Working Safely in Your NuAire Biological Safety Cabinet

an interactive guide
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Introduction

NuAire Biological Safety Cabinets are designed to provide personnel, environmental and product protection when appropriate practices and procedures are followed. Biological Safety Cabinets are only one part of an overall biosafety program which requires consistent use of good microbiological practices. This interactive guide is intended to give you more information about:

- Basic Use: Understanding the Functions of the BSC
- Using Proper Procedures and Techniques with the BSC
- Proper Preparation: The 4 Steps to Take Before You Begin Work in Your NuAire BSC
- Personal Hygiene Practices for Working Safely in a Biological Safety Cabinet
- Working With and In the Biological Safety Cabinet
- When Work is Completed
- Maintenance and Certification
Biological Safety Cabinets are the most widely used and accepted primary containment devices. Three kinds of Biological Safety Cabinets, designated as Class I, II and III, have been developed to meet varying research and clinical needs.

NuAire Class I or II cabinets can provide partial containment, and Class III cabinets have the potential for complete containment.

Biological Safety Cabinets must not be confused with other laminar flow devices or 'clean benches'; in particular, horizontal flow cabinets which direct air towards the operator should not be used for handling infectious, toxic or sensitizing materials.

For more information on classifications and types, use this link to see the Biological Safety Cabinet Types and Classes Chart on the NuAire Web site.
Chapter 1

Basic Use: Understanding the Functions of the Biological Safety Cabinet

From the earliest laboratory-acquired typhoid infections to the hazards posed by today's antibiotic-resistant bacteria and rapidly mutating viruses, threats to worker safety have stimulated the development and refinement of cabinets in which infectious microorganisms and potentially harmful aerosols could be handled safely. There has also been increasing demand to protect work in progress from particulate contamination, especially in the areas of microtechnologies.

Biological Safety Cabinets use High Efficiency Particulate Air (HEPA) filters in their exhaust and/or supply systems to protect personnel and products within the BSC from contamination from aerosols and particulates. They also protect the environment by isolating and containing the work in progress within the BSC.
In order for the BSC to do its job, laboratory personnel must be trained in the correct use and maintenance of biological safety cabinets, and follow the guidelines to ensure that personnel, product and environmental protection are maintained.

Your NuAire LabGard™ Biological Safety Cabinets will meet your laboratory requirements for life science research, clinical or industrial applications. Each comes with NuAire’s legendary reputation for a commitment to quality and dependability, and backed by a level of customer service available only from NuAire.
These are a few of the unique benefits of owning a NuAire Biological Safety Cabinet. Touch on any of the underlined text to link directly to the NuAire Web site for additional information. (This requires an active Internet connection from your computer, and a properly installed Internet browser, such as Netscape Navigator or Internet Explorer.)

**Work Better**
- Better Protection for Personnel, Product and Environment
- Exceptionally Quiet
- Easy, Efficient Operation
- Less Maintenance

**Designed Better**
- Ergonomically Designed for Comfort
- Flexible Configuration
- Safety Features
- Options and Accessories
Benefits of owning a NuAire Biological Safety Cabinet (continued).

**Built Better**
- True Laminar Airflow & HEPA Filters
- HEPEX™ Zero Leak Airflow System

**Backed Better**
- 36 Month Warranty is the longest and most comprehensive Warranty on Products of this kind in the world.
- Worldwide Customer Service
Airflow of an A/B3 Biological Safety Cabinet.

Click on the above frame to see the airflow demonstration video.
Purpose: What the BSC Does & Doesn’t Do

Biological Safety Cabinets were designed to remove or minimize exposures to hazardous biological materials and hazardous aerosols. The Biological Safety Cabinet is the principal device used to provide containment of infectious splashes or aerosols generated by many microbiological procedures.

Properly maintained Biological Safety Cabinets are used whenever:

a. Procedures with a potential for creating infectious aerosols or splashes are conducted. These may include centrifuging, grinding, blending, vigorous shaking or mixing, sonic disruption, opening containers of infectious materials whose internal pressures may be different from ambient pressures, inoculating animals intranasally, and harvesting infected tissues from animals or eggs.

b. High concentrations or large volumes of infectious agents are used.
What the BSC Does & Doesn’t Do (continued)

A Class II Biological Safety Cabinet, combined with proper microbiological technique, provides primary containment for low to moderate risk microorganisms. This containment is accomplished by laminar airflow and HEPA (high efficiency particulate air) filtration. With proper technique, the Class II Biological Safety Cabinet provides protection to the product, the worker and the environment.
Chapter 2

Using Proper Procedures and Techniques with the Biological Safety Cabinet

The use of proper procedures and equipment cannot be overemphasized in providing primary personnel and environmental protection. The Biological Safety Cabinet is not a substitute for good laboratory practice. Aerosols can escape. Chemical vapors will pass through HEPA filters and most biological safety cabinets do not provide protection from toxic chemicals or radionuclides.

Location of the Cabinet

Proper use begins with the proper location and installation of the BSC. Biological Safety Cabinets were developed as workstations to provide personnel, product and environmental protection during the manipulation of infectious microorganisms and hazardous aerosols. In order to accomplish the task, location considerations must be met to ensure maximum effectiveness of these primary barriers.
The Ideal Location

The ideal location for the Biological Safety Cabinet is remote from the entry (e.g., the rear of the laboratory away from traffic), since people walking parallel to the face of a BSC can disrupt the air curtain. The air curtain created at the front of the cabinet is quite fragile, amounting to a nominal inward and downward velocity of 1 mph. Open windows, air supply registers, or laboratory equipment that creates air movement (e.g., centrifuges, vacuum pumps) should not be located near the Biological Safety Cabinet. Similarly, chemical fume hoods must not be located close to Biological Safety Cabinets.
Clearance Around the BSC

Whenever possible, a 12-inch clearance should be provided behind and on each side of the cabinet to allow easy access for maintenance, and to ensure that the air return to the laboratory is not hindered. A 16- to 18-inch clearance above the cabinet may be required to provide for accurate air velocity measurement across the exhaust filter surface with a thermo-anemometer and for exhaust filter changes.

When the BSC is hard-ducted or connected by a thimble unit to the ventilation system, adequate space must be provided so that the configuration of the duct work will not interfere with airflow. The thimble unit must provide access to the exhaust filter for testing of the HEPA filter.
Additional Location Considerations

Here are some additional points which should be taken into consideration when deciding where to put the Biological Safety Cabinet.

1. Biological Safety Cabinets should not be installed as an integral part of a room(s) supply and exhaust system so that fluctuations of the room supply and exhaust air cause the Biological Safety Cabinets to operate outside their design parameters for containment.

2. Biological Safety Cabinets (or fume hoods) should not be used as the sole source of room exhaust.

3. The room supply air system should be equipped with dampers to prevent backflow if Biological Safety Cabinets are connected to exhaust ductwork.

4. If Biological Safety Cabinets are connected to exhaust ductwork, connections should be by thimble units where appropriate. Room exhaust ducts should be equipped with manual dampers to permit sealing for decontamination. NuAire recommendations for installation should be carefully followed.

continues...
Additional Location Considerations (continued)

5. Cabinets should be located away from doors, away from windows that can be opened, away from room supply air and away from heavily-traveled laboratory areas.

6. A minimum of 12 inches (30cm) clearance is recommended, where possible, on each side of the cabinet to allow easy access for cleaning. 16 to 18 inches (30cm) of clearance may be required above the cabinet’s exhaust filter to allow the airflow to be recirculated back into the room. This also provides an easy access to the exhaust filter for proper testing. If the cabinet’s exhaust air is ducted to an external source this minimum distance is still necessary to provide testing access for filter integrity.

If you have additional questions about the location or installation of your BSC, please refer to your Operation and Maintenance Manual, or visit the NuAire Web site (by touching this link) or navigate with your browser to http://www.nuaire.com.
Chapter 3

Proper Preparation: The 4 Steps to Take Before You Begin Work in Your NuAire Biological Safety Cabinet

Step 1:
Develop a Standard Operating Procedure and Manual

Each laboratory should develop or adopt a bio-safety or operations manual which identifies the hazards that will or may be encountered, and which specifies practices and procedures designed to minimize or eliminate risks.
Step 1 (continued)

Laboratory personnel must receive appropriate training on the potential hazards associated with the work involved, including the necessary precautions to prevent exposures, and the exposure evaluation procedures. Personnel should be advised of special hazards and should be required to read and to follow the required practices and procedures.
It’s also important that personnel receive annual updates, or additional training as necessary for procedural or policy changes.

A scientist trained and knowledgeable in appropriate laboratory techniques, safety procedures, and associated with handling infectious agents must direct laboratory activities. Access to the laboratory is limited or restricted at the discretion of this laboratory director when experiments or work with cultures and specimens are in progress. The laboratory director should establish policies and procedures so that only those who have been advised of the potential hazard and meet specific entry requirements (e.g. immunization) enter the laboratory or animal rooms.

Step 2: Prepare the Biological Safety Cabinet

Turning on the Cabinet
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.
Step 2 (continued)
Disinfecting the Work Surface

The work surface, the interior walls (not including the supply filter diffuser), and the interior surface of the window should be wiped with 70% ethanol (EtOH), a 1:100 dilution of household bleach (i.e., 0.05% sodium hypochlorite), or other disinfectant as determined by the operator to meet the requirements of the particular activity. 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 re-contaminate cabinet surfaces, a critical issue when sterility is essential (e.g., maintenance of cell cultures). DO NOT depend on the UV germicidal lamp to provide a sterile work surface.
Step 2 (continued)

Disinfecting Work Materials

The surfaces of all materials and containers placed into the cabinet should be wiped with 70% EtOH to reduce the introduction of contaminants to the cabinet environment. This simple step will reduce introduction of mold spores and thereby minimize contamination of cultures. You can further reduce microbial load on materials to be placed or used in your NuAire Biological Safety Cabinets by periodically decontaminating incubators and refrigerators.

Step 3: Prepare Experiment Equipment

Prepare a written checklist of materials necessary for a particular activity. Know what you’re working with.
Step 3 (continued)

Place everything needed to complete the particular procedure inside the cabinet prior to beginning work.

This minimizes the number of arm-movement disruptions across the fragile air barrier of the cabinet.

As much as possible, restrict the opening and closing of lab doors and walking traffic in the work area when the cabinet is being used. These activities will disturb the cabinet's airflow.
Step 3 (continued)

Remove unnecessary items; excessive materials may disrupt the airflow. Arrange implements in a logical manner to segregate clean and dirty materials. Remember to provide a container for wastes on the inside of the cabinet. Nothing should pass in or out through the air barrier until the procedure is complete.

Remove any items on the intake grilles that may block or disrupt the air supply.

The front grill **MUST NOT** be blocked with research notes, discarded plastic, wrappers, pipetting devices, etc.
Step 3 (continued)

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 (e.g., vortex mixers, tabletop centrifuges) should be placed toward the rear of the cabinet to take advantage of the air split.
Step 3 (continued)

Before beginning work, the operator should adjust the stool height so that his/her face is above the front opening.

Manipulation of materials should be delayed for approximately one minute after placing the hands/arms inside the cabinet. This allows the cabinet to stabilize and to "air sweep" the hands and arms to remove surface microbial contaminants. When the user's arms rest flatly across the front grille, room air may flow directly into the work area, rather than being drawn through the front grille. Raising the arms slightly will alleviate this problem.
Airflow of an A/B3 Biological Safety Cabinet

Click to play the airflow demonstration animation. Touch the pause button on controller bar to stop.
Step 3 (continued)

Plastic-backed absorbent toweling can be soaked in a disinfectant and placed on the work surface (but not on the front or rear grille openings). This toweling facilitates routine cleanup and reduces splatter and aerosol formation during an overt spill. It can then be folded and placed in an autoclaveable biohazard bag when work is completed.

Extra supplies (e.g., additional gloves, culture plates or flasks, culture media) should be stored outside the cabinet. Only the materials and equipment required for the immediate work should be placed in the BSC.

Upright pipette collection containers should not be used in Biological Safety Cabinets, nor placed on the floor outside the cabinet. The frequent inward/outward movement needed to place objects in these containers is disruptive to the integrity of the cabinet air barrier and can compromise both personnel and product protection.
Step 3 (continued)

Only horizontal pipette discard trays containing an appropriate chemical disinfectant should be used within the cabinet.

Potentially contaminated materials should NOT be brought out of the cabinet until they have been surface decontaminated. Instead of bringing them directly out, contaminated materials can be placed into a closable container for transfer to an incubator, autoclave or for other decontamination treatment.
Step 4: Dress Properly

Proper dress is essential for protection of personnel, product and environment. These guidelines are suggested minimums for safety when working in the BSC.

Laboratory coats should be worn buttoned over street clothing; latex gloves are worn to provide hand protection. (A solid front, back-closing lab gown provides better protection of personal clothing than a traditional lab coat.)

Gloves are necessary when handling infected animals and when hands may contact infectious materials, contaminated surfaces or equipment.
Step 4 (continued)

Gloves should be pulled over the knitted wrists of the gown, rather than worn inside. Elasticized sleeves can also be worn to protect the investigator's wrists. Wearing two pairs of gloves may be appropriate; if a spill or splatter occurs, the hand will be protected after the contaminated glove is removed. Dispose of gloves when contaminated, remove when work with infectious materials is completed, and DO NOT wear gloves outside the laboratory. Disposable gloves are not washed or reused.

Protective eyewear should be worn for anticipated splashes of microorganisms or other hazardous materials to the face.

Laboratory gloves and other necessary supplies are available from Scientific Visions at http://www.scientificvisions.com.
Chapter 4

Personal Hygiene Practices for Working Safely in a Biological Safety Cabinet

Laboratory Personnel must observe strict rules of personal hygiene practices in order to maintain even minimum levels of acceptable containment and protection.

Eating, drinking, smoking, handling contact lenses, and applying cosmetics are not permitted in the work areas where there is reasonable likelihood of exposure to potentially infectious materials.
Persons who wear contact lenses in laboratories should also wear goggles or a face shield.

Food must be stored outside the work area in cabinets or refrigerators designated and used for this purpose only.

Mouth pipetting is prohibited; mechanical pipetting devices must be used.

All procedures must be performed carefully to minimize the creation of splashes or aerosols.

Persons must wash their hands after they handle viable materials and animals, after removing gloves, and before leaving the laboratory.
Laboratory equipment and work surfaces should be decontaminated with an appropriate disinfectant on a routine basis, after work with infectious materials is finished, and especially after overt spills, splashes, or other contamination by infectious materials.
All cultures, stocks, and other regulated wastes must be decontaminated before disposal by an approved decontamination method, such as autoclaving. Materials to be decontaminated outside of the immediate laboratory must be placed in a durable, leak-proof container and closed for transport from the laboratory. Materials to be decontaminated at off-site from the laboratory must be packaged in accordance with applicable local, state, and federal regulations, before removal from the facility.

Spills and accidents which result in overt exposures to infectious materials must be immediately reported to the laboratory director. Medical evaluation, surveillance, and treatment must be provided as appropriate and written records are maintained.

Animals not involved in the work being performed are not permitted in the lab. Be sure an insect and rodent control program is in effect.
Chapter 5

Working With and In Your NuAire Biological Safety Cabinet

Movement within the Cabinet

Good technique when working within the BSC will minimize air turbulence and prevent splatter or unwanted spread of aerosols. Here are some guidelines for good technique that will maximize potential protection of personnel, product and environment.
Minimize Movement

The rapid movement of a worker's arms in a sweeping motion into and out of the cabinet will disrupt the air curtain and may compromise the partial barrier containment provided by the BSC. Moving arms in and out slowly, perpendicular to the face opening of the cabinet, will reduce this risk. Other personnel activities in the room (e.g., rapid movement, open/closing room doors, etc.) may also disrupt the cabinet air barrier.
Reduce Splatter

Many common procedures conducted in Biological Safety Cabinets may create splatter or aerosols. Good microbiological techniques should always be used when working in a Biological Safety Cabinet to minimize this potential. For example, techniques to reduce splatter and aerosol generation will minimize the potential for personnel exposure to infectious materials manipulated within the cabinet. Class II cabinets are designed so that horizontally nebulized spores will be captured by the downward flowing cabinet air within fourteen inches of travel. As a general rule of thumb, keeping clean materials at least one foot away from aerosol-generating activities will minimize the potential for cross-contamination.
Working with Equipment in the Cabinet

The middle third of the work surface (in red, below) is the ideal area to be used.

All operations should be performed at least 4 inches from the front grille on the work surface. Materials or equipment placed inside the cabinet may cause disruption to the airflow, resulting in turbulence, possible cross-contamination, and/or breach of containment.
Active work should flow from the clean to contaminated area across the work surface.

Materials and supplies should be placed in such a way as to limit the movement of "dirty" items over "clean" ones.

Maintain proper balance of materials from left to right in order to prevent an airflow imbalance within the work zone.
Bulky items such as biohazard bags, discard pipette trays and suction collection flasks should be placed to one side of the interior of the cabinet. Most procedures should not require use of a flame when combined with good aseptic technique and proper cabinet use. If a burner is required, use the "Touch-O-Matic" type with a pilot light.

Since a burner will produce air turbulence, place it to the rear of the workspace.
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 (e.g., vortex mixers, tabletop centrifuges) should be placed toward the rear of the cabinet to take advantage of the air split.

Working “Clean to Dirty” in 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). Reverse the arrangement for left-handed persons.
Preventing Cross-Contamination

Here are a number of things that should be done to reduce the chance for cross-contamination when working in a BSC:

• Opened tubes or bottles should not be held in a vertical position.

• Investigators working with Petri dishes and tissue culture plates should hold the lid above the open sterile surface to minimize direct impaction of downward air.

• Bottle or tube caps should not be placed on the toweling. Items should be recapped or covered as soon as possible.
• In almost all cases, open flames are not required in the near microbe-free environment of a Biological Safety Cabinet. On an open bench, flaming the neck of a culture vessel will create an upward air current which prevents microorganisms from falling into the tube or flask. An open flame in a BSC, however, creates turbulence which disrupts the pattern of air supplied to the work surface. When deemed absolutely necessary, touch plate micro-burners equipped with a pilot light to provide a flame on demand may be used. This will minimize internal cabinet air disturbance and heat buildup. The burner must be turned off when work is completed. Small electric "furnaces" are available for decontaminating bacteriological loops and needles and are preferable to an open flame inside the BSC. Disposable sterile loops can also be used.
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.

The left suction flask (A) is used to collect the contaminated fluids into a suitable decontamination solution; the right flask (B) serves as a fluid overflow collection vessel. A glass sparger in flask B minimizes splatter. An in-line HEPA filter (C) is used to protect the vacuum system (D) from aerosolized microorganisms.
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 the microorganisms as they are collected. Once inactivation occurs, liquid materials can be disposed of appropriately as noninfectious waste.
Chapter 6

When Work is Completed

Decontaminate

Enclose any items which have been in contact with the agent and decontaminate the surface.
All containers and equipment should be surface decontaminated and removed from the cabinet when work is completed. At the end of the work day, the final surface decontamination of the cabinet should include a wipe down of the work surface, the cabinet's sides and back, and the interior of the glass. If necessary, the cabinet should also be monitored for radioactivity and decontaminated when necessary.

Any spilled fluid and disinfectant solution on the work surface should be absorbed with paper towels and discarded into a biohazard bag.
The drain pan should be emptied into a collection vessel containing disinfectant. A flexible tube should be attached to the drain valve and be of sufficient length to allow the open end to be submerged in the disinfectant within the collection vessel. This procedure serves to minimize aerosol generation. The drain pan should be flushed with water and the drain tube removed.

**Cover Waste Containers**
Allow the cabinet to operate for five minutes with no activity in order to purge airborne contaminants from the work area.

**Remove all Equipment From the Cabinet**
Thoroughly decontaminate interior work surfaces. Twenty or thirty minutes is generally considered an appropriate contact time for decontamination, but this varies with the disinfectant and the microbiological agent. If using a chloride type disinfectant, after contact time, wipe down interior surfaces with a 70% alcohol solution to protect stainless steel interiors from corrosion.

The cabinet can be turned off and the vertically sliding window closed. If desired, the UV light may be turned on.
Thoroughly wash your hands and arms with warm, soapy water. Personnel should remove their gloves and gowns and wash their hands as the final step in safe microbiological practices.

**Leave the Biological Safety Cabinet Running**
The Biological Safety Cabinet may be left on at all times. If the unit is not left running continuously, turn the blower on and air purge for at least five minutes to remove airborne contamination before the next use. If the Biological Safety Cabinet is vented to the outside of the building, both the remote motor and the internal blower should be left on at all times.

**Turn Off the UV Light**
Never work in the unit with the UV light illuminated. (UV light will damage the human eye very quickly.)

**Spills Within the Cabinet**
Small spills within the BSC can be handled immediately by removing the contaminated absorbent paper toweling and placing it into the biohazard bag. Any splatter onto items within the cabinet, as well as the cabinet interior, should be immediately wiped with a towel dampened with decontaminating solution. Hands should be washed whenever gloves are changed or removed.
BIOHAZARDOUS SPILLS IN THE CABINET

Perform decontamination steps while the cabinet is operating to prevent the escape of airborne contaminants.

Spray or wipe all potentially contaminated walls, work surfaces, and implements with an appropriate disinfectant detergent. (Make sure to wear gloves while doing this.)

If the spill is large, flood the work surface with disinfectant and allow to stand 10 to 15 minutes before absorbing and wiping clean.
Changing HEPA Filters

Consult your operation and maintenance manual. HEPA filters, whether part of a building exhaust system or part of a cabinet, will require replacement when they become so loaded that sufficient airflow can no longer be maintained.

Filters must be decontaminated before removal. To contain the formaldehyde gas typically used for microbiological decontamination, exhaust systems containing HEPA filters require airtight dampers to be installed on both the inlet and discharge side of the filter housing. This ensures containment of the gas inside the filter housing during decontamination.

Access panel ports in the filter housing also allow for performance testing of the HEPA filter. (See photo at right.)

HEPA filters are typically constructed of paper-thin sheets of borosilicate medium, pleated to increase surface area, and affixed to a frame. Aluminum separators are often added for stability.
A bag-in/bag-out (BIBO) filter assembly can be used in situations where HEPA filtration is necessary for operations involving biohazardous materials and hazardous or toxic chemicals. This protects the technician handling the filter as well as the environment.

This shows the BIBO filter enclosure with (A) filters, (B) bag, (C) safety straps, (D) cinching straps, and (E) shock cord in the mouth of the PVC bag restricts the bag around the second rib of the housing lip.

The BIBO system is used when it is not possible to decontaminate the HEPA filters with formaldehyde gas, or when hazardous toxic chemicals have been used in the BSC. Note, however, that this requirement must be identified at the time of purchase and installation; a BIBO assembly cannot be added to a cabinet after the fact.
Certification and Annual UV Light Check

CERTIFICATION
A Biological Safety Cabinet MUST be certified by a qualified individual at the following times:

- Newly Installed
  It is extremely important that all new Biological Safety Cabinets be certified when they are received from the manufacturer. Failure to do so may lead to the use of a cabinet which is not functioning appropriately and cause the owner to pay for repairs which should be covered under the purchasing agreement.

- Moved or Relocated
  Relocating a BSC may break the HEPA filter seals or otherwise damage the filters or the cabinet.

- After a Major Repair such as replacement of HEPA filters or motor.

- Annually
  Each BSC should be tested and certified at least annually to ensure continued proper operation.
On-site testing following the recommendations for field testing (NSF Standard 49) must be performed by experienced, qualified personnel. Some basic information is included here to assist in understanding the frequency and kinds of tests to be performed.

**Background**

In 1993, NSF began a program for accreditation of certifiers based on written and practical examinations. Education and training programs for candidate accredited field certifiers are offered through the Eagleson Institute, Sanford, ME; the Harvard School of Public Health, Cambridge, MA; NuAire Inc., Plymouth, MN; Forma Scientific Inc., Marietta, OH; and Lab Conco Corporation, Kansas City, MO. Other training, education and certification programs may be developed in the future. Selecting competent individuals to perform testing and certification is important, and it is suggested that the institutional biosafety officer be consulted in identifying companies qualified to conduct the necessary field performance tests.
Use Accredited Field Certifiers

It is strongly recommended that accredited field certifiers be used to test and certify Biological Safety Cabinets whenever possible. If in-house personnel are performing the certifications, then these individuals should become accredited. The importance of proper certification cannot be emphasized enough, since persons who manipulate infectious microorganisms are at increased risk of acquiring an occupational illness when their Biological Safety Cabinets are functioning improperly.

The annual tests applicable to each of the three classes of Biological Safety Cabinets are listed and described below. There is also information regarding the conduct of selected tests. Biological Safety Cabinets perform consistently well when proper annual certification procedures are followed; cabinet or filter failures tend to occur infrequently.
Performance Testing Biological Safety Cabinets in the Field

Biological Safety Cabinets are the primary containment device that protect the worker, product and environment from exposure to microbiological agents, and their performance as specified by Standard No. 49 needs to be verified at the time of installation and annually thereafter. The purpose and acceptance level of the performance tests (below) are to ensure the balance of inflow and exhaust air, the distribution of air onto the work surface, and the integrity of the cabinet. Other tests check electrical and physical features of the BSC.

A. Down Flow Velocity and Volume Test: This test is performed to measure the velocity of air moving through the cabinet workspace, and is to be performed on all Class I and II Biosafety Cabinets.

B. Inflow Velocity Test: This test is performed to determine the calculated or directly measured velocity through the work access opening, to verify the nominal set point average inflow velocity and to calculate the exhaust airflow volume rate.
C. Airflow Smoke Patterns Tests: This test is performed to (1) determine if the airflow along the entire perimeter of the work access opening is inward, (2) if airflow within the work area is downward with no dead spots or refluxing, (3) if ambient air passes onto or over the work surface, and (4) if there is refluxing to the outside at the window wiper gasket and side seals. The smoke test is an indicator of airflow direction, not velocity.

D. HEPA Filter Leak Test: This test is performed to determine the integrity of supply and exhaust HEPA filters, filter housing, and filter mounting frames while the cabinet is operated at the nominal set point velocities. An aerosol in the form of generated particulates of dioctylphthalate (DOP) or an accepted alternative is required for leak-testing HEPA filters and their seals. Although DOP can be an irritant to most laboratory professionals, competent service personnel are trained to use this chemical in a safe manner. The aerosol is generated on the intake side of the filter, and particles passing through the filter or around the seal are measured with a photometer on the discharge side. This test is suitable for ascertaining the integrity of all HEPA filters.
E. Cabinet Leak Tests: The pressure holding test is performed to determine if exterior surfaces of all plenums, welds, gaskets, and plenum penetrations or seals are free of leaks. It need only be performed just prior to initial installation when the BSC is in a free-standing position (all four sides are easily accessible) in the room in which it will be used, after a cabinet has been relocated to a new location, and again after removal of access panels to plenums for repairs or a filter change. This test may also be performed on fully installed cabinets.

F. Electrical Leakage and Ground Circuit Resistance and Polarity Tests: These safety tests are performed to determine if a potential shock hazard exists by measuring the electrical leakage, polarity ground fault interrupter function, and ground circuit resistance to the cabinet connection. They may be performed by an electrical technician other than the field certification personnel at the same time the other field certification tests are conducted. The polarity of electrical outlets are checked using a polarity tester. The ground fault circuit interrupter should trip when approximately 5 milliamperes (mA) is applied.
G. Lighting Intensity Test: This test is performed to measure the light intensity on the work surface of the cabinet as an aid in minimizing cabinet operator's fatigue.

H. Vibration Test: This test is performed to determine the amount of vibration in an operating cabinet as a guide to satisfactory mechanical performance, as an aid in minimizing cabinet operator's fatigue, and to prevent damage to delicate tissue culture specimens.

I. Noise Level Test: This test is performed to measure the noise levels produced by the cabinets, as a guide to satisfactory mechanical performance and an aid in minimizing cabinet operator's fatigue.

J. UV Lamp Test: Some Biological Safety Cabinets have UV lamps. When used, they must be tested periodically to ensure that their energy output is sufficient to kill microorganisms. After having been turned off and allowed to cool, the surface on the bulb should be cleaned with 70% ethanol prior to performing this test. Five minutes after the lamp has been turned on, the sensor of the UV meter is placed in the center of the work surface. The radiation output should not be less than 40 microwatts per square centimeter at 254 nanometers (nm).
Finally, accurate test results can only be assured when the testing equipment is properly maintained and calibrated. It is appropriate to request the calibration information for the test equipment being used by the certifier.

Note
Federal Standard No. 209B, Clean Room and Work Station Requirements has evolved into Federal Standard No. 209E, Airborne Particulate Cleanliness Classes in Clean Rooms and Clean Zones. This standard does not apply to Biological Safety Cabinets and should not be considered a basis for their performance or integrity certification. However, the methodology of 209E can be used to quantify the particulate count within the work area of a BSC. 209E defines how to classify a clean room / clean zone. Performance tests and procedures needed to achieve a specified cleanliness classification are outlined by the Institute of Environmental Sciences–Testing Clean Rooms (IES-RP-CC-006-84-T) and Laminar Flow Clean Air Devices.
The standard can be ordered from NSF for a nominal charge:

NSF International,
789 Dixboro Road
Ann Arbor, Michigan, 48105 U.S.A.
Telephone: 734-769-8010
Fax: 734-769-0109
Email: information@nsf-isr.org

If you have further questions about setup, preparation, operation, finishing work in the BSC or certification, check with your industrial hygienist or bio-safety officer.

You can also send your questions to NuAire via email using this link: service@nuaire.com. Or you can visit the NuAire Web site for answers about Biological Safety Cabinets.

You have purchased one of the finest pieces of laboratory equipment available, a NuAire Biological Safety Cabinet.

Proper preparation, use and maintenance are your assurance of protection for personnel, product and the environment.