PROCEDURES – GUIDELINES FOR EFFECTIVE USE OF BIOLOGICAL CABINET

This Guidelines/Procedures is intended as a minimum requirement to be applied by operators to protect the user, the environment, the product.

REQUIREMENTS

1. Notify the Biosafety Coordinator before a new biosafety cabinet is to be ordered or an existing cabinet moved to a new location. The proposed location for the cabinet must be known.
2. Notify the Biosafety Coordinator if a biosafety cabinet or clean air bench is to be relocated from another institution. The cabinet must be decontaminated before being moved to laboratories at UWO. Documentation will be required
3. New cabinets or cabinets which have been moved must be recertified after they are installed in the new location.
4. Biosafety cabinets used for work with Level 2 agents, which includes unpreserved human blood and tissues, must be recertified annually by an approved testing service (See section 5 below). OHS must receive copies of the test reports and records will be kept by the Biosafety Coordinator.
5. Natural gas and propane are not allowed to be used inside Class II cabinets.

DEFINITION

BIOLOGICAL SAFETY CABINET

A biological safety cabinet is a ventilated cabinet which uses a variety of combinations of HEPA filtration, laminar air flow and containment to provide either personnel, product or environmental protection or protection of all components against particulates or aerosols from biohazardous agents.

It is distinguished from a chemical fume hood by the presence of HEPA filtration and the laminar nature of the air flow. For fume hood use please see " Laboratory Safety Manual", OHS, 1997; sec 9.3 pp 15,16 Biological safety cabinets must NOT be used for procedures with hazardous chemicals.

1. Types of cabinets

There are three classes of biological safety cabinet:

Class I:

Open fronted cabinets with laminar air flow directed away from the user through a HEPA filter. The cabinet may be ducted to exhaust system or may exhaust into the room. Class 1 cabinets provide partial protection to the user and protection of the environment but no protection to the work.

Class 1 cabinets are suitable for some work procedures at Containment 1 and 2.

Class II types A, B1, B2, B3 :

All Class II cabinets use recirculated HEPA filtered vertical laminar air flow within a partially contained cabinet with a glass sash leaving 8-10 inch work opening. The component of the air flow which is exhausted through HEPA filters may be ducted to the outside or recirculated to the room. Class II cabinets provide a high degree of protection to the worker, the work and the environment.

Suitable for work at Containment Level 1, 2 and 3

Class III:
Totally enclosed cabinets with HEPA filtered supplied air, non-recirculated HEPA filtered laminar flow air over the work surface and hard ducted to outside. The work surface is accessed only through glove ports or sealed air locks. Provides a totally contained area to protect the worker, the work and the environment.

Suitable for work at Containment Level 4 Clean Air Bench:

Reverse HEPA filtered laminar air flow towards worker. Product protection only. NOT a biological safety cabinet. The worker is directly exposed to aerosols and particulates from the work. Not be used for Containment Level 2,3 or 4 agents or radioisotopes. To be used where only a flow of clean air is required.

RECOMMENDATIONS FOR THE USE OF BIOSAFETY CABINETS

1. Purchase

The class of cabinet selected will depend on

- a) the Risk Level of the agent to be used
- b) the degree of protection required for the work
- c) whether volatile chemicals or radioisotopes will be used in the cabinet
- d) The type of procedure to be carried out in the cabinet

Other factors:

- a) cost
- b) the other uses of the room into which the cabinet will be installed
- c) the amount of space available for installation of the cabinet.
- d) size of doorways which cabinet must pass through to the final point of use.

- e) adequate headspace for installation and for the maintenance of the filter boxes, exhaust ducts, alarm systems and fans. There should be, preferably, a 30 cm clearance from the walls and ceiling.
- f) Availability of service connections, such as electricity

The Biosafety Coordinator should be consulted before purchasing a cabinet for installation in University facilities. A University Biohazardous Agents Registry Form must have been completed for all of the agents which will be used in the cabinet.

Physical Plant and Capital Planning Services must be consulted for installation requirements.

The following guidelines refer to Class II cabinets only:

1. Installation and Location of cabinet

The installation of the cabinet in its final location is not usually included in the purchase price. This has to be arranged separately and will incur an extra cost. Cabinets are very heavy and awkward and it is recommended that professional movers are hired to do this. If the cabinet is damaged during the move which is not covered by insurance, the repairs may result in more expense. The correct location of the cabinet in the room in which it is to be operated will improve the efficiency of operation of the cabinet. The cabinet should be situated away from doors, windows and main traffic areas in the lab which could create air currents which can disrupt the laminar flow characteristics inside the cabinet. The cabinet should be installed with at least 30 cm space on either side and behind the cabinet. A minimum of 40 cm should be available between the top exhaust filter and the ceiling to allow access for certification. (ref: CSA Standard Z316.3). The cabinet must not be situated directly under or adjacent to the room air supply as the exhaust air from the cabinet will disrupt the air supply. Also note that these cabinets generate heat and situating two or more in a small room may require specialized ventilation and heat control.

1A. Relocation of Cabinets

The Biosafety Coordinator must be informed before a biological safety cabinet is relocated.

Generally this will involve a decontamination with formaldehyde before the cabinet is moved and recertification after relocation as required in NSF Standard 49 and CSA Standard Z316.3.

If a cabinet is to be relocated to a laboratory at UWO from a laboratory at another institution, documentation will be required by the Biosafety Coordinator to prove decontamination at the previous location before the cabinet can be moved to a laboratory at UWO. Recertification will then be required at UWO after the cabinet is installed.

2. Maintenance and cleaning

The stainless steel surfaces inside the cabinet should be wiped with 70% ethanol or isopropanol on completion of work or after any spill. A small amount of detergent may be added to the ethanol to assist in the removal of soil. Bleach is not recommended for this use as it may lead to corrosion of the steel surfaces unless they are thoroughly rinsed after the bleach has been used. Periodically the work surface should be lifted (with the cabinet running) and the spill area underneath should be cleaned and disinfected. Spilled medium in this area will allow microbial growth and can be a source of contamination.

3. Certification

New biological safety cabinets MUST be certified by an approved certification company on installation and before use. Please consult the Biosafety Coordinator to arrange this.

4. Recertification

Biological safety cabinets which are being used for work with agents at Containment Level 2 or 3 MUST be recertified annually. (Laboratory Biosafety Guidelines: Health Canada, 5.2.2., 2nd ed. 1996. NSF Standard 49, CSA Standard Z316.3.). Cabinets MUST also be recertified if they are moved to a new location.

Recertification will usually be preceded by cabinet decontamination by formaldehyde gas for 4 hours.

HEPA filtered infected animal enclosures MUST also be recertified annually.

The Biosafety Coordinator co-ordinates recertification of cabinets in use on campus at UWO twice a year in January and August. Recertification can be arranged at other times by the researcher provided that an approved company is used. In this case copies of the test reports must be sent to the Biosafety Coordinator in Stevenson Lawson Rm 60. Recertification is also recommended annually for biological safety cabinets which are being used to maintain sterile work conditions for agents at Level 1, but is not mandatory. Leaks in or damage to the HEPA filters may result in increased microbial contamination in the work area. Over time the HEPA filters become loaded with dust and contaminants and although this will not compromise the sterility of the air, it may result in low air flow problems which may compromise the sterility of the work area and lead to a shortening of the life of the electric motor. Clean air enclosures for SPF or immune compromised animals should also be recertified annually.

5. The use of gas burners inside the cabinet.

Gas burners (natural or propane) are NOT PERMITTED for use inside a Class II biological safety cabinet because if the burner goes out gas may build up inside the cabinet resulting in an explosive atmosphere. The heat from the flame will disrupt the laminar air flow pattern and may result in leakage of microbial agents from

the work space into the laboratory and also allow contaminants to enter the sterile work space. Alcohol burners may be used with caution inside the cabinet but the volume of alcohol used must be kept to a minimum. The alcohol should be in a metal (not glass) container. Alternative techniques for sterile work include using sterile pasteur pipettes as an aspirator and using electronic bacterial loop incinerators. Sufficient equipment should be available so that a fresh supply of sterile equipment replaces the need to flame items.

Where gas lines have been permanently or temporarily connected to a cabinet, these must be disconnected.

6. The use of Ultraviolet light inside the cabinet

UV irradiation produced by germicidal lamps can be used to decontaminate the work surfaces inside a cabinet since UV irradiation is effective in killing many microorganisms. There are, however, several factors to take into consideration before using a germicidal lamp as an aid to maintaining sterile conditions inside the cabinet.

a) The 253.7 nm wavelength emitted by the germicidal lamp has limited penetrating power and will not penetrate soil or dust. It is thus only effective against microbes in the air in the cabinet or exposed on the work surface.

As soon as the cabinet is turned on the clean air produced in the cabinet by the UV is replaced by sterile air which has passed through the HEPA filter and therefore the gain is brief.

b) The intensity or destructive power of the radiation decreases as the square of the distance from the lamp.

Thus, exposure time is related to the distance from the lamp.

c) The intensity of the lamp also decreases with time. Lamp intensity must be checked with a meter if the sterilizing capacity of the UV is to be verified.

d) The intensity of the lamp is drastically affected by an accumulation of dust and dirt on the lamp. Therefore the lamp must be cleaned at least weekly. The lamp must never be on while an operator is working at the cabinet. The UV light will reflect off the steel surfaces and some will reflect out into the room through the work opening, the lamp must be turned off if a worker is in the same room. Contact Occupational Health and Safety if you need a biological safety cabinet certified.