Laboratory
Biological Safety Cabinet
and
Fume Hood
Manual

For NEBRASKA MEDICINE and UNMC

SERIOUS MEDICINE. EXTRAORDINARY CARE.

BREAKTHROUGHS FOR LIFE.

Table of Contents

Introduction

This manual will cover two basic types of equipment:

- Biological Safety Cabinet (BSC)
- Fume Hood

Biological Safety Cabinets are designed to minimize dangers inherent in work with biological materials assigned to biosafety levels 1, 2, or 3. These cabinets also provide physical isolation and containment of biological hazards and their by-products. **BSC's offer product, personnel, and environmental protection.**

Fume Hoods are designed to protect laboratory personnel by capturing, conveying, and/or containing contaminants such as chemical vapors, gases, dust, mists, and fumes before they escape into the laboratory environment. **Fume Hoods offer personnel protection only.**

This Laboratory Manual has been developed to provide information regarding the selection, performance, safety, and inspection of fume hoods and biological safety cabinets.

1. Biological Safety Cabinet General Description

A1	Class II, Type A1	• 70% air recirculated; 30% exhausted from a common plenum to the room; • 75FPM intake; • may have biologically contaminated positive pressure plenum
A2	Class II, Type A2	• 70% air recirculated; 30% exhausted from a common plenum to the room; • 100FPM intake; • biologically contaminated plenum under negative pressure or surrounded by negative pressure
A2	Class II, Type A2 With Canopy	• 70% air recirculated; 30% exhausted from a common plenum to a facility exhaust system; • 100FPM intake; • biologically contaminated plenum under negative pressure or surrounded by negative pressure
B1	Class II, Type B1	• 40% air recirculated; 60% exhausted from cabinet; • exhaust air pulled through dedicated exhaust duct into facility exhaust system; • 100FPM intake • all biologically contaminated plenums are negative to the room or surrounded by negative pressure plenums
B2	Class II, Type B2	• 0% air recirculated; 100% exhausted from cabinet exhaust air pulled through dedicated exhaust duct into facility exhaust system; • 100FPM intake • all ducts and plenums are under negative pressure • all contaminated ducts are under negative pressure or surrounded by directly exhausted negative pressure ducts or plenums

Note:

- Class A1 BSC's with positive pressure plenums are not allowed on site.
- Class A2 BSC's WITH CANOPY (ducted) must not be hard connected. Must have a canopy connection allowing room air to be drawn into the exhaust duct.
- An audible/visual alarm is required to warn of exhaust system failure.

2. Biological Safety Cabinet Selection

Class II, Type A2 Cabinet

- Cabinet air may be recirculated back into the laboratory or ducted out of the building by means of a "thimble or canopy" connection (i.e., a small opening around the cabinet exhaust filter housing) whereby the balance of the cabinet is not disturbed by fluctuations in the building exhaust system. The canopy must be designed to allow for proper certification of the cabinet (canopy must provide access to permit scan testing of the HEPA filter).
- Provides a microbe free work environment necessary for cell culture propagation.
- Type A2 cabinets used for work with minimal amounts of volatile chemicals and radionuclides required as an adjunct to microbiological studies must be exhausted through a properly functioning exhaust canopy with alarm.

Class II, Type B1 Cabinet

- Hard ducted through a dedicated duct exhausted to the atmosphere after passage through a HEPA filter.
- Maintain a minimum average face velocity of 100 FPM.
- Recirculate 30% of the air within the cabinet.
- Suitable for work with volatile chemicals and radionuclides required as adjuncts to biological studies.

Class II, Type B2 Cabinet

- Does not recirculate air within the cabinet.
- Maintain a minimum average face velocity of 100 FPM.
- Hard-ducted through a dedicated duct exhausted to the atmosphere. 100% of cabinet air is exhausted out of the building after passage through a HEPA filter. Contains negative pressure plenums.
- Suitable for work with volatile chemicals and radionuclides required as adjuncts to biological studies.
- The cabinet internal fan is interlocked to shut down when the building exhaust fan fails to prevent pressurization of the cabinet.

Class III Cabinet

- Class III cabinets are totally enclosed and gas-tight with HEPA filtered supply and exhaust air.
- Work is performed with attached long-sleeved gloves.
- The cabinet is kept under negative pressure of at least 120 Pa (0.5 in. w.g.) and airflow is maintained by a dedicated exterior exhaust system.
- Class III cabinets protect the worker and the product. They are designed for work with level 4 pathogens and provide an alternative to the positive-pressure suit made for maximum containment laboratories.
- The exhaust air is double HEPA filtered or treated by HEPA filter and incineration.
- Removal of materials from the cabinet must be through a dunk tank, double door autoclave or air-lock pass-through for decontamination.
- Interlock or protocols must be used for the autoclave and pass-through doors to prevent both doors from being open at the same time.

3. Procurement of a BSC

Before purchasing a BSC, the manufacturer and model should be approved by the UNMC Bio Safety Equipment Engineer or Research Resource Manager.

A form is available online to assist in purchasing, installation, relocation, and decommissioning of a BSC or fume hood.

Link to the form:
https://info.unmc.edu/safety/safety-office/PURCHASE_INSTALL_RELOCATE_DECOMMISSION_HOOD_BSC_LFB_FORM.pdf

There are some BSC's that do not meet NSF/ANSI 49. The most current version of **"NSF/ANSI 49 – Biosafety Cabinetry: Design, Performance, and Field Certification"** is the industry standard all BSC's and certifications must follow.

Once BSC is approved, your department will be able to order.

Prior to delivery of a new BSC, a work request must be sent to UNMC Facilities Management & Planning (7100). The work request should include the department's cost center #, building, and room the BSC is to be installed. Any special requests, such as vacuum to be run to the new BSC, should be included. Facilities Management & Planning can be contacted by FAX 402-559-8282 or PHONE 402-559-4050.

Note:

- Before selecting a location for a new BSC, consider the space around the BSC. The BSC should be installed as far as possible from ceiling diffusers, doors, high traffic areas, fume hoods, and windows that may be opened. Allow enough space around the BSC to access removable panels and control panels. Sufficient height (12") must be maintained above the BSC to scan the exhaust HEPA filter during annual certification. Many BSC's require a 20 amp wall outlet. Check in advance to see if a 20 amp outlet is located in the area the new BSC is to be installed.

- Upon arrival of a new BSC onsite, the new BSC will be delivered to the dedicated lab by our hood contractor on the date requested. The hood contractor is under contract by UNMC. After installation, the BSC will be certified by the hood contractor or the UNMC Bio Safety Equipment Engineer. After certification, the new BSC will be entered into a database. UNMC will schedule annual certifications based on the initial certification date.

4. Certification

Certification is to be performed annually, whenever a BSC is moved, or after repairs. Certification will be performed by a certifier that has met the requirements of the NSF International Class II Biosafety Cabinet Field Certifier Accreditation Program. Bio Safety Cabinets are certified in accordance of the latest version of NSF/ANSI 49, Annex F.

Field certification tests include:
- down flow velocity profile test
- inflow velocity test
- HEPA/ULPA filter leak test
- air flow smoke patterns test
- noise level tests
- site installation assessment tests

Note:

- Site installation assessment tests include alarm functions, blower interlock, and exhaust system performance (proper exhaust duct negative pressure and canopy performance).

- Optional tests related to worker comfort and safety, **not** performed are: lighting intensity, vibration, electrical leakage, ground circuit resistance, and polarity tests.

5. BSC Policies for NE Medicine and UNMC

Decontamination:
BSC decontamination at UNMC is achieved by using the chlorine dioxide (CD) method. The BSC should be emptied before the four hour decontamination process.

BSC must be decontaminated:
1) Before it is opened up for repair
2) Before it is moved to a new location
3) Before it is taken out of service

Noncompliance BSC's:
A BSC in need of repair must be addressed in a timely manner. If the repair of a BSC is not addressed in a timely matter, a report will be filed with the Safety Office. A yellow notice shall be placed on the front of a BSC indicating why it does not meet current standards. *(see an example in this manual: page 16 - 12.3 Sample Fail Notice.)* A non-functioning BSC will be removed from the space or sealed, if repairs are not scheduled. All BSC's must be currently certified and in full working order i.e., fluorescent lamps and failed sash springs.

BSC Decommissioning:
A decommissioning form from The Nebraska Medical Center Policies and Procedures Manual shall be implemented in the process of removing a BSC to insure complete communication of contaminates and decontamination have been addressed by all individuals involved.

A form is available online to assist in purchasing, installation, relocation, and decommissioning of a BSC or fume hood.

Link to form:
https://info.unmc.edu/safety/safety-office/PURCHASE_INSTALL_RELOCATE_DECOMMISSION_HOOD_BSC_LFB_FORM.pdf

Work Requests:
All work requests must be submitted at least one week in advance.
Shipping times for BSC parts varies among manufacturers. Most parts are 7-10 working days out.

Recommendations:
Shut off all BSC's and close the sash at the end of the work day. This will save energy and prolong the life of the BSC HEPA filters and motor.

6. Biological Safety Cabinet Safety

Do not confuse laminar flow cabinets with biological safety cabinets. Laminar flow cabinets can look very similar to biological safety cabinets, but they only protect samples inside the work zone from external airborne contamination. Laminar flow cabinets do not protect the user.

Do not use the cabinet as a storage area. Overloading the cabinet with unnecessary items can affect cabinet airflow and containment.

A Bunsen burner shall not be used in the cabinet. The resulting buoyancy effects will affect cabinet airflow and containment. When absolutely necessary, electric burners may be used.

Open flames in a BSC:

- Create turbulence in the airflow compromising protection of both the worker and the work.
- Present a potential fire or explosion hazard, especially when using a gas burner in conjunction with ethanol.
- Cause excessive heat build-up which may damage HEPA filters and compromise the cabinet's integrity.
- May inactivate the manufacturer's warranty.

BSCs should not be used in place of a chemical fume hood. Volatile or toxic chemicals should not be used in Class II Type A cabinets without ducting because vapor build-up inside the cabinet presents a fire or explosion hazard. In addition, this type of cabinet recirculates air from the cabinet work space into the room, potentially exposing the operator and other room occupants to toxic chemical vapors

Minimize disturbances to the airflow barrier. Work in a controlled and steady manner. Avoid rapid movements.

Avoid overloading the work surface. Work within the safe areas. Avoid blocking the air pathway through the grills in the front and back of the work surface area. **Tall items and/or pipet-aid mounts on the side and back walls seriously disrupt the air flow and can compromise the function of the BSC.**

Observe proper surface decontamination. Decontaminate the work zone with cleaning agents before and after every use. The germicidal UV-lamp is not a substitute for good cleaning practices.

Note:

- **A BSC is not a hood!** A Fume Hood is a hood. A BSC is commonly referred to as a hood but should be identified as a Cabinet, BSC, or Biological Safety Cabinet.

- A laminar flow cabinet designed to protect product only is most commonly referred to as a Clean Bench, Laminar Flow Clean Bench, or Sterile Work Station.

- Certification tags are hole punched identifying the month and year **when tested** and type of equipment.

7. Fume Hood General Description

Definition:
A Laboratory Fume Hood is a safety device specifically designed to carry undesirable effluents (generated within the Hood during a laboratory procedure) away from laboratory personnel and out of the building when connected to a properly designed laboratory ventilation system. A Laboratory Fume Hood shall be made primarily from flame resistant materials including the top, three fixed sides, and a single face opening. Face opening is equipped with a sash and sometimes an additional protective shield. Face opening will have a profiled entry and usually an airfoil designed to sweep and reduce reverse airflows on the lower surface. A Laboratory Fume Hood will be equipped with a baffle and in most cases a bypass system designed to control airflow patterns within the hood and manage the even distribution of air at the opening. The bypass system may be partially blocked to accommodate Variable Air Volume (VAV) Systems. A Laboratory Fume Hood is given here as the proper terminology. Other widely used terms include --- Fume Hood, Chemical Hood, Chemical Fume Hood, and Hood. A Laboratory Fume Hood will be set on a bench, a pedestal or on the laboratory floor.

Constant Volume: The conventional hood is a basic enclosure with an interior baffle and movable front sash. The conventional hood generally operates at a constant exhaust volume with the majority of exhaust air entering the hood through the sash opening. Closing the sash increases the speed of the air through the sash opening so that high face velocities are to be expected with the sash in the near closed position. The conventional hood is generally the least expensive but its performance depends largely on sash position. With the sash in the near closed position, high velocity air passing through the sash opening can damage fragile apparatus, disturb instrumentation, slow distillation rates, cool hot plates, and disperse valuable sample materials or result in turbulence inside the hood.

By-Pass: The by-pass hood generally operates at a constant volume and is designed so that as the sash is closed the air entering the hood is redistributed thereby minimizing the high velocity air streams encountered in conventional hoods. The by-pass openings above and below the sash area reduce fluctuations in face velocity as the sash is raised or lowered. Therefore the face velocity in by-pass hoods does not generally reach levels which might be detrimental to lab fume hood procedures. By-pass type hoods comprise the majority of hoods on the market.

Auxiliary Air: This fume hood sometimes referred to as a makeup air fume hood, was developed as a variation on the bypass fume hood and reduces the amount of conditioned room air that is consumed. The auxiliary fume hood is a bypass hood with the addition of direct auxiliary air connection to provide unconditioned or partially conditioned outside makeup air. Auxiliary air hoods were designed to save heating and cooling energy costs but tend to increase the mechanical and operational costs due to the additional ductwork, fans, and air tempering facilities. In general, installation of this type of hood is discouraged since the disadvantages usually outweigh the benefits. **Auxiliary air fume hoods are not allowed onsite.**

Variable Air Volume (VAV): A VAV hood maintains a constant face velocity regardless of sash position. To ensure accurate control of the average face velocity, VAV hoods incorporate a closed loop control system. The system continuously measures and adjusts the amount of air being exhausted to maintain the required average face velocity. The addition of the VAV fume hood control system significantly increases the hood's ability to protect against exposure to chemical vapors or other contaminants. Many VAV hoods are also equipped with visual and audible alarms to notify the laboratory worker of hood malfunction or insufficient face velocity.

Walk-in Hood: A walk-in hood is a hood which sits directly on the floor and is characterized by a very tall and deep chamber that can accommodate large pieces of equipment. Walk-in hoods may be designed as conventional, bypass, auxiliary air, or VAV.

Perchloric Acid: A perchloric acid hood has the general characteristics of a bench-top hood; however, the interior lining must be coved and welded seamless stainless steel (other non-reactive material such as CPVC or polypropylene have been used when heat is not a concern). Non-reactive and corrosion resistant material should extend all the way through the exhaust system. In addition, the hood, duct, and fan must have a water wash down system to remove perchlorates and prevent the build-up of potentially explosive perchlorate salts. Drain outlet shall be designed to handle a minimum of 15 gallons per minute. The work surface on perchloric acid hoods typically has a water trough at the back of the hood interior under the baffle. The fume hood liner in a perchloric acid fume hood shall have no access holes such as those which may be used for plumbing access. Access panels should be considered in the lab layout for access through the hood exterior. In nearly all other respects however, the design of a perchloric acid hood is the same as conventional or bypass fume hoods. A perchloric acid hood shall never be tied to a manifold system.

Sash is the term used to describe the movable glass panel that covers the face area of a fume hood. Sashes can be vertical, horizontal, or a combination of the two. Many hoods are installed with a vertical sash stop which stops the sash at approximately an 18" inch work level. Sash stops should never be removed, overridden, or modified. It is recommended that all lab work in a properly functioning fume hood be performed at the sash stop level or lower whenever possible.

Alarms, sensors, controls, and gauges may be installed on many of the newer VAV hoods. These features provide lab personnel with a constant reading of fume hood performance. If the face velocity falls below an acceptable range the hood sensors will trigger an alarm to notify lab personnel. Low velocity alarms activate when the sash has been raised to a height at which the hood can no longer exhaust a sufficient amount of air, the building air exhaust system is not working properly, or there has been a power outage. When a low velocity alarm is activated, no hazardous chemical work should be performed until the exhaust volume is increased. Additionally, laboratory workers should not attempt to stop or disable hood alarms. UNMC's Facilities Management & Planning office (402-559-4050) or Nebraska Medicine Facilities Management & Planning office (402-552-3347) should be notified for adjustment of air handling system exhaust and fume hood maintenance.

8. Procurement of a Fume Hood

Before purchasing a Fume Hood, the manufacturer and model must be approved by Facilities Management & Planning. A fume hood is part of the HVAC system, so an air survey may be performed prior to the engineer's final approval.

A form is available online to assist in purchasing, installation, relocation, and decommissioning of a BSC or fume hood.

Link to the form:
https://info.unmc.edu/safety/safety-office/PURCHASE_INSTALL_RELOCATE_DECOMMISSION_HOOD_BSC_LFB_FORM.pdf

Once fume hood is approved, your department will be able to order.

Prior to installation of a new fume hood, a work request must be sent to Facilities Management & Planning. The work request should include the department's cost center #, building, and room the fume hood is to be installed. Any special requests should be noted. Facilities Management & Planning can be contacted by phone at Nebraska Medicine 402-552-3347 or UNMC 402-559-4050.

9. Certification

Fume hoods are certified in accordance to the guidelines of **"SEFA 1-2010" Recommended Practices for Laboratory Fume Hoods.** Fume hoods are certified annually. Fume hood contents should be removed for testing.

10. Fume Hood Policy

Air is drawn in through the opening face of the hood, where the user works, and out through the exhaust. The constant flow of air inwards towards the face prevents hazardous vapors, fumes, and particles from escaping out through the hood opening, keeping the user and other laboratory workers safe.

Facial flow velocity must be high enough for the hood to be effective. A low flow velocity allows harmful fumes, vapors, or particles to escape through the opening of the hood toward the user. One cause of low flow velocity is having a sash, too high. When this happens a low flow velocity alarm and sash height alarm may sound. Here are the rules for safe hood operation:

1) Never put your head into the hood space, since inserting your head inside the hood can expose you to harmful materials. Only your arms should be present in the hood.
2) Wear appropriate personal protective equipment (PPE) at all times, regardless of the protection afforded by the fume hood. Contact the Environmental Health & Safety (EHS) office for proper PPE recommendations, if they are unknown.
3) Place all materials in the fume hood at least 6 inches away from the edge of the fume hood face. When work is carried out within 6 inches of the edge, vapors, fumes, and particles are more likely to escape.
4) When not in use and at night, the sash should be closed. Closing the sash ensures a safer working environment for all in the laboratory. Additionally, this saves energy.
5) Do not tamper with any fumehood alarms and/or sensors.
6) Do not use the hood for chemical storage. Store chemicals in appropriate locations, such as a flammable cabinet, and bring them into the fume hood only when needed.
7) If collecting flammable waste the waste containers must be put in the flammable liquids cabinets at the end of the workday.
8) Do not store or stack large objects in a fume hood that can restrict air flow.
9) Do not disrupt fume hood air flow by placing objects close to the hood such as refrigerators and/or shelves. Also, do not hang items such as lab coats on the fume hood accessory handles.
10) Do not place items on top of the airfoil.
11) Always work with the sash height at or below the maximum safe height. 18" is the maximum working sash height allowed unless approved by the Bio-Safety Equipment Engineer.
12) Face velocity set point is 100 FPM, 90 FPM minimum.
13) A fume hood must be emptied and thoroughly cleaned prior to a lab moving out of the space.

Note:

- Fume hood roof exhaust stacks must extend at least ten feet above the roof or at least two feet above the top of a parapet wall, whichever is greater.
- Discharge must be directed vertically upward.

11. Definitions

Baffle – Panel or panels located at the rear of a fume hood interior that aid in distributing the desired air flow pattern of air moving into and through the hood.

Biohazard - A risk to human health or the environment arising from biological work, especially with microorganisms.

Cabinet – Biological Safety Cabinet (BSC)

Canopy – A cover over the HEPA filter on top of a BSC allowing access to the HEPA filter and for room air to exhaust through slots.

Clean Bench – Laminar flow workstation for product protection only.

CFM – Cubic Feet of air per Minute (Volume)

Damper – A device installed in a duct to control air flow volume.

Decontamination – Removal or destruction of infectious agents. Removal or neutralization of toxic agents.

Diffuser – Supply air register in the ceiling.

Face Velocity – The speed of air (in FPM) entering the work surface of a fume hood or biosafety cabinet.

FPM – Feet of air Per Minute (Velocity)

HEPA – High-Efficiency Particulate Air filter. A filter designed to remove 99.97% of airborne particles measuring 0.3 microns or greater in diameter passing through it. Most BSC's contain 99.99% eff. HEPA filters.

Hood – Fume Hood

IEST – IEST-RP-CC002.3. Institute of Environmental Sciences and Technology. Unidirectional-Flow, Clean-Air Devices. Testing standard used on laminar flow clean benches.

Laminar Air Flow – Unidirectional air flow through the work area often referred to as turbulence free steady air flow.

NSF/ANSI 49 - National Sanitation Foundation, Biosafety Cabinetry: Design, Construction, Performance, and Field Certification of Class II (laminar flow) biosafety cabinetry, which is designed to minimize the hazards inherent in working with biosafety agents of levels 1, 2, 3, or 4.

Pa - The unit of measurement called standard atmosphere (atm) is defined as 101.325 kPa and approximates to the average pressure at sea-level at 45° N. Meteorological reports typically state atmospheric pressure in hectopascals.

Plenum - A chamber intended to contain air, gas, or liquid at positive or negative pressure.

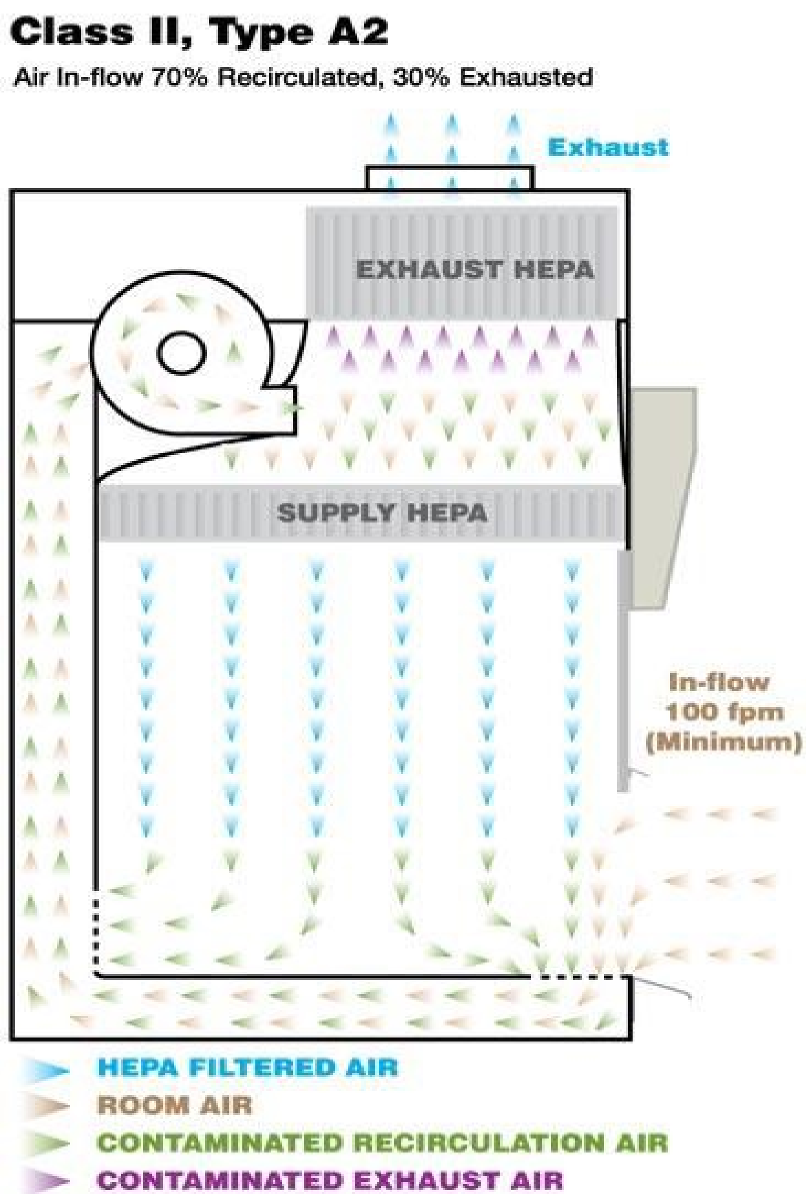
Sash – A moveable transparent panel set in a fume hood or biosafety cabinet entrance to form a protective shield and control face velocity.

SEFA 1-2010 - S.E.F.A. Scientific Equipment and Furniture Association. SEFA 1-2010 *Laboratory Fume Hoods Recommended Practices.* Standard used for testing fume hoods.

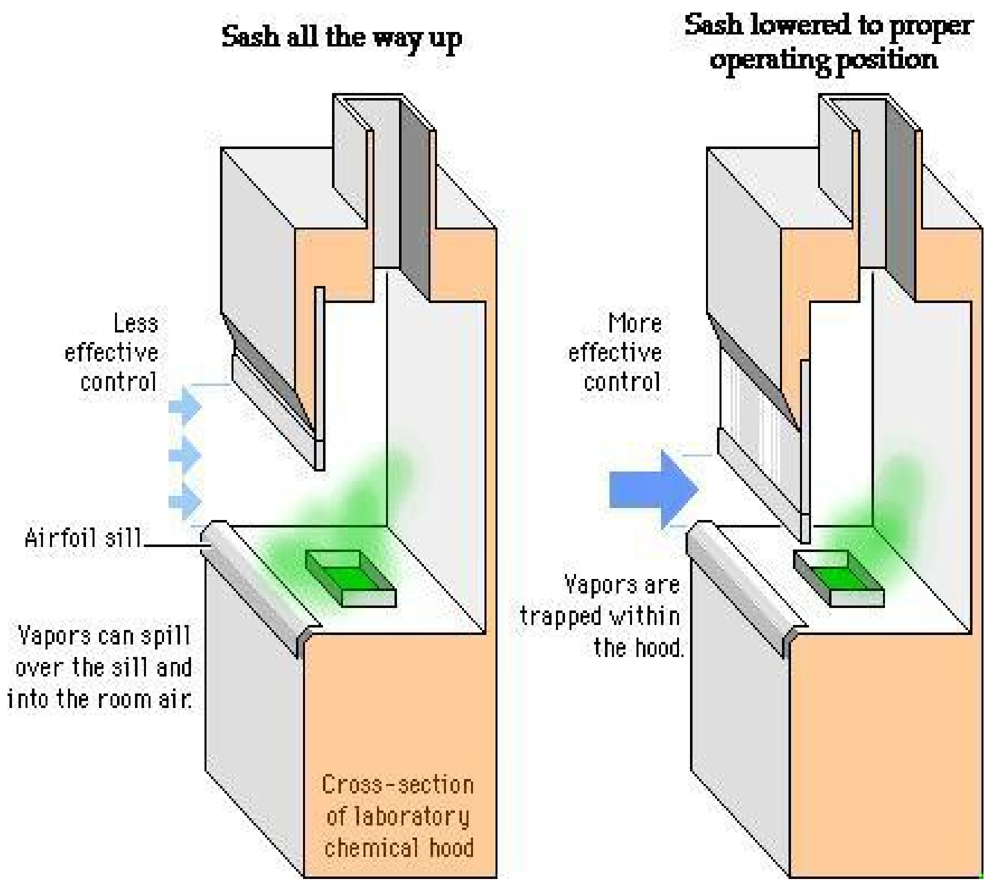
w.g. - inches of water / **wc** - inch water column (water gage), in Aq, or in H_2O is a non-SI unit for pressure. The units are by convention and due to the historical measurement of certain pressure differentials. It is used for measuring small pressure differences across an orifice, pipeline, air duct, or shaft.

12. Illustrations

12.1 Typical Class II, Type A2 Biological Safety Cabinet



12.2 Typical Laboratory Fume Hood



12.3 Sample of a Fail Notice if unit fails inspection

NOTICE

- HEPA Filters Are Plugged -

THIS EQUIPMENT DOES NOT MEET

CURRENT STANDARDS

USE OF THIS EQUIPMENT COULD BE HAZARDOUS

PLEASE SEND WORK REPAIR REQUEST TO
UNMC FACILITIES MANAGEMENT & PLANNING

402-559-4050

For any questions Contact ----- ----- in the Bio Safety Office
Cell: (000) 000-0000