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NSF International Standard/
American National Standard
for Biosafety Cabinetry –

Biosafety Cabinetry: Design,
Construction, Performance, and
Field Certification

Standard Developer

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Foreword

The purpose of this Standard is to establish minimum requirements for materials, design, construction, and performance of Biosafety Cabinetry that are designed to protect personnel, product, and the environment. This Standard details requirements for performance testing as well as field certification testing.

This edition of the Standard (NSF/ANSI 49-2008) includes the following revisions:

Issue 12
The revisions from this issue added language in Section 3.13 stating a specification for use in class II biosafety cabinets.

Issue 13
The update from this issue provided a clarification in language in Section 3.4.2.2.

Issue 14
The addition of language in the standard provided for a listing process for the concurrent balance value test. ASHRAE, Standard 111-2008 was updated, and concurrent balance value was defined.

Issue 16
The name of the standard was updated to include all types of cabinets.

Issue 17
These revisions added interlock requirements for both type B cabinets in section F.7.3.2.

Issue 18
This modification provided language to F.1 regarding downflow velocity readings.

Several changes to Annex F were made, they include but are not limited to:

Issue 19
Clarification of Sound Level Measurement requirements.

Issue 20
A requirement was added that supply fan interlocks on B2 cabinets be tested at the time of alarm verification.

Issue 21
This revision updated language in various sections regarding the definition of a type A1/A2 biosafety cabinets and language in annex F regarding integrity testing.

Issue 22 and 25
This revision added an informational annex (G).

Issue 26
This revision added language to specify reported values that must be documented for all tests in annex A and annex F.

2 The information contained in this Foreword is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI’s requirements for an ANS. As such, this Foreword may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.
Issue 31
This issue added the Helium Leak Test and the Sulfur hexafluoride ($\text{SF}_6$) leak test as an informational annex (J). These were removed from the main body via the approval of issue 21.

This Standard was developed by the NSF Joint Committee on Biosafety Cabinetry using the consensus process described by the American National Standards Institute.

Suggestions for improvement of this Standard are welcome. Comments should be sent to Chair, Joint Committee on Biosafety Cabinetry, c/o NSF International, Standards Department, PO Box 130140, Ann Arbor, Michigan 48113-0140, USA.
1 General

1.1 Scope

This Standard applies to Class II (laminar flow) biosafety cabinetry designed to minimize hazards inherent in work with agents assigned to biosafety levels 1, 2, 3, or 4. It also defines the tests that shall be passed by such cabinetry to meet this Standard. This Standard includes basic requirements for the design, construction, and performance of biosafety cabinets that are intended to provide personnel, product, and environmental protection; reliable operation; durability and structural stability; cleanability; limitations on noise level; illumination; vibration; and motor/blower performance.

1.2 Minimum requirements

Cabinets qualifying under this Standard shall have passed all of the designated tests. Units with component parts covered under existing NSF standards or criteria shall conform to those applicable requirements.

1.3 Variations in design and construction

Cabinetry varying in design, construction, or installation of accessory equipment may qualify under this Standard, if appropriate tests and investigations indicate that the equipment is durable and reliable, can be cleaned and decontaminated, and performs in conformance to this Standard. Such equipment shall meet the requirements for materials and finishes in this Standard.

Major modifications require appropriate tests for conformance. Major modifications include, but are not limited to, changes in the following: location or capacity or quantity or all three of blower/motor(s); size or design or both of air plenums; position of High Efficiency Particulate Air (HEPA) filters; position or redesign of work surface; work area intake and exhaust air grilles; window placement or design; access opening size; location and size of exhaust port; and built-in accessory equipment (centrifuges, ultraviolet lighting, supports for intravenous drug container, arm rests, etc.). Relocation of utility service equipment (electrical outlets, petcocks, etc.) is not considered a major modification if other provisions of this Standard are not compromised.

2 Normative references

The following documents contain requirements that, by reference in this text, constitute requirements of this Standard. At the time of publication, the indicated editions were valid. All documents are subject to revision, and parties are encouraged to investigate the possibility of applying the most recent editions of the documents indicated below.
ACGIH, Industrial Ventilation, A Manual of Recommended Practice

ANSI 226.1 – Test No. 17

ANSI/NFPA 70, 1999, National Electrical Code

APHA, Compendium of Methods for Microbiological Examination of Foods, 1976 (Spore staining techniques)

APHA, Standard Methods for the Examination of Water and Wastewater, Seventeenth Edition (Standard dilution plate methods)


IES, Illuminating Engineering Society Lighting Handbook

IEST-RP-CC-001, Recommended Practice for HEPA Filters

IEST-RP-CC007, Testing ULPA Filters

IEST-RP-CC-013, Institute of Environmental Sciences Recommended Practice, Tentative, August, 1986

IEST-RP-CC021, Testing HEPA and ULPA Filter Media

MIL-F-51079B, Filters, Particulate, High Efficiency, Fire Resistant, Biological Use

NIOSH, Department of Health and Human Services (DHHS) reports in "Hazard Review of Bis(chloromethyl)ether (BCME)


3 American Conference of Governmental Industrial Hygienists, 1330 Kemper Meadow Dr., Cincinnati, OH 45240 www.acgih.org

4 American National Standards Institute, 25 West 43rd Street, Fourth Floor, New York, NY 10036 www.ansi.org

5 National Fire Protection Association, 1 Batterymarch Park, Quincy, MA 02269 www.nfpa.org

6 American Public Health Association, 800 I Street, NW, Washington, DC 20001 www.apha.org


8 Illuminating Engineering Society, 345 E. 47th St., New York, NY 10017 www.iesna.org

9 Institute of Environmental Sciences and Technology, 5005 Newport Drive, Suite 506, Rolling Meadows, IL 60008-1699 www.iest.org


11 NIOSH, Department of Health and Human Services (DHHS), Publications Office, 4676 Columbia Pkwy., Cincinnati, OH 45226 www.cdc.gov/niOSH/

3 Definitions

3.1 accessible: Fabricated to be exposed for cleaning and visual inspection using simple tools (screwdriver, pliers, open-end wrench, etc. [Also see 3.18, "readily accessible."]).

3.2 biohazard (a contraction of the words biological and hazard): Infectious agent(s), or part thereof, presenting a real or potential risk to the well-being of man, animals, and/or plants, directly through infection or indirectly through disruption of the environment.

3.3 biosafety levels:14 The combination of laboratory practices and techniques, safety equipment, and laboratory facilities appropriate for the operations performed and the hazard posed by the infectious agents and the laboratory function or activity. These biosafety levels are described in Biosafety in Microbiological and Biomedical Laboratories.15

3.3.1 biosafety level 1: Practices, safety equipment, and facility design and construction are appropriate for undergraduate and secondary educational training and teaching laboratories and for other laboratories in which work is done with defined and characterized strains of viable microorganisms not known to consistently cause disease in healthy adult humans. *Bacillus subtilis*, *Naegleria gruberi*, infectious canine hepatitis virus, and exempt organisms under the NIH Guidelines for Research Involving Recombinant DNA Molecules16 are representative of those microorganisms meeting these criteria. Many agents not ordinarily associated with disease processes in humans are, however, opportunistic pathogens and may cause infection in the young, the aged, and immunodeficient or immunosuppressed individuals. Vaccine strains that have undergone multiple in vivo passages should not be considered avirulent simply because they are vaccine strains.

Biosafety Level 1 represents a basic level of containment that relies on standard microbiological practices with no special primary or secondary barriers recommended, other than a sink for hand washing.

3.3.2 biosafety level 2: Practices, equipment, and facilities are applicable to clinical, diagnostic, teaching, and other facilities in which work is done with the broad spectrum of indigenous moderate-risk agents present in the community and associated with human disease of varying severity. With good microbiological techniques, these agents can be used safely in activities conducted on the open bench, if the potential for producing aerosols is low. Hepatitis B virus, human immunodeficiency virus, the salmonellae, and *Toxoplasma spp*. are representative of microorganisms assigned to this containment level. Biosafety Level 2 is appropriate when work is done with any human-derived blood, body fluids, tissues, or primary human cell lines where the presence of an infectious agent may be unknown. (Laboratory personnel working with human-derived materials should refer to the OSHA Bloodborne Pathogen Standard for specific required precautions.)

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14 Previously referred to as risk levels (low, moderate, and high)


16 Department of Health and Human Services, National Institute of Health, 6705 Rockledge Drive, Suite 750, MSC 7985, Bethesda, MD 20892-7985  [www.nih.gov](http://www.nih.gov)
Primary hazards to personnel working with these agents may include accidental percutaneous or mucous membrane exposures or ingestion of infectious materials. Extreme caution should be taken with contaminated needles and sharp instruments. Even though organisms routinely manipulated at Biosafety Level 2 are not known to be transmissible by the aerosol route, procedures with aerosol or high splash potential, which may increase the risk of such personnel exposure, must be conducted in primary containment equipment or in devices such as biosafety cabinets (BSCs) or safety centrifuge cups. Other primary barriers should be used as appropriate, such as splash shields, face protection, gowns, and gloves.

Secondary barriers such as hand washing sinks and waste decontamination facilities must be available to reduce potential environmental contamination.

3.3.3 biosafety level 3: Practices, safety equipment, and facility design and construction are applicable to clinical, diagnostic, teaching, research, or production facilities in which work is done with indigenous or exotic agents with a potential for respiratory transmission and that may cause serious and potentially lethal infection. *Mycobacterium tuberculosis*, *St. Louis encephalitis virus*, and *Coxiella burnetii* are representative of the microorganisms assigned to this level. Primary hazards to personnel working with these agents relate to autoinoculation, ingestion, and exposure to infectious aerosols.

At Biosafety Level 3, more emphasis is placed on primary and secondary barriers to protect personnel in contiguous areas, the community, and the environment from exposure to potentially infectious aerosols. For example, all laboratory manipulations should be performed in a BSC or other enclosed equipment, such as a gas-tight aerosol generation chamber. Secondary barriers for this level include controlled access to the laboratory and ventilation requirements that minimize the release of infectious aerosols from the laboratory.

3.3.4 biosafety level 4: Practices, safety equipment, and facility design and construction are applicable for work with dangerous and exotic agents that have a high individual risk of life-threatening disease, which may be transmitted via the aerosol route and for which there is no available vaccine or therapy. Agents with a close or identical antigenic relationship to Biosafety Level 4 agents also should be handled at this level. When sufficient data are obtained, work with these agents may continue at this level or at a lower level. Viruses such as Marburg or Congo-Crimean hemorrhagic fever are manipulated at Biosafety Level 4.

The primary hazards to personnel working with Biosafety Level 4 agents are respiratory exposure to infectious aerosols, mucous membrane or broken skin exposure to infectious droplets, and autoinoculation. All manipulations of potentially infectious diagnostic materials, isolates, and naturally or experimentally infected animals pose a high risk of exposure and infection to laboratory personnel, the community, and the environment.

The laboratory worker’s complete isolation from aerosolized infectious materials is accomplished primarily by working in a Class III BSC or in a full-body, air-supplied, positive-pressure personnel suit. The Biosafety Level 4 facility itself is generally a separate building or completely isolated zone with complex, specialized ventilation requirements and waste management systems to prevent release of viable agents to the environment.

3.4 cabinet classification: Although this Standard covers only Class II biosafety cabinetry, Class I and Class III cabinets are currently defined and known to be commercially available. Biosafety cabinets can be used for work with biological agents assigned to biosafety levels 1 through 4, depending on the facility design as described in *Biosafety in Microbiological and Biomedical Laboratories*. Special note should be taken that BSL 4 agents should only be used in Maximum Containment Laboratories and that Class I and Class II biosafety cabinets are only acceptable in Maximum Containment Laboratories with positive pressure containment suits.

3.4.1 Class I: A ventilated cabinet for personnel and environmental protection, having an un-recirculated inward airflow away from the operator that exhausts all air to the atmosphere after filtration
through a HEPA filter. Class I cabinets are suitable for work where no product protection is required.

NOTE – Although the traditional Class I BSC is exhausted to the atmosphere without recirculation into the lab, it is recognized that some of the benefits of the Class I BSC can be obtained even when the unit’s HEPA filtered exhaust is vented back into the laboratory.

3.4.2 Class II: A ventilated cabinet for personnel, product, and environmental protection having an open front with inward airflow for personnel protection, downward HEPA filtered laminar airflow for product protection, and HEPA filtered exhausted air for environmental protection.

NOTE – When toxic chemicals or radionuclides are used as adjuncts to biological studies or pharmaceutical work, Class II cabinets designed and constructed for this purpose should be used.

3.4.2.1 Class II Type A1 cabinets (formerly designated Type A): cabinets that

– maintain minimum average inflow velocity of 75 ft/min (0.38 m/s) through the work access opening;

– have HEPA filtered downflow air that is a portion of the mixed downflow and inflow air from a common plenum (i.e., a plenum from which a portion of the air is exhausted from the cabinet and the remainder supplied to the work area);

– may exhaust HEPA filtered air back into the laboratory or to the environment through an exhaust canopy; and

– have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums.

Type A1 cabinets are not suitable for work with volatile toxic chemicals and volatile radionuclides.

3.4.2.2 Class II, Type A2 cabinets (when exhausted to the environment were formerly designated Type B3): cabinets that

– maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening;

– have HEPA filtered downflow air that is a portion of the mixed downflow and inflow air from a common exhaust plenum;

– may exhaust HEPA filtered air back into the laboratory or to the environment through an exhaust canopy; and

– have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums.

Type A2 cabinets used for work with minute quantities of volatile toxic chemicals and tracer amounts of radionuclides required as an adjunct to microbiological studies must be exhausted through properly functioning exhaust canopies.

3.4.2.3 Class II Type B1 cabinets: cabinets that

– maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening;

– have HEPA filtered downflow air composed largely of uncontaminated recirculated inflow air;
– exhaust most of the contaminated downflow air through a dedicated duct exhausted to the atmosphere after passing through a HEPA filter; and

– have all biologically contaminated ducts and plenums under negative pressure or surrounded by negative pressure ducts and plenums.

Type B1 cabinets may be used for work treated with minute quantities of volatile toxic chemicals and tracer amounts of radionuclides required as an adjunct to microbiological studies if work is done in the direct exhausted portion of the cabinet, or if the chemicals or radionuclides will not interfere with the work when recirculated in the downflow air.

3.4.2.4 Class II, Type B2 cabinets: cabinets that

– maintain a minimum average inflow velocity of 100 ft/min (0.51 m/s) through the work access opening;

– have HEPA filtered downflow air drawn from the laboratory or the outside air (i.e., downflow air is not recirculated from the cabinet exhaust air);

– exhaust all inflow and downflow air to the atmosphere after filtration through a HEPA filter without recirculation in the cabinet or return to the laboratory; and

– have all contaminated ducts and plenums under negative pressure or surrounded by directly exhausted (nonrecirculated through the work area) negative pressure ducts and plenums.

Type B2 cabinets may be used for work with volatile toxic chemicals and radionuclides required as adjuncts to microbiological studies.

3.4.3 Class III: A totally enclosed, ventilated cabinet of leak-tight construction. Operations in the cabinet are conducted through attached rubber gloves. The cabinet is maintained under negative air pressure of at least 0.50 in w.g. (120 Pa). Downflow air is drawn into the cabinet through HEPA filters. The exhaust air is treated by double HEPA filtration or by HEPA filtration and incineration.17

3.5 calibration: Comparison of the measurement of a standard or instrument of unknown accuracy with another standard or instrument of known accuracy to detect, correlate, report, or eliminate by adjustment any variation in the accuracy of the unknown standard or instrument.

3.6 certification, cabinet design: Cabinet design certification is formal validation by a qualified design testing organization that a designated cabinet model meets all the requirements of annex A of this standard.

3.7 certification, cabinet field: Cabinet field certification is formal verification by a qualified field-testing certifier that an installed cabinet meets all the requirements of annex F of this standard.

3.8 chemical resistance: Capability of materials to maintain their original surface characteristics under prolonged contact with cleaning compounds, decontaminating agents, and normal conditions of the use environment.

3.9 closed: Fabricated with no openings exceeding 0.031 in (0.079 cm).

3.10 concurrent balance value: This value is determined using the duct traverse measurement method as specified in ASHRAE Standard 111-2008, a minimum of 7.5 duct diameters downstream of a

direct connected BSC. Prior to determining the concurrent balance value, it shall be confirmed that the cabinet is operating at its nominal setpoints for inflow and downflow velocity +/-3 fpm. The primary DIM method shall be used for setting the inflow velocity. The accuracy of the DIM shall be better than or equal to +/- 3% and +/- 7 cfm. The static pressure is also measured approximately two duct diameters from the cabinet exhaust connection. Appropriate filter load and tolerance values shall be added to the base static pressure value to accommodate filter loading: 0.3” w.g. shall be added for Type B1 cabinets and 0.7” w.g. shall be added for Type B2 cabinets. The resulting values may be used for design and balance exhaust/supply HVAC requirements.

3.11 decontamination: Inactivation or destruction of infectious agents or neutralization of toxic agents.

3.12 direct inflow measuring device (DIM): A volumetric airflow measuring device consisting of a capture hood with a sensing component that provides a readout as a single value for volumetric flow rate and meets the requirements of annex B.

3.13 downflow velocity – 4 inches: The flow of air in the cabinet coming from the downflow HEPA filters down into the work area at a point 4 in (10 cm) above the lower level of the window sash.

3.14 high efficiency air filters (for use in class II biosafety cabinets):

3.14.1 high efficiency particulate air (HEPA) filter: A throwaway, extended/pleated medium, dry-type filter with the following:

- rigid casing enclosing the full depth of the pleats;
- minimum particulate removal of 99.99% for thermally generated monodisperse dioctylphthalate (DOP) smoke particles or equivalent with a diameter of 0.3 µm (Type C);
- minimum particulate removal of 99.99% and determination as the lower efficiency when tested for particle size ranges of 0.1 to 0.2 µm or 0.2 to 0.3 µm in accordance with IEST-RP-CC007 (Type J);
- minimum particulate removal of 99.95% and determination as the lower efficiency when tested for particle size ranges of 0.1 to 0.2 µm or 0.2 to 0.3 µm in accordance with IEST-RP-CC007 (Type K);
- maximum pressure drop of 1.0 in w.g. (250 Pa) when clean and operated at rated airflow capacity; and
- no area showing a penetration exceeding 0.01% when scan tested with a polydisperse aerosol having a light scattering median size of 0.7 µm and a geometric standard deviation of 2.4.

These filters conform to all the performance and construction requirements of a Type C, a Type J, or a Type K filter respectively, contained in IEST-RP-CC001.4. Filter media shall be tested in accordance with the methods of IEST-RP-CC021 with performance levels to meet the minimum efficiency requirements as specified above and the pressure drop requirements as required by the specific application.

3.14.2 ultra-low-penetrating air (ULPA) filter: A throw away, extended/pleated medium, dry-type filter with the following:

- rigid frame enclosing the full depth of the pleats;
- minimum particle removal of 99.999% and determination as the lower efficiency when tested for particle size ranges of 0.1 to 0.2 µm or 0.2 to 0.3 µm when tested in accordance with IEST-RP-CC007;
– maximum pressure drop of 1.0 in w.g. (250 Pa) when clean and operated at rated airflow capacity. ULPA filters may have higher airflow resistance than HEPA filters for the same rated airflow; therefore, care shall be taken to ensure that the pressure drop is compatible with the cabinet motor/blower capability; and

– no area showing a penetration exceeding 0.01% when scan tested with a polydisperse aerosol having a light scattering median size of 0.7 µm and a geometric standard deviation of 2.4.

This filter conforms to all requirements of a Type F filter contained in IEST-RP-CC001.4, HEPA and ULPA filters.

3.15 laminar airflow: Unidirectional airflow through the work area often referred to as turbulence-free airflow; steady, unidirectional micro-turbulence flow; or mass airflow.

3.16 leak tight: Free of leaks at 2 in w.g. (500 Pa) of air pressure as described in annex A.

3.17 nominal set point velocities: The cabinet downflow and inflow velocities that the manufacturer designates as the settings at which the cabinet is intended to operate and the settings at which it passed the tests listed in 6.7 and annex A, section A.7.

3.18 polydisperse aerosol: Aerosol with a light scattering median size of 0.7 µm and a geometric standard deviation of 2.4.

3.19 readily accessible: Fabricated to be exposed for cleaning and visual inspection without using tools.

3.20 readily removable: Capable of being taken away from the main unit without using tools.

3.21 removable: Capable of being taken away from the main unit using simple tools (screwdriver, pliers, open-end wrench, etc. [also see 3.19, "readily removable"]).

3.22 sealed: Fabricated with no openings that will permit entry or leakage of air (leak-tight).

3.23 smooth: A surface free of pits and inclusions, with cleanability equal to or exceeding the following.

3.23.1 interior work surfaces and exposed interior surfaces: Number 3 (100 grit) finish on stainless steel.

3.23.2 other interior surfaces and exterior surfaces: Commercial grade cold-rolled, hot-rolled, or combination cold/hot-rolled steel free of visible scale.

3.24 surfaces: (figure 1)

3.24.1 interior work surfaces: Surfaces used when performing a task, operation, or activity.

3.24.2 exposed interior surfaces: Exposed interior surfaces, other than work surfaces, that are subject to splash, spillage, or airborne contamination during normal use.

3.24.3 other interior surfaces: Interior surfaces not exposed to splash or spillage but exposed to vapor or volatile toxic substances or both.

3.24.4 exterior surfaces: All exposed surfaces not defined as interior.

3.25 toxic: Having an adverse physiological effect on biological systems.
3.26 work area: The horizontal plane inside the cabinet extending from sidewall to sidewall and from back wall to the inside of the window or sash at a point approximately 2 in (5 cm) above the lower level of the window sash.

3.27 work tray: The solid floor of the work area identified by the manufacturer as the location for the user's activity. This is differentiated from work area.

4 Materials

4.1 General

Materials shall withstand normal wear, corrosive action of gases or liquids, cleaning compounds, and decontaminating agents and procedures. Materials shall be structurally sound, dimensionally stable, fire and moisture resistant, and compatible with other materials used in the laboratory.18

4.2 Interior work surfaces

Interior work surfaces shall be smooth, 300-series stainless steel.

4.3 Exposed interior surfaces

Exposed interior surfaces shall be smooth and abrasion- and corrosion-resistant or shall be rendered corrosion-resistant with nontoxic material that resists crazing, cracking, and chipping. Recirculated air diffuser materials shall be tested in accordance with UL Standard 94. Nonrigid diffuser materials shall conform to Class 94HBF; rigid diffuser materials shall conform to Class 94HB.

4.4 Other interior and exterior surfaces

Other interior and exterior surfaces shall be smooth and abrasion- and corrosion-resistant or shall be rendered corrosion-resistant with nontoxic materials that resist crazing, cracking, and chipping.

4.5 Materials and finishes

4.5.1 Windows

Windows shall be optically clear and not adversely affected by accepted cleaning methods and decontaminating agents. Glazing materials shall be laminated glass, tempered glass, safety plastic, or equivalent. Edges shall be ground or provided with protective stripping.

4.5.1.1 Flammability

Safety plastic view screens shall be tested in accordance with UL Standard 94 and conform to Class 94HB.

4.5.1.2 Abrasion resistance

Windows shall be abrasion-resistant and show no more than 5% change in haze when tested in accordance with 5.17, Test No. 17 of ANSI Standard 226.1.

18 See Annex H for material selection guidance.
4.5.2 Protective coatings

4.5.2.1 Chemical resistance

Protective coatings shall be resistant to prolonged contact to liquids, cleaning compounds, and procedures. Specifically, the protective coatings used shall be resistant to the following chemicals, when tested in accordance with annex D:

- 1N hydrochloric acid;
- 1N sodium hydroxide;
- 1% quaternary ammonium compound;
- 5% formaldehyde;
- 5000 ppm hypochlorite;
- 2% iodophor;
- 5% phenol; and
- 70% ethyl alcohol (ethanol).

When a coating is exposed to these chemicals following the test methods in annex D, there shall be no visible effect on the finish other than a slight change of gloss, discoloration, and/or temporary softening of the finish, with no loss of adhesion or film protection.

NOTE – When special chemical solutions are intended to be used, the resistance of the material thereto shall also be evaluated.

4.5.2.2 Abrasion resistance

Protective coatings for exposed interior, other interior, and exterior surfaces shall meet the following requirements when tested in accordance with annex D:

- maximum weight loss – 100 mg; and
- minimum wear value – 500 cycles.

4.5.3 Plastics

Plastics shall meet the applicable requirements of 4.1, 4.3, 4.4, and 4.5.1.

4.5.4 Welding

Welded seams and deposited weld material shall meet the applicable requirements of 4.1, 4.2, 4.3, and 4.4.

4.5.5 Gaskets and sealants

Gaskets and sealants shall be closed cell, durable, resistant to cleaning and disinfecting agents, and resistant to general use. They shall be made of materials that do not release halogens and are non-hardening, nontoxic, stable, odor free, not detrimentally absorbent, and unaffected by exposure to gases, liquids, cleaning compounds, and decontamination agents listed in 4.5.2.

Exposed surfaces of gaskets for all access panels, doors, structural seams, and windows shall be skinned and smooth. Gaskets supplied with HEPA filters shall be exempt from this requirement.

4.5.6 Sound dampening

Sound-dampening materials shall conform to the requirements for the area in which they are used. They shall not be used in areas subject to contamination. Non-hardening and porous types shall not be accepted.
4.5.7 Hard solder

Hard (silver) solder shall be formulated to be corrosion-resistant.

5 Design and construction

5.1 General

Cabinets shall be designed and constructed to function properly and operate in a safe manner, minimize contamination, provide personnel and product protection, and be capable of being cleaned and decontaminated. Exposed burrs and sharp edges (including, but not limited to, sheet metal screws) shall be eliminated from surfaces of the cabinet that are subject to normal operation, field certification, and maintenance (including those maintained with simple tools).

5.2 Cleanability

Interior work, exposed interior, and the other interior surfaces subject to splash or spillage shall be readily accessible and easily cleanable as assembled or when removed. Interior work, exposed interior, and other interior surfaces, including plenums, shall be capable of being vapor or gas decontaminated.

5.3 Decontamination

Cabinets shall be designed to be decontaminated with an inactivating agent (such as formaldehyde gas) without being moved. Closure to contain decontaminating agents should be limited to gas-tight sealing of air intake and exhaust openings with metal plates, or plastic film and tape, or equivalent. Pressure tight valves, if provided, suitable for decontamination shall be located on the clean side of the HEPA filter.

5.4 Plenum design

5.4.1 Type A1

Type A1 cabinets can have biologically contaminated plenums under positive or negative pressure to the room.

5.4.2 Type B1 and A2

All biologically contaminated ducts and plenums in Types B1 and A2 cabinets shall be maintained under negative pressure or enclosed within a negative pressure zone.

5.4.3 Type B2

Plenums or ducts carrying contaminated air shall be maintained under negative pressure or enclosed within a directly exhausted (nonrecirculated) negative pressure zone.

5.5 Internal corners and angles

5.5.1 Interior work surfaces

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19 See Annex G.
5.5.1.1 Two-plane intersection

An internal angle of 2 rad (110°) or less formed by the intersection of two planes, which is subject to manual cleaning, shall have a minimum continuous and smooth radius of 0.13 in (3.2 mm) (see figure 2).

5.5.1.2 Three-plane intersection

An internal corner formed by the intersection of three planes at 2 rad (110°) or less, subject to manual cleaning, shall have a minimum continuous and smooth radius of 0.25 in (6.3 mm) for a vertical or horizontal intersection. The alternate intersections shall have a minimum continuous and smooth radius of 0.13 in (3.2 mm) (see figure 2).

5.5.1.3 Fillet material

Parent material or hard solder may be used as fillet material in structurally sound seams.

5.6 External corners and angles

All external corners and angles subject to splash or spillage or both shall be sealed as smooth as the surfaces being joined, and formed to eliminate sharp edges that may interfere with use, cleaning, or maintenance (see figure 3).

5.7 Joints and seams

5.7.1 Interior work and exposed interior surfaces

All joints and seams subject to routine manual cleaning shall be sealed as smooth as the surfaces being joined. Perimeter drain spillage trough joints and seams shall be welded and sealed. All other seams shall be sealed. Equipment parts shall be stamped, extruded, formed, or cast in one piece. Joints shall be fabricated to eliminate dirt-c catching horizontal ledges.

5.7.2 Other interior and exterior surfaces

All joints and seams subject to routine splash or spillage or both shall be sealed and smooth. All joints and seams subject to exposure to vapor or toxic volatile substances or both shall be sealed. All other seams shall be closed.

5.8 Fastening methods

5.8.1 Exposed fastenings

Exposed screw threads, projecting screws, and studs shall not be used on interior work surfaces. They shall only be used on exposed interior and other interior surfaces when other fastening methods are impractical. All metal fasteners and studs subject to maintenance shall not be subject to excessive overspray.

5.8.2 Exterior fastenings

Fasteners for exterior removable panels that are gasketed and subject to pressure shall be studs with solid acorn nuts, or equivalent, so that the gasket is sealed. Fasteners for other removable panels may be low profile-type fasteners (truss, round counter sunk, flat counter sunk head [see figure 4]), or studs with solid acorn nuts. All metal fasteners and studs subject to maintenance shall not be subject to excessive overspray.
5.8.3 Interior fastenings

In areas subject to cleaning, interior fastenings and joinings shall be fabricated to minimize projections, ledges, and recesses. All metal fasteners and studs subject to maintenance shall not be subject to excessive overspray.

5.9 Welds

Welds shall meet the smoothness requirements of the applicable surface.

5.10 Solder

Solder shall only be used to seal structurally sound seams or as a fillet material (see 5.5.1.3).

5.11 Removable panels

All maintenance panels to access the blower/motor assemblies and filters shall be front access. Panels shall remain in place when sealing fasteners are removed. All cabinets shall be provided with a blower access panel. Cabinets fabricated without an access panel large enough to allow removal of the blower motor assembly as one piece shall be prohibited. The design and construction of removable panels shall minimize projections and openings. Removable panels for access into contaminated areas shall be designed so that upon reassembly, a seal is provided as required in 6.2.

5.12 Stability

Cabinets shall stand on the floor or bench top in a stable and secure manner and not tip or fall when tested in accordance with annex A, section A.8.

5.13 Provision for mounting

Provision shall be made for cleaning, and where necessary, cleaning underneath the unit. All cabinets shall be designed and constructed with one of the following provisions for mounting.

5.13.1 Mounting

The cabinet base shall be designed to be sealed to the mounting surface (floor, raised base, bench top).

5.13.2 Clear space beneath

The cabinet shall be mounted on adjustable legs, or other acceptable means, to ensure a minimum of 4 in (10 cm) of unobstructed clearance beneath the unit. A 2 in (5 cm) minimum clearance beneath the ends of the cabinet is acceptable if the front is open for cleaning and the side panel is equal to or less than 2 in (5 cm) thick (see figures 5, 6, and 7).

5.14 Legs and feet

Legs and feet shall be sufficiently rigid to provide support with a minimum of cross bracing. They shall be fastened to the cabinet and shaped at floor or bench top contact to minimize the accumulation of splash and spillage. Legs and feet shall be of simple design, with no exposed threads. The minimum contact diameter of the foot shall be 0.75 in (19 mm). The foot shall be fabricated with a smooth material to prevent floor damage.

5.15 Reinforcing and framing

Reinforcing and framing members, not totally enclosed or within walls, shall be easily cleanable. Reinforcing and framing members shall not provide harborage for vermin. The ends of all hollow sections,
not subject to gas decontamination, shall be closed. Reinforcing and framing members subject to splash or spillage or both shall be sealed. Horizontal angle reinforcing and gussets shall not be placed where soil may accumulate. Where angles are used horizontally, they shall have one leg turned down wherever the equipment permits or be formed integrally with the sides. All vertical channel sections shall be completely closed or open.

5.16 Fixed panels

Fixed panels shall be designed, constructed, and fastened to eliminate projections and openings.

5.17 Doors and covers

Doors and covers shall fit properly and close completely. Horizontal sliding doors shall not be used for the work area. When used for storage areas, doors shall slide easily and be readily removable. Piano and butt-type hinges are acceptable. Handles shall be designed, constructed, and installed to eliminate sharp edges or unnecessary projections. Latches and hold-open mechanisms shall provide even and secure support.

5.17.1 Single panel

Single panel doors (see figure 8) and covers shall be fabricated to minimize the collection of foreign matter and be designed without channel sections at the bottom. Channel sections, if used, shall be inverted or shallow and wide enough to be easily cleanable. Clean-out holes shall be provided in all channels that are not inverted.

5.17.2 Double panel

Double panel doors and covers shall be fabricated to minimize the collection of foreign matter. Openings to hollow sections shall be closed. If subject to splash, spillage, or both, openings shall be sealed.

5.17.3 Viewing panel

Viewing panels shall be fabricated to prevent particles from entering the workspace by induction through joints, tracks, or guides.

5.17.4 Sliding sash alarm

Sliding sash enclosures shall include an audible alarm activated when the sash is raised above the manufacturer's specified opening height.

5.18 Louvers and openings

All louvers and openings outside the work area and air plenums shall comply with one or more of the following:

- be of drip deflecting design;
- not be subject to routine splash, spillage, or overhead drippage;
- be designed and constructed to be readily accessible and the space behind easily cleanable;
- or
- louvers through double panel doors and covers shall be sleeved.

5.19 Tracks and guides

All tracks and guides for doors, window covers, and access panels shall be designed and constructed to be easily cleaned.
5.20 Filters

- HEPA or ULPA filters shall be required for the downflow and exhaust air systems.

- HEPA and ULPA filters for downflow and exhaust systems shall conform to the materials, construction, and aerosol efficiency requirements of IEST-RP-CC-001.4 for type C, type J, type K, or type F filters. Filter media shall be tested in accordance with the methods of IEST-RP-CC021 with performance levels to meet the minimum efficiency requirements as specified above and the pressure drop requirements as required by the specific application. In addition, HEPA and ULPA filters shall be scan tested for a leakage not to exceed 0.01% when tested in accordance with annex A, section A.3.

- The cabinet shall be designed to provide accessibility for filter installation, testing, and sealing.

- HEPA and ULPA filters shall be mounted to prevent air bypass of the filters. When required, one or more 0.4 in (1 cm) IPS threaded plugged penetrations shall be located in the plenum upstream of the HEPA or ULPA filters and accessible from the front of the cabinet. These penetrations are used to measure the aerosol concentration upstream of the HEPA and ULPA filters during the HEPA or ULPA filter leak test (see 6.3). When the penetration enters a potentially contaminated space, it shall be labeled “Decontaminate Cabinet Before Opening.”

- Cabinets exhausting into the room shall be provided with a perforated exhaust filter guard (see figure 9) to prevent damage to the filter and blockage of exhaust air.

  NOTE – An additional airflow sensor may be provided to indicate blockage of exhaust air.

- HEPA and ULPA filter patches shall not exceed 3% of the total face area of the side being patched. The maximum width of any one patch shall not exceed 1.5 in (4.0 cm).

5.21 Gaskets and sealants

Exposed surfaces of gaskets shall be easily cleanable and shall not contain internal angles (angles less than \(2.4\,\text{rad} \, [135^\circ]\)). All corner joints and hollow sections of gaskets shall be sealed.

- Fixed gaskets shall be securely fastened and sealed in place.

- HEPA filter seals shall be leakproof when tested in accordance with annex A, section A.3. Gaskets on HEPA filters shall have interlocking corners or sealed joints.

- Gaskets used in cabinet seams or on the facing of service panels shall have sealed joints. Structural strength of seams and service panel joints shall be independent of the seal produced by the gasket.

- The structural strength of joints or assemblies where sealant bonding has been applied shall be independent of the sealants.

5.22 Stopcocks and service outlets

Stopcocks and service outlets shall be readily accessible. Electrical outlets on exposed interior surfaces shall have drip-proof caps or gasket seal blade openings.
5.23 Alarms

5.23.1 Sliding sash alarm

Sliding sash enclosures shall include an audible and visual alarm, activated when the sash is raised above the manufacturer’s specified opening height.

5.23.2 Internal cabinet supply/exhaust fan interlock alarm

When a cabinet contains both an internal downflow and exhaust fan, they shall be interlocked so that the downflow fan shuts off whenever the exhaust fan fails. An audible and visual alarm shall signal the failure. If the downflow fan fails, the exhaust fan shall continue to operate, and an audible and visual alarm shall signal the failure.

5.23.3 Type B exhaust alarm

Type B cabinets shall be exhausted by a remote fan. Once the cabinet is set or certified in its acceptable airflow range, audible and visual alarms shall be required to indicate a 20% loss of exhaust volume within 15 sec. The internal cabinet fan(s) shall be interlocked to shut off at the same time the alarms are activated.

5.23.4 Type A1 or A2 exhaust alarm (informative)

Type A1 or A2 cabinets, when canopy connected and exhausted by a remote fan, should have an audible and visual alarm to indicate a loss of exhaust airflow.

5.24 Electrical components

5.24.1 Motor

– A thermal protector shall be provided. It shall not trip at 115% of the rated voltage under maximum load and ambient temperature conditions. The motor shall be rated for continuous operation.

– Fan motors shall be sized to operate at a static pressure sufficient to meet the requirements of 6.13.

– All fan motors shall be variable speed and shall have controls that can be secured. Controls shall be installed behind a removable or locked panel. Motor controls shall permit the adjustment of fan speeds to achieve proper airflow balance.

– Motors and lights shall be separately protected from the receptacles. Circuit overload protection conforming to the National Electrical Code shall be provided. Flexible power cords for single-phase power shall be 3 wire, with the ground wire connected to the frame, unless otherwise specified and sized in accordance with the National Electrical Code for the specified load(s).

5.24.2 Electrical wiring, switches, etc.

Replaceable electrical components shall not be located in contaminated air plenums, except for fan motors, sealed nonporous or jacketed wiring, and necessary airflow sensors. All wiring penetrations of contaminated spaces shall be sealed in accordance with 6.2. Circuit overload protection shall be provided for all receptacles. Switches shall be mounted outside the work area. A wiring diagram showing connection of all electrical components shall be permanently attached to the unit in an accessible location.
outside of air plenum systems. A statement providing starting current, running power, and circuit
requirements shall be provided with the installation instructions.

5.25 Lighting

5.25.1 Work lighting

The light intensity at the work surface shall conform to 6.5. Lamps, ballasts, and starters shall be
accessible and not installed in contaminated areas. Lamps shall be located so reflection does not
interfere with visibility through the window, and the operator's eyes are shielded from direct radiation.

5.25.2 Ultraviolet lighting

UV lighting is not recommended in Class II (laminar flow) biosafety cabinetry. If requested by the
purchaser, it shall be installed in such a manner that it does not reduce the required performance as
specified in 6. This Standard does not provide any performance verification of UV lighting.

5.26 Gauges

Pressure gauges indicating the differential pressure across the recirculated air filter, if provided, shall be
installed in accordance with the manufacturer's instructions. Hose connections to the gauge and sampling
port shall be secured by positive compression clamps. If threaded connections are used to penetrate the
plenum, an engagement of three continuous threads shall be required.

5.27 Drain spillage trough

A drain spillage trough shall be provided below the work surface to retain spillage from the work area; the
trough shall be easily cleanable. A drainpipe shall be connected to the drain spillage trough and fitted with
a 0.37 in (0.94 cm) or larger ball valve. The drainpipe and valve shall conform to the material
requirements of the drain pan or trough. The drain spillage trough shall accommodate at least 1 gal (4 L).
The drain valve shall be identified with a label and operating instructions placed in close proximity to, or
on, the valve.

5.28 Diffuser placement

Removable diffusers shall be designed and constructed to ensure reassembly in the proper operating
position.

5.29 Work area components placement

Readily removable interior work area work surfaces, intake air grills, and exhaust air grills shall be
designed and constructed to ensure fixed reinstallation in their proper operating positions.

5.30 Height and width

The cabinet, excluding removable light fixtures, exhaust filter housings and guards, and adjustable legs or
feet, shall be sized to fit through a 79 by 35 in (201 x 89 cm) doorway using commonly available furniture
moving equipment (jacks and dollies) (see figure 10).

5.31 Data plate(s)

A data plate(s) indicating the following shall be readily visible on the front of the cabinet:

- manufacturer's name and address;

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UV irradiation can cause erythema of skin and eye damage.
- cabinet model;
- cabinet serial number;
- nominal set point for downflow and inflow velocities (DIM and thermal anemometer);
- type classification;
- downflow velocity test grid dimensions (annex A, section A.9.3);
- indication that the cabinet has potentially contaminated plenums that are at positive pressure directly to the room (if applicable);
- voltage requirements; and
- inflow velocity test grid and method (annex A, section A.10.3).
Figure 1 – Surfaces

- Interior work surfaces
- Exposed interior surfaces
- Exterior surfaces
- Other interior surfaces
Intersection of three planes (internal corner), two intersections may have a minimum radius of 1/8 in (3.2 mm), the third must have a minimum radius of ¼ in (6.3 mm)

Intersection of two planes 1/8 in (3.2 mm) minimum radius, vertical or horizontal

Figure 2 – Internal corners and angles

This

Not this

All external corners or angles are to be sealed and finished smooth.

Figure 3 – External Corners or Angles
Figure 4 – Low profile type fasteners
Figure 5 – Clear space beneath

Figure 6 – Clear space beneath

Figure 7 – Clear space beneath
Figure 8 – Single panel door
Figure 9 – Exhaust filter guard
Figure 10 – Height and width

less than 79 in (2 m)

less than 35 in (0.9 m)
6 Performance

6.1 General

For qualification by the testing organization, biosafety cabinetry shall meet the performance requirements listed in 6.2 through 6.15 when tested in accordance with annex A. All removable components within the cabinet that are offered as optional equipment by the manufacturer shall be in place during testing except during nominal set point downflow velocity determination.

6.2 Pressure decay / soap bubble / tracer gas leak

The periphery and penetrations of all plenums shall be leak tight when tested by the pressure decay or soap bubble test (see annex A, section A.1).

6.2.1 The cabinet shall hold 2 in w.g. (500 Pa) within ± 10% for 30 min or all welds, gaskets, penetrations, or seals on exterior surfaces of air plenums shall be free of soap bubbles when at 2 in w.g. (500 Pa) ± 10% pressure above atmospheric.

6.3 HEPA filter leak

6.3.1 HEPA filters, filter housings, and mounting frames shall be tested with dioctyl phthalate (DOP) or equivalent and determined to be leak tight when cabinet is operating at the nominal set point velocities.

6.3.2 Polydisperse DOP or equivalent sustained penetration shall not exceed 0.01% of the upstream concentration at any point when measured on a linear or logarithmic scale photometer (see 3.17).

6.4 Noise level

6.4.1 The noise level shall be determined with the cabinet operating at the nominal set point velocities.

6.4.2 The overall noise level 12 in (30 cm) in front of the cabinet and 15 in (38 cm) above the plane of the work surface at the vertical centerline of the cabinet shall not exceed 67 dbA with a maximum background level of 57 dbA.

6.5 Lighting intensity

6.5.1 The lighting intensity at the work surface shall be determined with a background lighting intensity in the room of 10 ± 5 ft-candles (110 ± 50 lux) at the work surface elevation.

6.5.2 The average lighting intensity shall be a minimum of 60 ft-candles (650 lux). Individual readings shall be a minimum of 40 ft-candles (430 lux).

6.6 Vibration

The net displacement shall not exceed $2 \times 10^{-4}$ in ($5 \times 10^{-6}$ m) rms amplitude at frequencies between 10 Hz and 10 kHz in the center of the work surface when the cabinet is operating at the nominal set point velocities.

6.7 Personnel, product, and cross-contamination protection

The cabinet shall meet the requirements of 6.7.1, 6.7.2, and 6.7.3 and annex A, section A.7, when operating with the airflows specified in that annex.
6.7.1 Personnel protection
The system shall be challenged by $1 \times 10^8$ to $8 \times 10^8$ *Bacillus subtilis* var. *niger* (*B. subtilis*) spores for 5 min. The number of *B. subtilis* colony-forming units (CFU) recovered from the collection suspension of all six glass impinger samplers (AGI-30) shall not exceed 10 CFU per test. Total slit-type air sampler plate counts shall not exceed five *B. subtilis* CFU for a 30 min sampling period. Three replicate tests shall be performed. The control plate shall be positive for *B. subtilis* CFU.

6.7.2 Product protection
The system shall be challenged by $1 \times 10^6$ to $8 \times 10^6$ *B. subtilis* spores for 5 min. The number of CFU recovered on agar settling plates shall not exceed 5 CFU for each test. Three replicates shall be performed. The control plate shall be positive for *B. subtilis* CFU.

6.7.3 Cross-contamination protection
The system shall be challenged by $1 \times 10^4$ to $8 \times 10^4$ *B. subtilis* spores for 5 min. Some agar plates within 14 in (36 cm) from the challenge sidewall will recover *B. subtilis* CFU and shall be used as positive controls. The number of CFU recovered on agar plates with centers greater than 14 in (36 cm) shall not exceed 2 CFU per test. Three replicates each shall be performed from the left and right sides of the cabinet.

6.8 Stability
The cabinet shall be designed and constructed to resist overturning and distortion under applied forces, resist deflection of the work surfaces under load, and resist tipping under workload.

6.8.1 Resistance to overturning
Cabinets shall conform to the requirements of UL 61010A-1, section 7.3.

6.8.2 Resistance to distortion
The top front edge and the top of the sides shall not move forward more than 0.063 in (1.6 mm) from the static position when a 250 lb (110 kg) lateral force is applied to the top rear edge and top of the opposite side, respectively.

6.8.3 Resistance to deflection of work surface
The work surface shall not be permanently deflected by a 50 lb (23 kg) test load distributed uniformly over an area 10 x 10 in (25 x 25 cm) in the center of the work surface.

6.8.4 Resistance to tipping
The rear bottom of the cabinet shall not lift off the floor more than 0.062 in (1.6 mm) when a 250 lb (110 kg) test load is applied to the leading edge of the cabinet.

6.9 Downflow velocity
The average downflow velocity or velocities (non-uniform) and the calculated and measured average inflow velocities of the cabinet shall be set at the nominal set points for testing. Subsequent production cabinets of the initial model and size conforming to 6.7 may also qualify when the average downflow velocity (or velocities, if so specified) is provided within ± 5 ft/min (± 0.025 m/s) (see annex A, section A.9).
6.9.1 Uniform downflow velocity

Cabinets intended to be operated with a uniform downflow velocity shall have individual point velocities that do not vary more than ± 20% or ± 16 ft/min (± 0.08 m/s), whichever is greater, from the average downflow velocity.

6.9.2 Non-uniform downflow velocity

The manufacturer shall designate the velocity gradient in terms of design velocity and distance from the cabinet front for every zone within which velocity is intended to be uniform. The individual point velocities shall not vary more than ± 20% or ± 16 ft/min (± 0.08 m/s), whichever is greater, from the average within each designated zone.

6.10 Inflow velocity

The velocity of the inflow air through the work access opening shall be determined. Subsequent production cabinets of the initial model and size conforming to 6.7 may also qualify if the directly measured and calculated inflow velocities are within ± 5 ft/min (± 0.025 m/s) of the nominal set point velocities.

6.10.1 The minimum directly measured and calculated inflow velocities of Type A1 cabinets shall be 75 ft/min (0.38 m/s).

6.10.2 The minimum inflow quantity per 1 ft (0.3 m) of work area width of Type A1 cabinets shall be 45 ft³/min (0.02 m³/s) (see 6.7 and 6.9).

6.10.3 The minimum directly measured and calculated inflow velocities of Type A2 cabinets shall be 100 ft/min (0.51 m/s).

6.10.4 The minimum inflow quantity per 1 ft (0.3 m) of work area width of Type A2 cabinets volume rate shall be 65 ft³/min (0.03 m³/s) (see 6.7 and 6.9).

6.10.5 The minimum directly measured and calculated inflow velocities of Type B1 and B2 cabinets shall be 100 ft/min (0.51 m/s).

6.10.6 The minimum inflow quantity per 1 ft (0.3 m) of work area width of Type B1 and B2 cabinets volume rate shall be 65 ft³/min (0.03 m³/s).

6.11 Airflow smoke patterns

Smoke patterns shall be determined with the cabinet operating at the nominal set point velocities.

6.11.1 Airflow within the work area of the cabinet shall be downward, with no dead spots, reflux, or escape from the cabinet.

6.11.2 Airflow along the entire perimeter of the work access opening shall be inward, with no reflux out of the cabinet or smoke penetration over or onto the work surface.

6.11.3 Airflow within the work area of cabinets shall be downward (no reflux), with no escape to the outside of the cabinet at the sides and top of the window.

6.12 Drain spillage trough leakage

Drain spillage troughs shall hold a minimum of 1 gal (4 L) of water with no visible leakage after a 1 h holding period.
6.13  Motor/blower performance

When the cabinet is operated at the nominal set point velocities and without readjusting the fan speed control, a 50% increase in pressure drop across the new filter shall not decrease total air delivery more than 10%.

6.14  Electrical safety

The cabinet shall conform to the requirements of UL 61010A-1.

6.15  Performance data

The manufacturer shall provide a performance data sheet with each cabinet. The following quality control tests shall be conducted in accordance with annex A and reported for each unit:

– pressure decay / soap bubble / tracer gas leak;
– HEPA filter leak;
– downflow velocity;
– inflow velocity; and
– airflow smoke patterns.

The following additional quality control tests shall be conducted in accordance with annex A and reported on every tenth unit produced:

– noise;
– lighting; and
– vibration.

6.16  Record maintenance

Quality control test results shall be maintained on file at the plant location for a minimum of three years. Current calibration records (obtained within one year) for all quality control test instruments shall be maintained on file at all times.
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Annex A
(normative)

Performance tests

NOTE – Before any performance tests are run, the cabinet shall be properly installed and leveled and airflows adjusted to the nominal set point (± 3.0 ft/min [± 0.015 m/s]). These tests are intended for the qualification of a new cabinet model by the testing organization. The testing organization also requires and performs appropriate tests during periodic requalification. Cabinet models undergoing major redesign shall be requalified as stated in 1.3 of this Standard. Field tests are provided in Annex F.

Until certified under NSF/ANSI 49 – 2002, all new cabinets shall be factory tested using the procedures described in NSF/ANSI 49, annex A – 2002, with the exception of the downflow velocity test. When the downflow velocity test is performed, the procedure in NSF Standard 49, 1992 should be used; however, the acceptance criteria outlined in the 2002 standard shall be applied.

A.1 Pressure decay / soap bubble / tracer gas leak test

A.1.1 Pressure decay or soap bubble test

A.1.1.1 Purpose

This test on exterior surfaces of all plenums determines whether welds, gaskets, plenum penetrations, and seals are free of leaks.

A.1.1.2 Apparatus

– manometer, pressure gauge, or pressure transducer system with a minimum range of 0 – 2 in w.g. (0 – 500 Pa) and accurate to ± 0.02 in w.g. (5 Pa);
– liquid leak detector;
– plastic sheet (0.02 in [0.5 mm] extruded high-impact styrene); and
– duct tape.

A.1.1.3 Method (pressure decay)

a) Prepare the cabinet as a sealed system, i.e., seal the front window and exhaust port.

b) Remove decorative panels and other access obstructions, where necessary, to expose plenums to be tested.

c) Attach a manometer, pressure gauge, or pressure transducer system to the test area to indicate the interior pressure (see annex A, figure A1a).

d) Pressurize the cabinet with air to a reading of 2 in w.g. (500 Pa), turn off the pressurizing air, and measure the pressure after 30 min. A leakage of 10% of the original pressure is allowable. If a cabinet does not hold 2 in w.g. (500 Pa), use the soap bubble method to locate leaks.

A.1.1.4 Method (soap bubble)

a) Prepare the cabinet as a sealed system, i.e., seal the front window and exhaust port.
b) Remove decorative panels and other access obstructions, where necessary, to expose plenums to be tested.

c) Attach a manometer, pressure gauge, or pressure transducer system to the test area to indicate the interior pressure (see annex A, figure A1a).

d) Pressurize the cabinet with air to ensure a continuous reading of 2 in w.g. (500 Pa) ± 10%.

e) Spray or brush the liquid leak detector along all welds, gaskets, penetrations, and seals on exterior surfaces of cabinet plenums. Small leaks will be indicated by bubbles. Large leaks will occur that blow the detection fluid from the hole without forming bubbles and may be detected by slight feel of airflow or sound.

A.1.1.5 Acceptance

The cabinet shall hold 2 in w.g. (500 Pa) ± 10% for 30 min or all welds, gaskets, penetrations, and seals on exterior surfaces of air plenums shall be free of soap bubbles when at 2 in w.g. (500 Pa) ± 10% pressure above atmospheric.

A.2 HEPA filter leak test

A.2.1 Purpose

This test determines the integrity of downflow and exhaust HEPA filters, filter housings, and filter mounting frames. The cabinet shall be operated within ± 3.0 ft/min (0.015 m/s) of the nominal set point, with the exception of the downflow HEPA filters on B1 cabinets.

A.2.2 Apparatus

A.2.2.1 An aerosol photometer with linear or expanded logarithmic scale shall be used. The instrument shall be capable of indicating 100% upstream concentration with an aerosol of 10 μg/L of polydisperse dioctylphthalate (DOP) particles, or an equivalent fluid, which provides the same particle size distribution (e.g., polyalpha olefin [PAO], di[2-ethylhexyl] sebocate, polyethylene glycol, and medicinal-grade light mineral oil) \(^{21}\) produced by the generator described in annex A, section A.3.2. It shall also be capable of detecting an aerosol of 1 \(\times\) 10^{-5} % of the same particles. The sampling rate of air shall be at least 1 ft\(^3\)/min (5 \times 10^{-4} m^3/s) ± 10%. The probe area shall have a maximum open area of 1.7 in\(^2\) (11 cm\(^2\)) and a minimum dimension of 0.5 in (1.3 cm). The photometer shall be calibrated in accordance with the photometer manufacturer's instructions, or with IEST-RP-CC-013 if instructions are not provided.

A.2.2.2 An aerosol generator of the Laskin Nozzle type conforming to annex A, figure A2 or equivalent shall be used to create an aerosol by flowing air through liquid DOP or an equivalent substitute. When a Laskin nozzle generator is used, the compressed air supplied to the generator should be adjusted to a minimum of 20 psi (140 kPa), measured at the generator manufacturer’s recommended location. The nozzles shall be covered with liquid to a depth not to exceed 1 in (2.5 cm).

A.2.2.3 A pressure gauge for the generator having a maximum range of 0 – 80 psi (550 kPa) with a resolution and accuracy of 1 psi (7 kPa) calibrated by the manufacturer or in accordance with the manufacturer’s instructions shall be used.

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A.2.3 Method

A.2.3.1 Filters that can be scanned

a) Turn on the cabinet blower and lights (types A1/A2 and B2 – downflow filter test). Remove filter diffusers and protective covers if they are present. Place the generator so the aerosol is introduced into the cabinet, as specified by the manufacturer, to provide uniform distribution upstream of the HEPA filter. When the manufacturer has not identified the aerosol introduction point(s), introduce the aerosol in such a manner as to ensure thorough mixing in the cabinet airflow. For example, a T-connection can be fitted to the aerosol generator output to enable distribution of challenge into both entrances of a single blower, or entrances of multiple blowers. The manufacturer shall determine the aerosol introduction point that provides the most uniform distribution (reference IEST-RP-CC-03422).

b) Turn on the photometer and adjust in accordance with the manufacturer's instructions.

c) Sample the aerosol concentration upstream of the HEPA filter and verify that the concentration gives a light scattering intensity at least equal to that produced by 10 $\mu$g/L of DOP.

   – For linear readout photometers (graduated 0 – 100), adjust the instrument to read 100 on the 100% scale.

   – For logarithmic readout photometers, adjust the upstream concentration to $1 \times 10^4$ above the concentration needed to produce one scale division (use the instrument calibration curve).

d) With the nozzle of the probe held not more than 1.0 in (2.5 cm) from the area being tested, scan the entire downstream side of the HEPA filters, and the perimeter of each filter pack, by passing the photometer probe in slightly overlapping strokes at a traverse rate of not more than 2 in/s (5 cm/s). Separate passes shall be made around the entire periphery of the filter, along the bond between the filter pack and frame, and around the seal between the filter and the device.

A.2.3.2 Filters that cannot be scanned

When a cabinet is ducted so that the exhaust filter cannot be scanned, it may be leak tested by drilling a hole approximately 0.3 in (1 cm) in diameter in the duct at a downstream location that will produce a well-mixed aerosol, and inserting the photometer sampling probe with rigid extension tubing through the hole.

A.2.4 Acceptance

A.2.4.1 Filters that can be scanned

Sustained aerosol penetration shall not exceed 0.01% of the upstream concentration at any point.

A.2.4.2 Filters that cannot be scanned

Sustained aerosol penetration shall not exceed 0.005% of the upstream concentration.

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22 HEPA and ULPA Filter Leak Tests. Institute of Environmental Sciences and Technology, 940 East Northwest Highway, Mount Prospect, IL 60056 www.iest.org
A.3 Noise level test

A.3.1 Purpose

This test provides a uniform method for measuring the noise level produced by the cabinet. The methods can be performed in most acoustically ordinary rooms, such as a factory, where walls are neither sound absorbing nor completely sound reflecting. The cabinet shall be operated at the nominal set point velocities within ± 3.0 ft/min (± 0.015 m/s).

A.3.2 Apparatus

The measuring instrument shall be a sound level meter having a minimum accuracy of ± 1 db and resolution of 1 db with a minimum range of 50 to 100 db and an "A" weighting scale set up in accordance with the manufacturer's instructions.

A.3.3 Method

a) Turn on the cabinet blower and lights.

b) Set the instrument to the "A" weighting mode.

c) Measure the noise level 12 in (30 cm) in front of the cabinet leading front edge of the access opening and 15 in (38 cm) above the plane of the work surface, in line with the vertical centerline of the cabinet (see annex A, figure A3).

d) To measure the ambient noise level, turn the cabinet blower and lights off, and if applicable, leave the remote exhaust blower on and measure as in c) above.

A.3.4 Acceptance

Overall noise level in front of the cabinet shall not exceed 67 dbA when measured where the maximum ambient sound level is 57 dbA. When the ambient sound level is greater than 57 dbA, the reading obtained in annex A, section A.4.3 c) shall be corrected in accordance with curves or tables provided in the instrument operator's manual. If this information is not available, use standard correction curves or tables (see below).

<table>
<thead>
<tr>
<th>Difference between total and background sound readings in dbA</th>
<th>Number to subtract from total to yield corrected noise level</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-2</td>
<td>reduce background levels</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>4-5</td>
<td>2</td>
</tr>
<tr>
<td>6-10</td>
<td>1</td>
</tr>
<tr>
<td>&gt;10</td>
<td>0</td>
</tr>
</tbody>
</table>

A.4 Lighting intensity test

A.4.1 Purpose

This test determines the light intensity on the work surface of the cabinet in foot-candles (lux).
A.4.2 Apparatus

A portable photoelectric illumination meter approved for field measurement in accordance with the Illuminating Engineering Society (IES) Lighting Handbook and accurate to ± 10% shall be used. The illumination meter shall be calibrated in accordance with the manufacturer's instructions.

A.4.3 Method

a) With the cabinet lights off, measure the background lighting intensity along the side-to-side centerline of the work tray on a uniform linear pattern in increments close to but no greater than 12 in (30 cm), starting 6.0 in (15 cm) from the side walls (see annex A, figure A4).

b) Turn on the lights and blower.

c) Measure the cabinet light intensity along the side-to-side centerline of the work tray on a uniform linear pattern in increments close to but not greater than 12 in (30 cm), starting 6.0 in (15 cm) from the side walls (see annex A, figure A4).

A.4.4 Acceptance

Lighting intensities shall average a minimum of 60 ft-candles (650 lux) on the work surface, and individual readings shall not be below 40 ft-candles (430 lux) when measured where the background light levels average 10 ± 5 ft-candles (110 ± 50 lux) at the work surface.

A.5 Vibration test

A.5.1 Purpose

This test determines the amount of vibration in the operating cabinet. The cabinet shall be operated within ± 3.0 ft/min (± 0.015 m/s) of the nominal set point velocities.

A.5.2 Apparatus

A vibration analyzer with a minimum reliable reading of 1.0 \times 10^{-6} \text{in} (2.5 \mu \text{m}) \text{rms amplitude} or the ability to detect differences of this magnitude, set up in accordance with manufacturer's instructions.

A.5.3 Method

a) To determine the vibration displacement on the vertical axis, affix the sensing element of the vibration pickup unit firmly onto the geometric center of the work surface(s) by:

– clamping;
– bolting; or
– using an integral magnet with petroleum jelly film, or a double-faced adhesive tape.

The test position is shown in annex A, figure A5.

b) Determine the gross vibration amplitude with the cabinet operating.

c) Determine the background vibration amplitude with the cabinet blower(s) off and, if applicable, the exhaust blower on.
d) Subtract the background from the gross vibration amplitude to determine the net vibration amplitude attributable to the cabinet.

A.5.4 Acceptance

Net displacement shall not exceed $2 \times 10^{-4}$ in ($5 \times 10^{-6}$ m) rms amplitude at 10 Hz to 10 kHz in the center of the work surface(s).

A.6 Personnel, product, and cross-contamination protection (biological) tests

A.6.1 Purpose

These tests determine whether aerosols will be contained within the cabinet, outside contaminants will not enter the cabinet work area, and aerosol contamination of other equipment in the cabinet will be minimized. The cabinet shall be operated at the airflow velocities indicated in the specific test methods with removable equipment installed. The cabinet shall be turned on at least 30 min before the start of any test and operated continuously throughout all test methods. Cabinets meeting these test requirements shall then meet airflow characteristics as measured in annex A, sections A.9 and A.10.

A.6.2 Materials

– spores of Bacillus subtilis var. niger ($B. \ subtilis$), ATCC 9372$^{24}$, or NCTC No. 10073$^{25}$; and

– sterile diluent prepared as follows:

a.1) Step 1: concentrated diluent phosphate buffer solution (PBS):

– dissolve 34 g KH$_2$PO$_4$ in 500 mL distilled water;

– adjust pH to 7.2 ± 0.5 with 1 N NaOH at 77 °F (25 °C); and

– dilute to 1 L with distilled water.

a.2) Step 2: final diluent PBS:

– distilled H$_2$O – 1 L;

– stock PBS step 1 – 1.25 mL;

– final pH – 7.2 ± 0.5;

– autoclave at 250 °F (120 °C) for 15 min; and

– optional – magnesium sulfate (50 g MgSO$_4$ · 7H$_2$O per L distilled water) – 5.0 mL.

or

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$^{24}$ American Type Culture Collection, Rockville, MD  [www.atcc.org](http://www.atcc.org)

$^{25}$ National Collection Type Culture, London, England  [www.ukncc.co.uk](http://www.ukncc.co.uk/)
b)  

- distilled water – 1 L;  
- adjust pH to $7.0 \pm 0.1$ at $77^\circ F (25^\circ C)$;  
- autoclave at $250^\circ F (120^\circ C)$ for 15 min;  

NOTE – Formula b) is suitable for diluent when spore suspension is prepared for immediate use. When storage of diluent suspension at $39.2^\circ F (4^\circ C)$ is required, formula a) should be used;  

- petri plates (100 x 15 mm and 150 x 22 mm) containing nutrient agar, trypticase soy agar$^{26}$, or other suitable growth medium with no inhibitors or other additives;  
- six AGI-30 samplers (flow rate calibrated at 12.3 to 12.6 L/min) containing 20 mL of sterile diluent. The AGI-30 samplers shall be Ace Glass, Inc., Vineland, NJ, Catalog Number 7540-10, air sampling impingers, or equivalent;  
- two slit-type air samplers operating at a rated flow of $1.0 \pm 0.05$ ft$^3$/min ($28 \pm 1.4$ L/min);  
- refluxing 6-jet modified MRE-type short-form collision nebulizer (available as Model CN-38 Nebulizer [Model NSF CN-31/I] from BGI, Inc., Waltham, MA) or any other nebulizer that can be demonstrated to produce a bacterial aerosol of equivalent characteristics.  
- one 2.5 in (63 mm) outside diameter stainless steel, steel, or aluminum cylinder with closed ends shall be used to disrupt the airflow. The length is to be determined by the size of the cabinet interior. One end butts against the back wall of the work area and the other end protrudes at least 6.0 in (15 cm) into the room through the work access opening of the cabinet;  

- suspension of *B. subtilis* var. *niger* spores prepared as follows:  
  
  Method A (using previously harvested *B. subtilis* spores)  
  
  a) Aseptically inoculate (by streak plating technique) several tryptic soy agar (Difco$^{27}$ or equivalent) petri plates (100 x 15 mm).  
  
  b) Incubate for $48 \pm 2$ h at $99 \pm 1^\circ F (37 \pm 0.5^\circ C)$.  
  
  c) Remove characteristic (pigmented dark orange) colonies and transfer them to ten 220-mL sterile screw-capped bottles each containing approximately 50 mL of tryptic soy agar.  
  
  d) Incubate for $48 \pm 2$ h at $99 \pm 1^\circ F (37 \pm 0.5^\circ C)$.  
  
  e) Add 10 mL of PBS to each slant and gently wash the bacteria from the agar surface.  

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$^{26}$ BBL Microbiological Systems, Cockeysville, MD 21030 [www.bd.com](http://www.bd.com)

$^{27}$ Difco Laboratories, P. O. Box 331058, Detroit, MI 48232-7058 [www.vgdllc.com](http://www.vgdllc.com)
f) Combine the bacterial suspensions to yield approximately 100 mL in a sterile 150-mL screw-cap bottle. Heat the stock culture at 149 ± 1 °F (65.0 ± 0.5 °C) for 15 min. If cell debris interferes with nebulizer dissemination, the suspension may be clarified by washing three times in PBS by centrifugation at 2500 rpm for 15 min. Re-suspend in PBS to the original volume.

g) Determine spore concentration by standard dilution-plate methods\(^{28}\) using PBS and tryptic soy agar. Spores prepared as above should yield an average count of 2 \(\times\) 10\(^9\) to 4 \(\times\) 10\(^9\)/mL.

h) Incubate plates for 48 ± 2 h at 99 ± 1 °F (37 ± 0.5 °C).

i) Dilute the spore suspension with PBS to obtain a final spore concentration of 5 \(\times\) 10\(^8\) to 8 \(\times\) 10\(^8\)/mL if the spores are to be used immediately.

j) Store the stock spore suspension (2 \(\times\) 10\(^9\) to 4 \(\times\) 10\(^9\)/mL) at 39 °F (4 °C) or divide it into aliquots to store in screw-capped vials at -94 °F (-70 °C). Make frequent checks of spore viability by surface plating and of spore predominance by an acceptable spore staining technique.\(^{29}\)

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**Method B**

a) Inoculate 250-mL portions of sterile tryptose broth with aliquots of previously harvested *B. subtilis* spores, or rehydrated freeze-dried cultures per ATCC or NCTC instructions.

b) Incubate on a reciprocating shaker for 48 ± 2 h at 99 ± 1 °F (37 ± 0.5 °C).

c) Heat the stock cultures at 149 ± 1 °F (65 ± 0.5 °C) for 15 min.

d) Transfer the suspensions to screw-cap test tubes and wash at least three times in sterile distilled water by centrifugation at 2500 rpm for 15 min. Use PBS in the last washing if storage is required.

e) Determine spore concentration by standard dilution-plate methods using PBS and tryptic soy agar. Spores prepared as described above should average 1.5 \(\times\) 10\(^9\)/mL.

f) Incubate the plates for 48 ± 2 h at 99 ± 1 °F (37 ± 0.5 °C).

g) If the spore suspension is to be used promptly, dilute the spore suspension with PBS to obtain a final suspension concentration of 5 \(\times\) 10\(^8\) to 8 \(\times\) 10\(^8\)/mL.

h) To store the stock spore culture, divide it into aliquots and store it at 39 °F (4 °C) in sterile screw-cap vials or store it in a freezer at -94 °F (-70 °C). Before use, check the viability of the spore suspension as described in annex A, section A.7.3.1.

### A.6.3 Personnel protection test (system challenged with 1 \(\times\) 10\(^8\) to 8 \(\times\) 10\(^8\) *B. subtilis* spores in 5 min)


A.6.3.1 Method

a) Set the cabinet at the nominal set point airflow velocities.

b) A nebulizer containing up to 55 mL of spore suspension \((5 \times 10^8 \text{ to } 8 \times 10^8 /\text{mL})\) shall be centered between sidewalls of the cabinet. The horizontal spray axis shall be placed 14 in (35 cm) above the work surface; the opening of the nebulizer shall be 4 in (10 cm) behind the front window. The spray axis shall be parallel to the work surface and directed toward the front window (see annex A, figure A6).

c) The cylinder shall be placed at the cabinet center. The axis of the cylinder shall be 2.75 in (7.0 cm) above the work surface. Around the cylinder, 4 AGI-30s shall be positioned with the sampling inlets 2.5 in (6.3 cm) outside the cabinet front. Two AGI-30s shall be placed so that their inlet axes are 6.0 in (15 cm) apart and in a horizontal plane tangent to the top of the cylinder. Two AGI-30s shall be positioned so that their inlet axes are 2.0 in (5.0 cm) apart and lie in a horizontal plane 1.0 in (2.5 cm) below the cylinder. As a positive control, an agar plate shall be placed under the center of the cylinder, and supported a minimum of 0.50 in (1.3 cm) above or below the front intake grill, to minimize the obstruction of airflow into the grill (see annex A, figures A7 and A8).

d) Two slit-type air samplers shall be placed so that the horizontal plane of the air inlets is at the work surface elevation, and the vertical axes of the inlets are 6.0 in (15 cm) in front of the cabinet and 8.0 in (20 cm) from each interior sidewall. Two AGI-30 samplers shall be placed so that the horizontal plane of the air inlets is 14 in (36 cm) above the work surface, the vertical axes are 2.0 in (5.0 cm) outside the front edge of the cabinet, and there are 6.0 in (15 cm) on each side of the cabinet centerline (see annex A, figure A9).

e) Duration of the test shall be 30 min. The test sequence shall be as follows:

<table>
<thead>
<tr>
<th>Time remaining (min)</th>
<th>Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>30</td>
<td>start slit samplers</td>
</tr>
<tr>
<td>25</td>
<td>start nebulizer</td>
</tr>
<tr>
<td>24</td>
<td>start impingers</td>
</tr>
<tr>
<td>19</td>
<td>stop impingers</td>
</tr>
<tr>
<td>18.5</td>
<td>stop nebulizer</td>
</tr>
<tr>
<td>0</td>
<td>stop slit samplers</td>
</tr>
</tbody>
</table>

Three replicate tests shall be performed.

f) Filter the sampling fluid from all of the AGI-30 samplers\(^{30}\) through a 1.85 in (47.0 mm) diameter 0.22 \(\mu\)m membrane filter, remove the filter aseptically, and place it on appropriate media. Incubate plates containing the filters and plates from the slit-type air samplers at 98.6 \(^\circ\)F (37.0 \(^\circ\)C). Examine them at 24 – 28 h, and if negative, re-incubate and read at 44 – 48 h.

g) For new and major modification redesign cabinet models, repeat the above steps after setting the cabinet airflow velocities at \(-10 \pm 3.0 \text{ ft/min } (-0.051 \pm 0.015 \text{ m/s})\) inflow using a direct airflow reading instrument and \(+10 \pm 3.0 \text{ ft/min } (+0.051 \pm 0.015 \text{ m/s})\) downflow above and below the nominal set points.

- Airflow velocity readjustments shall be made per the manufacturer’s procedure.
- The overall average downflow velocity shall be used in making downflow adjustments.

\(^{30}\) For research and field applications, the sampling fluid may be filtered separately from each AGI sampler to provide information on specific areas within the cabinet.
– Removable equipment not essential to cabinet operation shall be removed to set the downflow velocity.

h) For new and major modification redesign cabinet models, repeat the above steps setting the airflow velocities at -10 ± 3.0 ft/min (0.051 ± 0.015 m/s) from the nominal set point for both downflow and inflow.

A.6.3.2 Acceptance

The number of *B. subtilis* CFU recovered from the 6 AGI-30 samplers shall not exceed 10 CFU per test. Total slit-type air sampler plate counts shall not exceed five *B. subtilis* CFU for a 30 min sampling period. Three replicate tests shall be performed. The control plate shall be positive. A plate is “positive” when it contains greater than 300 CFU of *B. subtilis*.

A.6.4 Product protection test (system challenged by 1 x 10⁶ to 8 x 10⁶ *B. subtilis* spores in 5 min)

A.6.4.1 Method

a) Set the cabinet at nominal set point airflow velocities.

b) Cover the work surface with open agar plates 100 x 15 mm with the cylinder at the midpoint (see annex A, figure A10).

c) Position the horizontal spray axis of the nebulizer containing 55 mL of 5 x 10⁶ to 8 x 10⁶ spores/mL at the level of the top edge of the work opening, and center it between the two sides of the cabinet, with the opening of the nebulizer 4 in (10 cm) outside the window. The spray axis shall be parallel to the work surface and directed toward the open front of the cabinet.

d) A 2.5 in (6.3 cm) outside diameter cylinder, with closed ends, shall be placed in the center of the cabinet. The cylinder shall be positioned in the cabinet so that one end butts against the back wall of the work area, the other end extends at least 6.0 in (15 cm) into the room through the front opening of the cabinet, and the axis of the cylinder is 2.75 in (7.0 cm) above the work surface.

e) As a positive control, an agar plate shall be placed under the center of the cylinder and supported 0.5 in (1 cm) above or below the front intake grill to minimize the obstruction of airflow into the grill (see annex A, figure A11).

f) The nebulizer shall be operated for 5 min. 5 min after nebulization is terminated, lids shall be placed on the agar plates.

g) The plates shall be incubated at 98.6 °F (37.0 °C) and examined at 24 – 28 h. If negative, they shall be re-incubated and read at 44 – 48 h.

h) For new and major modification redesign cabinet models, the above steps shall be repeated after the cabinet airflow velocities are set at +10 ± 3.0 ft/min (+ 0.051 ± 0.015 m/s) inflow using a direct airflow reading instrument and -10 ± 3.0 ft/min (-0.051 ± 0.015 m/s) downflow from nominal set points.

– Airflow velocity readjustments shall be made per the manufacturer’s procedure.

– The overall average downflow velocity shall be used in making downflow adjustments.

– Removable equipment not essential to cabinet operation shall be removed to set the downflow velocity.
A.6.4.2 Acceptance

The number of *B. subtilis* CFU on agar settling plates shall not exceed 5 CFU for each test. Three replicates shall be performed. The control plates shall be positive. A plate is “positive” when it contains more than 300 CFU of *B. subtilis*.

A.6.5 Cross-contamination test (system challenged by $1 \times 10^4$ to $8 \times 10^4$ *B. subtilis* spores for 5 min)

A.6.5.1 Method

a) Set the cabinet at the nominal set point airflow velocities.

b) Position the horizontal spray axis of the nebulizer containing 55 mL of $5 \times 10^4$ to $8 \times 10^4$ spores/mL 3.0 – 5.0 in (76 – 130 mm) above the work surface, with the back of the nebulizer located against the midpoint of the left interior side wall. The spray axis shall be parallel to the work surface and directed toward the opposite sidewall.

c) Place open agar settling plates (100 x 15 mm) on the work surface in the following manner (see annex A, figure A12):

- two rows of control plates with the centerline under the outlet of the nebulizer;
- one row of plates with their centers on a line drawn front to back 14 in (36 cm) from the side wall being tested; and
- at least one more row of plates nested beyond the 14 in (36 cm) row; two rows when there is room.

d) Start the nebulizer. After 5 min, stop the nebulizer.

e) After 15 min, place the covers on the open agar plates. Incubate the plates at 98.6 °F (37.0 °C) and examine them at 24 – 28 h. If negative, re-incubate and read at 44 – 48 h.

f) Perform the same procedure [a) to e]), but place the nebulizer against the midpoint of the right interior wall.

A.6.5.2 Acceptance

Some agar plates, from the challenge sidewall to 14 in (36 cm) from the sidewall, will recover *B. subtilis* CFU and shall be used as positive controls. The total number of CFU recovered on agar plates with centers greater than 14 in (36 cm) shall not exceed 2 CFU per test. Three replicates each shall be performed from the left and right sides of the cabinet.

A.7 Stability tests

A.7.1 Purpose

These tests demonstrate the structural integrity and stability of a biosafety cabinet for the following:

- resistance to overturning under applied forces (refer to UL 61010A-1) cited in 6.8.1 of this Standard);
- resistance to distortion under applied forces;
– resistance to deflection of work surfaces under load; and
– stability with respect to tipping under load.

Tests are performed by applying static force loads, as described below, and measuring the distortion or deflection within the cabinet.

A.7.2 Apparatus

– compression force gauge or extension spring balance, calibrated in pounds, with an accuracy of ± 5% full scale; or

NOTE – Where an extension type spring balance is used, force shall be applied as "pull" at opposite side of device from that specified in methods below.

– test loads;
– 250 lb (110 kg) uniformly distributed over an area of 10 x 10 in (25 x 25 cm); and
– 50 lb (23 kg) uniformly distributed over an area 10 x 10 in (25 x 25 cm).

A.7.3 Resistance to overturning

A.7.3.1 Method

a) Block the cabinet (adjusted to manufacturer's tallest rated service position on stand if applicable) at front or rear bottom edge to prevent lateral movement.

b) Tilt the cabinet 10° from horizontal in the direction most likely to cause overturn.

A.7.3.2 Acceptance

The cabinet shall not initiate overturn when tilted 10° from horizontal in the direction most likely to cause overturn.

A.7.4 Resistance to distortion under applied forces

A.7.4.1 Method

a) Bolt the device securely to a firm base or floor to prevent overturning and lateral movement.

b) Apply a force of 250 lb (110 kg) at top rear and one top side edge. Measure the forward deflection of the top front edge and opposite top side edge with a dial micrometer (see annex A, figures A13 and A14).

Report the deflection.

A.7.4.2 Acceptance

The top front edge and the top of the sides shall not move forward more than 0.062 in (1.6 mm) from a static position when a 250 lb (110 kg) lateral force is applied to the top rear edge and top of the opposite side, respectively.
A.7.5 Resistance to deflection of work surface under load

A.7.5.1 Method

a) Measure the distance from the center point of the front edge of the work surface to the floor.

b) Place the 50 lb (23 kg) test load at the center of the work surface, distributed over an area 10 x 10 in (25 x 25 cm). Remove the test load and measure the distance from the center point of the front edge of work surface to the floor (see annex A, figure A15).

A.7.5.2 Acceptance

There shall be no permanent deflection of the work surface after the 50 lb (23 kg) test load is applied and removed.

A.7.6 Resistance to tipping under load (applicable only to freestanding devices with work surfaces)

A.7.6.1 Method

Place the 250 lb (110 kg) test load centered from right to left of the work surface on the leading edge of the cabinet (see annex A, figure A16).

A.7.6.2 Acceptance

The rear bottom of the cabinet shall not lift off the floor more than 0.062 in (1.6 mm)) when a 250 lb (110 kg) test load is applied.

A.8 Downflow velocity

A.8.1 Purpose

This test measures the velocity of air moving through the cabinet work space 4 in (10 cm) above the bottom edge of the window, and is performed on all cabinets accepted under annex A, section A.7. Individual point readings shall be taken and reported on a specified grid with removable components removed (nominal set point set up), and the average for each designated zone shall be calculated (uniform downflow represents a single zone).

A.8.2 Apparatus

– A thermal anemometer with an accuracy of ± 3.0 ft/min (± 0.015 m/s) or 3% of the indicated velocity, whichever is larger, shall be used. The device shall be calibrated in accordance with the thermal anemometer manufacturer's instructions, or with IEST-RP-CC-013 if instructions are not provided. When barometric pressure and air stream temperature (where velocity readings are taken) deviate from standard conditions listed for the thermal anemometer being used, correction factors from the manufacturer's manual for the thermal anemometer shall be consulted for the appropriate correction calculation.

– The air measurement probe shall be held rigidly in a freestanding fixture that permits accurate positioning and does not distort the airflow pattern (ring-stand and clamp).
A.8.3 Method: Setting nominal set point

A.8.3.1 Uniform downflow cabinets

Measure the air velocity at multiple points across the workspace, using equal points in the horizontal plane defined 4 in (10 cm) above the bottom edge of the window frame (certified height) using the following spacing:

- A uniform rectangular grid with spacings as close to but no greater than 6.0 x 6.0 in (15 x 15 cm) and containing a minimum of three rows and seven readings per row.

- Perimeter air velocity readings shall be taken 6.0 in (15 cm) away from the walls and window enclosing the work area (see annex A, figure A17).

- Removable equipment nonessential to cabinet operation (acceptable option components) shall be removed prior to the setting of the nominal set point.

The air measurement probe shall be held rigidly in a freestanding fixture that permits accurate positioning and does not distort the airflow pattern (ring-stand and clamp). Reported values shall be each of the readings included in the applicable grid and the overall average of these readings. The nominal set point shall be based on this average.

A.8.3.2 Non-uniform (zoned) downflow cabinets

Measure the air velocity at multiple points across the workspace in zones verified by the testing organization in the horizontal plane defined 4 in (10 cm) above the bottom edge of the window frame (height being tested). Manufacturer's instructions shall include locations of zone boundaries, the number of points within each zone, and the specific grid to be used with equidistant spacing. The removable equipment non-essential to cabinet operation (acceptable option components) shall be removed prior to setting the nominal set points. Reported values shall be each of the readings taken in each of the zones and the average of each zone. The nominal set point shall be based on the above data in accordance with the manufacturer's instructions.

A.8.4 Acceptance

The average downward airflow velocity through the cross section of the unobstructed work area (with removable acceptable option components removed) at the level 4 in (10 cm) above the bottom of the window of cabinets meeting the requirements of annex A, section A.7 shall be the values specified by the manufacturer. Subsequent production cabinets of the initial model and size conforming to annex A, section A.7 may also qualify if the measured downflow velocity set points are within ± 5 ft/min (± 0.025 m/s) of the nominal downflow velocity set point and any additional velocity readings agreed to by the testing organization are provided. Individual point readings in cabinets with uniform downflow shall not vary more than ± 20% or ± 16 ft/min (± 0.08 m/s) from the average downflow velocity, whichever is greater, as determined in annex A, section A.9.3. Individual point readings shall not vary more than ± 20% or ± 16 ft/min (± 0.08 m/s) from the average of each gradient zone, whichever is greater, as determined in annex A, section A.9.3, when the downflow is specified as non-uniform downflow (zoned) by the manufacturer.

A.9 Inflow velocity (face velocity) test

A.9.1 Purpose

This test determines the measured and calculated inflow velocity through the work access opening and the calculated exhaust flow volume rates. A minimum of five individual volumetric readings shall be taken
and averaged using a direct reading instrument and the calculated average intake velocity.

NOTE – Include instructions for the validated secondary method for measuring intake velocity.

A.9.2 Apparatus

The following devices may be used to carry out inflow velocity testing:

- a direct inflow measurement (DIM) instrument with an accuracy of ± 3% of reading ± 7 ft³/min (± 0.003 m³/s) or another acceptable source or in accordance with annex B;

- a thermal anemometer with an accuracy of ± 3.0 ft/min (± 0.015 m/s) or 3% of the indicated velocity (whichever is larger); and

- a pitot tube constructed according to the dimensions given in the Industrial Ventilation Manual.\(^{31}\)

The direct inflow measurement instrument shall be used to obtain direct measurement of inflow volume (primary method). Thermal anemometers or pitot tubes or both shall be used to determine calculated inflow velocity (secondary method).

A.9.3 Methods

A.9.3.1 General

The nominal set point average inflow velocity shall be determined by a direct inflow reading instrument measurement. After the nominal set point is determined by a direct inflow reading instrument measurement, readings shall be taken by the appropriate alternate calculated or measured method recommended by the manufacturer. Both of these set point values shall meet the requirements of annex A, section A.10.4.

A.9.3.2 Direct inflow measurement method (primary method)

a) Seal by taping the device to the center of the front opening of a biological safety cabinet. Seal the open areas on either side of the capture hood portion of the DIM as necessary.

b) All cabinet and exhaust blowers must be operating. Take at least five readings, and average them to determine inflow volume rate. Care should be taken not to restrict the airflow through the instrument intake area.

c) Calculate the average inflow velocity in feet/minute (meters/second) by dividing the average inflow volume rate in cubic feet/minute (cubic meters/second) by the work access opening area in square feet (square meters).

d) Determine the inflow quantity per linear foot of work area width by dividing the inflow volume rate by the width of the work area in feet (meters).

e) Include the following in the reported data: individual inflow volume rate readings, average inflow volume rate, work access opening dimensions and area, directly measured average inflow velocity, width of the work area, inflow quantity per 1 ft (0.3 m) of work area width, and the methods used to determine them.

A.9.3.3 Method to determine concurrent air balancing exhaust values for Type B cabinets only

a) This test shall be conducted before any previous test conditions are changed.

b) Measure and calculate exhaust volume by conducting a duct traverse in accordance with American Society of Heating, Refrigerating, and Air-Conditioning Engineers (ASHRAE)\textsuperscript{32} standards for air velocity measurements in round or rectangular ducts or with the Industrial Ventilation Manual.

c) Measure exhaust static pressure at a point approximately two duct diameters from the cabinet exhaust connection in accordance with ASHRAE standards for air velocity measurements in round or rectangular ducts or with the Industrial Ventilation Manual.

d) Include exhaust volume rate in cubic feet/minute (cubic meters/second) and exhaust static pressure in inches water gauge (Pascal) in the reported data.

A.9.3.4 Alternate inflow measurement methods

These methods, approved by the testing organization, shall be validated and provided by the manufacturer and shall be subject to review by the testing organization. Manufacturer validation procedures shall contain no fewer than ten replicate tests. The testing organization’s approval shall be based on review of data and successful reproduction of test results. The following methods have been found to be acceptable on some cabinets:

A.9.3.4.1 Method for Type A1 and A2 cabinets that use a thermal anemometer to measure exhaust velocity to determine inflow velocity

a) Take air velocity measurements at multiple points across the exhaust filter face as described by the manufacturer on a grid no larger than 4 x 4 in (10 x 10 cm), with the grid starting points and height above the filter validated by the testing organization (see annex A, figure A18).

b) The effective open area of the exhaust HEPA filter or exhaust port shall be determined and supplied by the manufacturer and validated by the testing organization. Cabinets in which the exhaust filter is not accessible or exhaust port flow is non-uniform, such as caused by a damper or exhaust filter housing design, shall be tested as approved by the testing organization.

c) To obtain the exhaust flow volume rate in cubic feet/minute (cubic meters/second), multiply the average exhaust air velocity in feet/minute (meters/second) by the exhaust area in square feet (square meters).

d) Calculate the average inflow velocity in feet/minute (meters/second) by dividing the average exhaust volume rate in cubic feet/minute (cubic meters/second) by the work access opening area in square feet (square meters).

e) Include the following in the reported data: individual exhaust velocity readings, average exhaust velocity, exhaust volume rate, exhaust opening dimensions and area, work access opening dimensions and area, calculated average inflow velocity, and the method used to determine them.

\textsuperscript{32} American Society of Heating, Refrigerating, and Air-Conditioning Engineers, Inc., 1791 Tullie Circle, N.E., Atlanta, GA 30329 www.ashrae.org
A.9.3.4.2 Method for Type A1, A2 and B2 cabinets using a thermal anemometer to measure velocity through a constricted access opening to determine average inflow velocity

a) Restrict the access opening as specified by the testing organization.

b) Air velocity measurements shall be taken at multiple points across the restricted opening as specified on the data plate. No fewer than two readings per 1 ft (0.3 m) of access opening width shall be taken.

c) Average the air velocity measurements. Multiply the average by the listed correction factor to obtain average inflow velocity.

d) Include the following in the reported data: height of restriction, individual velocity readings, average velocity, the listed correction factor, calculated inflow velocity, and methods used to determine them.

A.9.3.4.3 Method for Type B1 cabinets using a thermal anemometer to measure velocity through the access opening to determine average inflow velocity

a) Turn off blower(s) that recirculate air in the cabinet, if specified in the manufacturer's instructions.

b) Set the sash (viewing window) to manufacturer's recommended operating height.

c) Take two rows of air velocity measurements with an anemometer at multiple points in the plane of the access opening. Take one row at a distance below the top of the access opening equal to 25% of the opening height. Take the second row at a distance below the top of the access opening equal to 75% of the opening height (see annex A, figure A19).

d) Take the indicated velocity measurements every 4 in (10 cm) across the width of the front work access opening but no closer than 4 in (10 cm) from sides of the work opening. The average of all measurements represents the inflow velocity.

e) Include individual inflow velocity readings, average inflow velocity, and the methods used to determine them in the reported data.

A.9.3.4.4 Calculated method for Type B2 cabinets using an anemometer and pitot tube, if applicable

a) Turn on the cabinet downflow blower and exhaust system blower.

b) Set the sash (viewing window) at manufacturer's recommended operating height.

c) Measure and calculate exhaust volume in accordance with the testing organization’s verified methodology or with ASHRAE standards for air velocity measurements, in round or rectangular ducts or with the Industrial Ventilation Manual.

d) Measure the supply air velocity on an approximate 4 x 4 in (10 x 10 cm) grid in a horizontal plane 6.0 in (15 cm) below the face of the downflow diffuser, starting 2 in (5 cm) from each perimeter wall. The air measurement probe shall be held rigidly in a freestanding fixture (ring-stand and clamp) that permits accurate positioning and does not distort airflow pattern (see annex A, figure A20). Average the velocity readings and multiply the average by the area in square feet (square meters) of the plane in which the velocities were measured to determine the total filtered air supply in cubic feet/minute (cubic meters/second).
e) Subtract the supply air volume rate in cubic feet/minute (cubic meters/second) from the total exhaust volume rate in cubic feet/minute (cubic meters/second); the difference represents the calculated inflow volume rate in cubic feet/minute (cubic meters/second).

f) Divide the calculated inflow volume rate by the area of the access opening in square feet (square meters) to determine the average inflow velocity in feet/minute (meters/second).

g) Reported the individual exhaust velocity readings, calculated average exhaust velocity, exhaust duct area, calculated exhaust volume, individual supply velocity readings, average supply velocity, effective supply area, calculated supply air volume, area of the work access opening, calculated inflow air volume, calculated access opening average inflow velocity, and the methods used to determine them.

A.9.4 Acceptance

Acceptance criteria shall be based on inflow determined by the direct measurement. Subsequent production cabinets of the initial model and size may also qualify as meeting annex A, section A.7 when the directly measured inflow velocities are provided within ± 5 ft/min (± 0.025 m/s) of the nominal set point velocities.

The minimum inflow velocity of Type A1 cabinets shall be 75 ft/min (0.38 m/s). The minimum inflow volume shall be 45 ft³/min (76 m³/h) per foot (meter) of work area width (see annex A, sections A.7 and A.9).

The minimum inflow velocity of Type A2, B1, and B2 cabinets shall be 100 ft/min (0.51 m/s). The minimum inflow volume shall be 65 ft³/min (110 m³/h) per 1 ft (0.3 m) of work area width (see annex A, sections A.7 and A.9).

A.10 Airflow smoke patterns test

A.10.1 Purpose

This test determines that the airflow along the entire perimeter of the work access opening is inward, that airflow within the work area is downward with no dead spots or refluxing, that ambient air does not pass on or over the work surface, and that there is no escape to the outside of the cabinet at the sides and top of the window.

A.10.2 Apparatus

A source of visible cold smoke such as titanium tetrachloride.

NOTE – Titanium tetrachloride is corrosive and should be handled with care.

A.10.3 Method

A.10.3.1 Downflow test

Smoke shall be passed from one end of the cabinet to the other, along the centerline of the work surface, at a height of 4 in (10 cm) above the top of the access opening.

A.10.3.2 View screen retention test

Smoke shall be passed from one end of the cabinet to the other, 1 in (2.5 cm) behind the view screen, at a height 6.0 in (15 cm) above the top of the access opening.
A.10.3.3 Work opening edge retention test
Smoke shall be passed along the entire perimeter of the work opening edges, approximately 1.5 in (3.8 cm) outside the cabinet. Particular attention should be paid to corners and vertical edges.

A.10.3.4 Sash/window seal test
Smoke shall be passed up the inside of the window 2 in (5 cm) from the sides and along the top of the work area.

A.10.4 Acceptance

A.10.4.1 Downflow test
The smoke shall show smooth downward flow with no dead spots or reflux (upward flow).

A.10.4.2 View screen retention test
The smoke shall show smooth downward flow with no dead spots or reflux. No smoke shall escape from the cabinet.

A.10.4.3 Work opening edge retention test
No smoke shall be refluxed out of the cabinet once drawn in, nor shall smoke billow over the work surface or penetrate onto it.

A.10.4.4 Sash/window seal test
There shall be no escape of smoke from the cabinet.

A.11 Drain spillage trough leakage test

A.11.1 Purpose
This test demonstrates the containment capability of the spillage trough under the work surface.

A.11.2 Method
Fill the drain spillage trough with a minimum of 1 gal (4 L) of water and hold it for 1 h. Check for visible signs of water leakage after 1 h.

A.11.3 Acceptance
The drain spillage trough shall hold a minimum of 1 gal (4 L) of water and have no visible leakage after a 1 h holding period.

A.12 Motor/blower performance

A.12.1 Purpose
This test demonstrates that the motor/blower will operate at a static pressure sufficient to meet the requirements of 6.13.
A.12.2 Apparatus

Instrumentation required in annex A, sections A.9 and A.10 shall be used. A manometer with an accuracy of at least ± 2% of reading shall be used.

A.12.3 Method

a) Set the cabinet at the nominal set point, ± 3.0 ft/min (± 0.015 m/s).

b) Measure the total airflow volume rate, cubic feet/minute (cubic meters/second), and determine that the cabinet blower is delivering at the nominal set point (see annex A, sections A.9 and A.10). The cabinet supply air volume shall be determined as in annex A, section A.10.3.

c) Locate the testing organization approved positive and negative pressure taps. The manufacturer shall locate the positive pressure tap (see annex A, figure A23) directly above the downflow HEPA filter to allow conversion of velocity pressure to static pressure. The positive pressure tap shall not be located in the face of the blower outlet (see annex A, figure A21). If more than one pressure tap is used, as in a piezometer ring, pressure taps may be connected together for an average reading. The manufacturer shall locate the negative pressure tap not less than one-half equivalent diameter from the blower inlet. In the case of double inlet blowers, static measurements shall be made in both blower inlets and connected together for an average static pressure (see annex A, figure A22). If it is not possible to mount both static pressure taps due to cabinet design, one tap will be sufficient. For negative pressure tap, use a series pressure tap (see annex A, figure A23). Attach manometers to each pressure tap and record result. The positive pressure reading is the initial static pressure reference point. The sum of the positive and negative readings without reference sign is the total cabinet static pressure.

d) Increase the initial negative pressure reading by 50% or more of the initial positive pressure reading by restricting the cabinet’s negative airflow. To accomplish this, monitor the cabinet’s initial negative pressure, and load or restrict the cabinet’s negative airflow area (i.e., type A1, A2, B1-front grill or type B2-supply air inlet) until the initial negative pressure has increased by 50% of the initial positive pressure reading. In the case where the first loaded HEPA filter is under negative pressure (type B1), the 50% positive pressure value shall be considered 50% of the pressure drop of the first HEPA filter.

e) Measure the total volume of airflow (cubic feet/minute [cubic meters/second]) the restricted cabinet blower is delivering (see annex A, sections A.9 and A.10).

f) Record the initial negative and positive pressures, the final negative pressures, and the initial and final airflow volume rates.

A.12.4 Acceptance

The total airflow volume rate, cubic feet/minute (cubic meters/second), shall not decrease more than 10% meeting the requirements of section 6.13.

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33 Manufacturer to supply positive and negative pressure taps (see annex A, figures A21 and A22) on units submitted for laboratory certification.
a. Combination inclined and vertical manometer

b. Scanning for tracer gas leaks

Figure A1 – Tracer gas leak test
0.08 in (2 mm) diameter longitudinal holes next to tube in line with radial holes (4 required)

3/8 in (9.5 mm) OD x 0.065 in (1.7 mm) wall brass tubing

length variable to suit installation

5/8 in (15.9 mm) OD brass collar silver to tubing

0.04 in (1 mm) diameter radial holes 90° (1.6 RAD) apart; top edge of holes just touching bottom of collar (4 required)

approximately 0.5 in (12.7 mm) above bottom of can

brass plug – silver braze in place (full penetration)

Figure A2 – Details of Laskin nozzle
Figure A3 – Noise level test

Figure A4 – Lighting intensity test
**Figure A5 – Vibration test**

**Figure A6 – Personnel protection test**
Figure A7 – Personnel protection test
Figure A8 – Personnel protection test

Figure A9 – Personnel protection test
Figure A11 – Product protection test
Figure A12 – Cross contamination test
Figure A13 – Resistance to distortion

dial micrometer, center of front
apply 250 lb (110 kg) force, center of back

tie down
tie down
Figure A14 – Resistance to distortion
Figure A15 – Resistance to deflection

center of point, front edge of work surface, no permanent deflection after test load is removed

50 lb (23 kg) test load, center of work surface
Figure A16 – Resistance to tipping

250 lb (110 kg) applied to center of leading edge

1/16 in maximum (0.2 cm)
Figure A17 – Velocity profile test
The air velocity measurements shall be taken at multiple points across the exhaust filter face on a 10 cm (4 in) grid, with the points approximately 10 cm (4 in) from the filter frame and 10 cm (4 in) above the face of the filter.

Figure A18 – Calculated inflow velocity for Class II Type A
Take air velocity measurements at multiple points in the plane of the access opening. Take two rows of air velocity measurements. One row shall be taken at a distance below the top of the access opening equal to 25% of the opening height. The second row shall be taken at a distance below the top of the access opening equal to 75% of the opening height.

Indicated velocity measurements shall be taken every 4 in (10 cm) across the width of front work access opening but no closer than 4 in (10 cm) from edges of work opening.

Figure A19 – Another example of inflow velocity Class II Type B1
Figure A20 – Supply air volume
Figure A21 – Positive pressure tap placement

Figure A22 – Negative pressure tap placement
Figure A23 - Pressure Tap

- 0.40" diameter thru holes, typical 2 places
- 0.25" outside diameter tube
- 1/8" NPT x 1/4" tube connector, soldered to seal
- 3/16" outside diameter tube barb
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Annex B
(normative)

B.1 Method to verify fitness for use of potential direct inflow measurement devices

B.1.1 Calibrate the basic measuring portion of the device in a wind tunnel with National Institute of Standards and Technology (NIST) traceable calibration (e.g., for devices with removable hoods, calibrate the device without a hood installed; for devices using thermal anemometer, calibrate the thermal anemometer). A pitot tube constructed according to the dimensions given in the Industrial Ventilation Manual is a primary standard and needs no other verification.

B.1.2 Install the device using one of the two following methods:

– Method 1
  a) Seal the device to the front opening of a Class II Type B2 biological safety cabinet hard connected.
  b) Connect the exhaust of the cabinet to a duct containing an orifice meter or other flow meter calibrated traceable to NIST.
  c) Turn off the downflow blower and seal the downflow air opening.
  d) If the cabinet has a moveable sash, seal the sash.

– Method 2
  a) Seal the device to the front opening of a Class II Type A1 or A2 biological safety cabinet intended to be canopy connected.
  b) Seal a calibrated, NIST traceable flow hood, such as Shortridge model CFM-88, to the cabinet exhaust.
  c) If the cabinet has a moveable sash, seal the sash.
  d) The cabinet exhaust filter open area shall be larger than the section of the flow hood where readings are measured (14 x 14 in [36 x 36 cm] for the Shortridge unit).

B.1.3 Using the condition of annex B, section B.1.2, Method 1, measure the exhaust flow both with the device installed and removed. Record at least five readings in each instance. The difference should not exceed 2%. Then run the cabinet at no fewer than three airflow velocities in a range spanning the highest and lowest airflows the device will be required to measure. Record at least five readings of the device and of the flow meter, or orifice meter, and calculate the difference. The average difference should not exceed 2%.

B.1.4 Using the configuration of annex B, section B.1.2, Method 2, measure the exhaust flow both with the device installed and with it removed. Record at least five readings in each instance. The difference should not exceed 2%. Then, run the cabinet at no fewer than three airflow velocities in a range spanning the highest and lowest airflows the device will be required to measure. Record at least five readings of the device and of the flow hood on the cabinet exhaust and calculate the difference. The average difference should not exceed 2%.

34 Shortridge Instruments, 7855 E. Redfield Rd., Scottsdale, AZ 85260  www.shortridge.com
B.1.5 The calibration is valid for cabinets of the size used and smaller. It is recommended that 6 ft (2 m) cabinets be used in this procedure.
Annex C
(normative)

Nebulizer selection and calibration

C.1 Selection

C.1.1 Criteria

Nebulizers are acceptable when they:

- deliver $1 \times 10^8$ to $8 \times 10^8$ airborne spores of *Bacillus subtilis var. niger* in 5 min;
- deliver 94% ± 6% single cell spores; and
- have a spore aerosol discharge velocity of 100 ± 10 ft (30 ± 3 m) per minute.

NOTE 1 – Tests performed by First et al. demonstrated that a stainless steel six-jet collision refluxing nebulizer will deliver the bacterial spore aerosol required in 6.7.1 when the following conditions are met:

- the nebulizer is equipped with a glass flask 2.0 in (5.0 cm) in diameter, 3.5 in (9.0 cm) high and a 0.90 in (2.3 cm), ID horizontal discharge spout on top;
- the nebulizer is operated at 20 psi (140 kPa);
- 55 mL of a $5 \times 10^8$ to $8 \times 10^8$/mL spore suspension is placed in the flask;
- the bottom of the six-jet spray head is 0.71 in (1.8 cm) above the bottom of the flask; and
- six rosette patterns created by the air jets form on the inside of the glass flask. (These should be observed frequently for size and contour to verify that the jets are not clogged or obstructed.)

NOTE 2 – The six-jet collision refluxing nebulizer need not be retested for performance before use.

C.2 Calibration

C.2.1 Purpose

The purpose of this section is to demonstrate that a nebulizer conforms to all the criteria cited in annex C.

C.2.2 Site

The nebulizer shall be calibrated in the laboratory where it is being used.

C.2.3 Frequency

The nebulizer shall be calibrated prior to its first use and periodically thereafter.

C.2.4 Materials

- suspension of $5 \times 10^8$ to $8 \times 10^8$ *Bacillus subtilis var. niger* spores per mL;
- nebulizer to be calibrated;
- one all-glass ACI-30 impinger sampler (ACE Glass, Inc., Vineland, NJ, Catalog Number 7540-10, air sampling impingers, or equivalent);
- switching timer;
- Membrane filter funnel (47 mm filter size) with silicone rubber diaphragms sealed to each end with RTV. Diaphragms are perforated to insert the outlet of the nebulizer at the wider end and one impinger sampler at the other end. Insertions shall be tight on the impinger end. Insertion shall be loose on the nebulizer end so that the impinger is operating in atmospheric pressure, not in a closed system;
- flow meters;
- pressure gauge; and
- 37-mm aerosol type membrane filter in sampling cassette with an open face.

C.2.5 Method

a) Measure the nebulizer outlet dimensions and calculate the area in square feet.

b) Calculate the airflow in cubic feet/minute (cubic meters/second) through the nebulizer required to result in 100 ft/min (0.5 m/s) discharge velocity.

c) Add the manufacturer's recommended volume of spore suspension to the nebulizer.

d) Place the outlet of the nebulizer in the rubber diaphragm of the wide end of the filter funnel. Insert the collecting tube of the impinger sampler through the rubber diaphragm on the opposite end of the filter funnel. Ensure a tight fit at impinger end.

NOTE – The all-glass impinger (chemical corps type) comes in two versions: (1) an impinger with the tip submerged in liquid 4 mm from the flask bottom and passing 6.0 Lpm at a pressure drop of 8.0 psig or greater (ACE Glass No. 7541 impinger) and (2) an impinger with the tip above the liquid surface and passing 12.5 Lpm at a pressure drop of 8.0 psig or greater, known as AGI-30, (ACE Glass No. 7540 impinger). Either impinger may be used. When the air delivery rate of the nebulizer is not precisely 6.0 or 12.5 Lpm, select the impinger that samples at a higher rate and bleed in through an opening around the nebulizer insertion an amount of air equal to the difference in the two airflows. If the nebulizer and the impinger are to be operated at the same flow rate, a snug fit in the diaphragm at both ends is recommended.

e) Attach the hose to a pressure gauge attached to flow meter, then to the nebulizer.

f) Simultaneously turn on the nebulizer (maintain airflow through the nebulizer to result in a calculated 100 ft [30 m] per minute output velocity based on airflow [ft³/min] and diameter of the discharge spout - 12.5 L/m for the six-jet collision described in this annex) and the impinger sampler (operating according to manufacturer's instructions). Operate nebulizer for 5 min (using the switching timer) and the impinger sample for 5.25 min.

g) Aseptically transfer the impinger sampler collecting fluid to a sterile 500-mL graduated cylinder. Rinse the funnel, impinger stem, and bottle with sterile water to insure collection of all spores, and collect all rinse water in the graduated cylinder.
h) Measure and record the volume of fluid in the graduated cylinder. Transfer all the fluid aseptically to a sterile flask containing a magnetic stirrer and mix thoroughly.

i) Prepare serial dilutions and quantify spore concentration by five replicate platings.

j) Actively sample the bacterial aerosol with membrane filter located in its design mode. After sampling is completed, stain the membrane with an appropriate dye. Count the number of deposits containing single and more than one bacterium in representative fields under a microscope.

C.2.6 Calculations

a) number of spores delivered in 5 min = (dilution factor) x (average number of CFUs on the five plates);

b) velocity of air leaving nebulizer = the air volume measured in C.2.5 b) in cubic feet/minute (cubic meters/second) divided by nebulizer outlet area in square feet; and

c) Calculate the percent of single bacteria in the total aerosol sample.

C.2.7 Acceptance

– The average of five replicate calibration tests shall fall between $1 \times 10^8$ and $8 \times 10^8$ spores per 5 min nebulizer operation.

– The velocity of air leaving the nebulizer shall be $100 \pm 10$ ft ($30 \pm 3$ m) per minute.
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Annex D
(normative)

Evaluation of chemical resistance and abrasion resistance of surfaces

D.1 Chemical resistance

D.1.1 Chemicals

The following chemicals shall be used for resistance testing:

- 1N hydrochloric acid;
- 1N sodium hydroxide;
- 1% quaternary ammonium compound;
- 5% formaldehyde;
- 5,000 ppm hypochlorite;
- 2% iodophor;
- 5% phenol; and
- 70% ethyl alcohol.

D.1.2 Method

Chemical spot tests shall be made by applying 10 drops (approximately 0.5 mL) of each reagent to the surface to be tested. Each reagent shall be covered by a watch glass, convex side down, in the center of the puddle, to hold the reagent in place. Reagents shall be allowed to remain on the surface for 4 h, and tests shall be performed so the testing surface is wet throughout the entire test period. After 4 h, the surface shall withstand scrubbing with a stiff brush and hot water at 160 °F (72 °C). Samples shall be dried before examination. Surface stains of dyes shall be removed with an alcohol wash before examination.

D.1.3 Acceptance

When exposed to the chemicals listed above or special chemicals, the surface shall show no visible effect on the finish, other than a slight change of gloss, slight discoloration, or temporary slight softening of the finish, with no loss of adhesion and film protection.

D.2 Abrasion resistance

D.2.1 Method

A protective coating shall be applied in the recommended manner and properly cured on a panel of the prescribed substrate. It shall be evaluated on a Taber Abrader following the procedures of ASTM\textsuperscript{30} D1044-76 using a CS-IOS wheel, and a 1,000-g load for 500 cycles.

D.2.2 Acceptance

The maximum weight loss for 500 cycles shall not exceed 100 mg. The substrate shall not be exposed during the test.

\textsuperscript{30} ASTM, 100 Barr Harbor Drive, PO Box C700, West Conshohocken, PA 19428-2959  \url{www.astm.org}
Annex E
(informative)

E.1 Location

E.1.1 The Class II (laminar flow) biosafety cabinet should be located out of the traffic pattern and away from room air currents that could disrupt the containment provided by the work access opening air barrier. Annex E, figure E1 shows a suggested location after all air turbulence sources have been considered.

E.1.2 If there is a window in the laboratory, it should remain closed at all times. Cabinets should not be located where room ventilation air inlets blow across the front opening or onto the exhaust filter.

E.1.3 Where space permits, a 12 in (30 cm) clearance should be provided behind and on each side of the cabinet. If not feasible, a minimum 3 in (8 cm) clearance on each side and 1.5 in (3.8 cm) clearance in back are recommended. The electrical outlet for the cabinet should be accessible for the cabinet service and electrical safety testing without moving the cabinet.

E.2 Recommendations for installation

E.2.1 Type A1 and A2 cabinets

Type A1 and A2 cabinets are designed to return air to the laboratory and do not generally require external venting. It is critical that a minimum of 3 in (8 cm) clearance be provided between the exhaust opening on top of the cabinet and the ceiling. Less than 3 in (8 cm) clearance constricts the exhaust and reduces the flow into the cabinet at the front access opening. At least 12 in (30 cm) clearance is required between the exhaust opening on top of the cabinet and the ceiling to allow the use of a thermal anemometer to measure the exhaust velocity when calculating the cabinet inflow velocity.

When it is desirable to exhaust air to the atmosphere, exhaust should be via a 100% exhaust system (i.e., a system that does not recirculate its exhaust air into other parts of the building). The recommended exhaust system connection for types A1 and A2 cabinets is an exhaust canopy connection as shown in annex E, figures E2 and E3. Every canopy design must be tested to determine the airflow rate exhausted by the canopy that will ensure performance. Whenever the cabinet is field certified, the minimum exhaust flow by the canopy should be verified by measurements using the approved instruments and techniques cited in annex A, sections A.9 and A.10. No type A cabinet should ever be hard connected to an exhaust system (see figures E4 and E5).

It is preferable that cabinets be installed using an exhaust connection that allows for scan testing of the exhaust HEPA filter in accordance with annex F, section F.5.3.1.

A properly designed and installed exhaust canopy will allow a Type A1 or A2 cabinet to maintain acceptable inflow velocity at the front access opening even when the flow through the exhaust canopy is completely stopped. The performance of the exhaust canopy should be assessed by either the manufacturer of the exhaust canopy or the user to ensure awareness of the performance characteristics of the exhaust canopy with the particular model of cabinet being exhausted.

When the exhaust canopy is used to capture hazardous nonparticulate material being exhausted from the cabinet, the exhaust and associated alarm system should meet the same criteria as indicated for the

37 The information contained in this Annex is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI's requirements for an ANS. Therefore, this Annex may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.
Types B1 and B2 cabinets.

When Type A1 and A2 cabinets are found to be directly attached to the exhaust system and vented to the outside without the use of an exhaust canopy, it is recommended that the exhaust connection be modified to an exhaust canopy.

E.2.2 Types B1 and B2 cabinets

Type B1 and B2 cabinets are to be vented outside the building without recirculation. The venting system should include a leak-tight duct, a damper in the duct near the cabinet to permit flow adjustment closure and decontamination, and an external exhaust fan as the final system component (see annex E, figure E6). The exhaust fan should be sized to deliver the required exhaust airflow (as specified by the cabinet manufacturer), considering pressure losses in the duct and allowing at least 2 in w.g. (500 Pa) for a dirty HEPA filter. If a charcoal filter is used downstream of the HEPA filter, an additional pressure capacity equal to the manufacturer's recommended resistance should be provided. An alarm should be provided at the cabinet to indicate loss of exhaust flow. This can be an exhaust volume flow measuring device in the duct downstream of the exhaust filter, a sail switch at the fan discharge, or a flow measuring station in the exhaust duct. It is recommended that each Type B1 or B2 cabinet have its own (dedicated) exhaust system. The cabinet should be interlocked with the blower in the duct or the building system to prevent pressurization of the exhaust system. In addition, cabinets hard connected to an exhaust system should not be turned off.

It is recognized that there is interest in utilizing the increasingly sophisticated modulated flow exhaust ventilation systems where the exhaust from Type B1 or B2 cabinets, chemical fume hoods, flexible exhaust hoses, and/or room exhausts are modulated based on use to optimize containment, maintain appropriate pressure differentials, and maximize energy savings by reducing overall exhaust volume. These systems are required to maintain a high level of control of many complex factors over a number of years. Although the potential cost savings are great, the severity of the hazards contained by the biological safety cabinets requires the use of simpler and more reliable constant flow systems for the cabinet exhaust.

If a modulated flow exhaust system is used, it is recommended that the operation of the cabinet exhaust be verified under a variety of conditions over time. Furthermore, the type of exhaust alarm must be assessed in light of the type of sensors and controls used in the modulated flow system.

E.2.3 Roof exhaust systems

Roof exhaust systems serving biosafety cabinets should have a stack that extends straight upward at least 10 ft (3 m) above the roof surface to avoid re-entrainment by the building, and should be increased in elevation when necessary to avoid the influence of surrounding structures. Raincaps or any other structure that deflects the straight upward flow of the discharged air should be avoided. No precipitation can enter the stack when air is being exhausted at normal stack velocities. To take care of precipitation during periods when system is shut off, a 1 in (2.5 cm) hole can be drilled in the lowest point of the fan casing and the water allowed to drain onto the roof. It is recommended that roof exhaust fans be energized by direct-connected electric motors to avoid failures caused by slipping and breaking of belts. Another advantage of direct-connected fans is the ability to use the motor non-function to activate an alarm in the laboratory, whereas when a malfunctioning belted fan is employed, the motor can be operating when the fan is idle. A diagram illustrating a recommended roof exhaust facility is shown in annex E, figure E7.

E.3 Electrical

Variations in line voltage may affect the cabinet airflows. A voltage regulator should be installed in order to reduce the potential of variations in airflows.
Figure E1 – Suggested laboratory location for Class II (laminar flow) biosafety cabinet
Figure E2 – Suggested Class II (laminar flow) Type A biosafety cabinet venting system
Figure E3 – Suggested canopy venting for Class II (laminar flow)
Type A, biosafety cabinet

Notes:
1. Actual hood dimensions to be determined by designer
Figure E4 – Alternate venting methods Class II (laminar flow) biosafety cabinet with exhaust duct containing airflow monitoring system
Figure E5 – Alternate venting methods Class II (laminar flow) Biohazard Cabinet with exhaust duct containing airflow monitoring system
Figure E6 – Alternate venting methods Class II (laminar flow), Type B, biosafety cabinet with exhaust ducts
Figure E7 – Exhaust stack and blower

- Stack discharge directly upward
- Guy stack to roof if needed
- Directly connected totally enclosed weatherproof motor
- 2.5 cm diameter hole at low point in blower scroll for drainage to roof

Not for distribution or sale
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Annex F
(normative)

Field tests

F.1 Field certification preconditions and intervals

This annex contains the field tests that define the methods and acceptance criteria that are appropriately applied for determining qualification for field certification of all Class II biological safety cabinets. These field certification procedures are intended to confirm that an installed cabinet evaluated under the current version of the Standard has met all design criteria contained in NSF/ANSI 49 and currently meets all criteria contained in this annex. All cabinets shall be field tested using the procedures described in NSF/ANSI 49, annex F – 2002, with the exception of the downflow velocity test. When the downflow velocity test is performed, the procedure by which the cabinet was certified should be used; however, the acceptance criteria outlined in the 2002 standard shall be applied. Downflow velocity readings shall be taken four inches (ten centimeters) above the bottom edge of the window only when so stated on the manufacturer’s data plate label or when the manufacturer’s data plate label indicates the cabinet was listed to NSF 49-2002 or later.

To ensure that all cabinet operating criteria contained in this annex continue to be met, each cabinet should be field tested at the time of installation and at least annually thereafter. In addition, recertification should be performed whenever HEPA filters are changed, maintenance repairs are made to internal parts, or a cabinet is relocated. More frequent recertification should be considered for particularly hazardous or critical applications or workloads. It is customary for the person conducting the designated tests to affix to the cabinet a certificate of satisfactory performance when the cabinet meets all field test criteria.

Field certification of a cabinet is not intended to provide complete verification that the cabinet conforms to all of the requirements of NSF/ANSI 49.

F.1.1 Tests directly related to containment (i.e., personnel and environmental protection) and product protection.

The following physical tests shall be conducted on-site for a certification to be considered for the statement “Field Certified in accordance with NSF/ANSI 49”:

- downflow velocity profile test;
- inflow velocity test;
- airflow smoke patterns test;
- HEPA filter leak test;
- cabinet integrity test (positive pressure plenum cabinets only); and
- site installation assessment tests.

The site installation assessment tests shall include:

- alarm functions as required by this Standard;
- blower interlock; and
- exhaust system performance (proper exhaust duct negative pressure and canopy performance).

Microbiological equipment that has been used with microorganisms should be decontaminated prior to repair or replacement of components located in contaminated plenums, prior to cabinet relocation, and in some cases prior to recertification. See Annex G, Recommended Microbiological Decontamination Procedure. When equipment has been used with chemical or radioactive agents, appropriate protective clothing and safety procedures should be used during chemical decontamination.

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F.1.2 Tests related to worker comfort and safety

The following tests are for worker comfort and safety and are performed at the request of the customer or at the discretion of the certification provider:

- lighting intensity;
- vibration;
- noise level; and
- electrical leakage, ground circuit resistance, and polarity tests.

F.2 Downflow velocity

F.2.1 Purpose

This test measures the velocity of air moving through the cabinet workspace 4 in (10 cm) above the bottom edge of the window and shall be performed on all cabinets accepted under annex A, section A.7.

F.2.2 Apparatus

F.2.2.1 A thermal anemometer with an accuracy of $\pm 3.0$ ft/min ($\pm 0.015$ m/s) or 3% of the indicated velocity, whichever is larger, shall be used. The device shall be calibrated in accordance with the thermal anemometer manufacturer's instructions or IEST-RP-CC-013 if instructions are not provided. When the conditions vary from sea level by more than 1000 ft (300 m) and/or the temperature varies from 70 °F (21 °C) by more than 5 °F (2 °C), an appropriate correction for altitude and/or temperature should be used. The manufacturer's manual for the thermal anemometer or the Industrial Ventilation Manual shall be consulted for the appropriate correction calculation.

F.2.2.2 A freestanding fixture that permits accurate positioning of the thermal anemometer probe that does not distort the airflow pattern (ring-stand and clamp) shall be used.

F.2.3 Method: setting nominal set point

F.2.3.1 Uniform downflow cabinets

a) The air velocity shall be measured at multiple points across the workspace, using equal points in the horizontal plane 4 in (10 cm) above the bottom edge of the window frame, as specified on the data plate.

b) Removable equipment shall be removed prior to the test to replicate the as-manufactured conditions tested by the testing organization when required.

c) The air measurement probe shall be held rigidly in a freestanding fixture that permits accurate positioning and does not distort the airflow pattern (ring-stand and clamp).

d) Reported values shall be:

- Individual velocity readings in the applicable grid;
- Overall average of the velocity readings;
- Minimum velocity reading;
- Maximum velocity reading;
- Acceptance criteria for average airflow velocity;
- Acceptance criteria for airflow velocity uniformity; and
- Name of test (Uniform Downflow Velocity Test).
e) The nominal set point shall be based on the above data in accordance with the manufacturer’s instructions.

F.2.3.2 Non-uniform (zoned) downflow cabinets

a) The air velocity shall be measured at multiple points across the work space in zones, as specified on the data plate, 4 in (10 cm) above the bottom edge of the window frame.

b) Reported values for each zone shall be:

− Individual velocity readings in the applicable grid;
− Overall average of the velocity;
− Minimum velocity reading;
− Maximum velocity reading;
− Acceptance criteria for average airflow velocity;
− Acceptance criteria for airflow velocity uniformity; and
− Name of test (Non-uniform (zoned) Downflow Velocity Test).

c) The nominal set point shall be based on the above data in accordance with the manufacturer’s instructions.

F.2.4 Acceptance

F.2.4.1 Uniform downflow

A cabinet for which the cabinet manufacturer has specified a uniform downflow velocity qualifies for field certification when (1) the average downflow velocity is within ± 5 ft/min (± 0.025 m/s) of the value specified and (2) the individual point readings do not vary more than ± 25% or 16 ft/min (0.08 m/s), whichever is greater, from the average downflow velocity.

F.2.4.2 Non-uniform downflow

A cabinet for which the cabinet manufacturer has specified a non-uniform (zoned) downflow velocity qualifies for field certification when (1) the individual zone average downflow velocities are within ± 5 ft/min (± 0.025 m/s) of the values specified by the manufacturer and (2) the individual point readings do not vary more than ± 25% or 16 ft/min (0.08 m/s), whichever is greater, from the average downflow velocity of each zone.

F.3 Inflow velocity (face velocity) test

F.3.1 Purpose

This test determines the measured and calculated inflow velocity through the work access opening.

F.3.2 Apparatus

The following devices may be used to carry out inflow velocity testing:

− a direct inflow measurement (DIM) instrument with an accuracy of ± 3% of reading ± 7 ft³/min (± 0.003 m³/s) or in accordance with annex B;

− a thermal anemometer with an accuracy of ± 3.0 ft/min (± 0.015 m/s) or 3% of the indicated velocity, whichever is larger;
– a pitot tube constructed according to the dimensions given in the Industrial Ventilation Manual; and

– a freestanding fixture that permits accurate positioning of the thermal anemometer probe that does not distort the airflow pattern (ring-stand and clamp).

F.3.3 Methods

One of these methods was validated per cabinet model and provided by the manufacturer, which was reviewed and approved by the testing organization. Manufacturer validation procedures contained no fewer than ten replicate tests. The testing organization’s approval will be based on review of data and successful reproduction of test results. The validated alternate method is on the manufacturer’s data plate.

F.3.3.1 General

When the testing organization has determined the nominal set point on a given model and size of cabinet using a DIM device, and an appropriate alternative method has been validated for that cabinet by the testing organization, this alternate method may be used to establish the set point on the same model and size of cabinet in the field.

F.3.3.2 Direct inflow measurement method

a) Seal by taping the device to the center of the front opening of a biosafety cabinet. Seal the open areas on either side of the capture hood portion of the DIM as necessary.

b) All cabinet and exhaust blowers must be operating. Take at least five readings and average them to determine inflow volume rate. Care should be taken not to restrict the airflow through the instrument intake area.

c) Calculate the average inflow velocity in feet/minute (meters/second) by dividing the average inflow volume rate in cubic feet/minute (cubic meters/second) by the work access opening area in square feet (square meters).

d) Include the following in reported data: individual inflow volume rate readings, average inflow volume rate, work access opening dimensions and area, directly measured average inflow velocity, and the methods used to determine them.

e) Reported values shall be:

– Individual volume readings;
– Overall average of the volume;
– Calculated Inflow volume;
– Work access opening area;
– View screen opening height;
– Correction factor used (if applicable);
– Acceptance criteria for average airflow volume;
– Acceptance criteria for calculated inflow velocity;
– Inflow Velocity Test Method; and
– Name of Test (Inflow velocity test).

F.3.3.3 Alternate inflow measurement methods

In addition to the direct inflow method, one of the alternative methods was validated for each cabinet model and was reviewed and approved by the testing organization.
F.3.3.3.1 Method for Type A1 and A2 cabinets that use a thermal anemometer to measure exhaust velocity to determine inflow velocity

a) Take air velocity measurements at multiple points across the exhaust filter face on a grid as specified on the data plate.

b) Use the effective open area of the exhaust HEPA filter or exhaust port determined by the manufacturer and validated by the testing organization. Measure the effective exhaust area when the manufacturer has not provided it. Cabinets in which the exhaust filter is not accessible or exhaust port flow is non-uniform, such as caused by a damper or exhaust filter housing design, shall be tested as approved by the testing organization.

c) To obtain the exhaust flow volume rate in cubic feet/minute (cubic meters/second), multiply the average exhaust air velocity in feet/minute (meters/second) by the effective exhaust area in square feet (square meters).

d) Use the work access opening area as listed by the testing organization. Measure the work access opening area when the manufacturer has not provided it.

e) Calculate the average inflow velocity in feet/minute (meters/second) by dividing the average exhaust volume rate in cubic feet/minute (cubic meters/second) by the work access opening area in square feet (square meters).

f) Include the following in reported data: individual exhaust velocity readings, average exhaust velocity, exhaust volume rate, exhaust opening dimensions and area, work access opening dimensions and area, calculated average inflow velocity, and the methods used to determine them.

g) Reported values shall be:

- Individual exhaust velocity readings;
- Overall average of the exhaust velocity readings;
- Calculated exhaust volume;
- Calculated inflow velocity;
- Exhaust opening dimensions;
- Exhaust opening effective area;
- Work access opening area and dimensions;
- Correction factor used (if applicable);
- Acceptance criteria for calculated inflow velocity;
- Inflow Velocity Test Method; and
- Name of Test (Inflow velocity test).

F.3.3.3.2 Method for Type A1, A2, and B2 cabinets using a thermal anemometer to measure velocity through a constricted access opening to determine average inflow velocity

a) Restrict the access opening as specified by the testing organization.

b) Take air velocity measurements at multiple points across the restricted opening as specified on the data plate. No fewer than two readings per 1 ft (0.3 m) of access opening width shall be taken.

c) Average the air velocity measurements. Multiply the average by the listed correction factor to obtain the average inflow velocity.
d) Include the following in reported data: height of restriction, individual velocity readings, average velocity, the listed correction factor, calculated inflow velocity, and methods used to determine them.

e) Reported values shall be:

- Individual constricted velocity readings;
- Overall average of the constricted velocity readings;
- Calculated exhaust volume;
- Calculated inflow velocity;
- Constricted opening dimensions and area;
- Work access opening area and dimensions;
- Correction factor used (if applicable);
- Acceptance criteria for calculated inflow velocity;
- Inflow Velocity Test Method; and
- Name of Test (Inflow velocity test).

F.3.3.3.3 Method for Type B1 cabinets using a thermal anemometer to measure velocity through the access opening to determine average inflow velocity

a) Turn off the blower(s) that recirculate air in the cabinet, if tested that way by the testing organization.

b) Set the sash (viewing window) to the height tested by the testing organization.

c) Take air velocity measurements at multiple points across the work access opening on a grid as specified on the data plate.

d) Include individual inflow velocity readings, average inflow velocity, and methods used to determine them in the reported data.

e) Reported values shall be:

- Individual inflow velocity readings;
- Overall average of the inflow velocity readings;
- Calculated Inflow volume;
- Work access opening dimensions and area;
- Correction factor used (if applicable);
- Acceptance criteria for average inflow velocity;
- Inflow Velocity Test Method; and
- Name of Test (Inflow velocity test).

F.3.3.3.4 Calculated method for Type B2 cabinets using an anemometer and pitot tube if applicable

a) Turn on the cabinet downflow blower and exhaust system blower.

b) Set the sash (viewing window) at the height specified by the testing organization.

c) Measure and calculate the exhaust volume in accordance with the testing organization’s verified methodology, or with ASHRAE standards for air velocity measurements in round or rectangular ducts, or with the Industrial Ventilation Manual.

d) Measure the supply air velocity on a grid as specified on the data plate. The air measurement probe shall be held rigidly in a freestanding fixture (ring-stand and clamp) that permits accurate
positioning and does not distort the airflow pattern (see annex A, figure A20). Average the velocity readings and multiply by the area in square feet (square meters) of the plane in which the velocities were measured to determine the total filtered supply air volume flow rate in cubic feet/minute (cubic meters/second).

e) Subtract the supply air volume rate in cubic feet/minute (cubic meters/second) from the total exhaust volume rate in cubic feet/minute (cubic meters/second); the difference represents the calculated inflow volume rate in cubic feet/minute (cubic meters/second).

f) Divide the calculated inflow volume rate by the area of the access opening in square feet (square meters) to determine the average inflow velocity in feet/minute (meters/second).

g) Include the following in reported data: individual exhaust velocity readings, calculated average exhaust velocity, exhaust duct area, calculated exhaust volume, individual supply velocity readings, average supply velocity, effective supply area, calculated supply air volume, area of the work access opening, calculated inflow air volume, calculated average inflow velocity, and methods used to determine them.

h) Reported values shall be:

- Individual duct velocity readings;
- Overall average of the duct velocity readings;
- Calculated exhaust volume;
- Duct size, shape and area;
- Work access opening dimensions and area;
- Dimensions and area of the supply velocity measurement location (used to determine supply volume);
- Individual supply velocity readings (not to be confused with downflow velocities);
- Calculated supply air velocity and volume;
- Calculated inflow velocity and method used for calculations;
- Correction factor used (if applicable);
- Acceptance criteria for average inflow velocity;
- Inflow Velocity Test Method; and
- Name of Test (Inflow velocity test).

INFORMATIVE NOTE – Canopy connected A1 and A2 cabinets must be tested with a method that measures the inflow volume at the work access opening.

F.3.4 Acceptance

A cabinet qualifies for field certification when the average work access opening inflow velocity is within ± 5 ft/min (± 0.025 m/s) of the nominal set point verified by the testing organization using the same method.

F.4 Airflow smoke patterns test

F.4.1 Purpose

This test determines that the airflow along the entire perimeter of the work access opening is inward, that airflow within the work area is downward with no dead spots or refluxing, that ambient air does not pass on or over the work surface, and that there is no escape to the outside of the cabinet at the sides and top of the window.
F.4.2 Apparatus

A source of visible cold smoke such as titanium tetrachloride.

NOTE – Titanium tetrachloride is corrosive and should be handled with care.

F.4.3 Method

F.4.3.1 Downflow test

a) Smoke shall be passed from one end of the cabinet to the other, along the centerline of the work surface, at a height of 4 in (10 cm) above the top of the access opening.

b) Reported values shall be:
   − Name of test (Smoke Pattern Downflow Test); and
   − Pass or fail.

F.4.3.2 View screen retention test

a) Smoke shall be passed from one end of the cabinet to the other, 1.0 in (2.5 cm) behind the view screen, at a height 6.0 in (15 cm) above the top of the access opening.

b) Reported values shall be:
   − Name of test (View Screen Retention Test); and
   − Pass or fail.

F.4.3.3 Work opening edge retention test

a) Smoke shall be passed along the entire perimeter of the work opening edges, approximately 1.5 in (3.8 cm) outside the cabinet. Particular attention should be paid to corners and vertical edges.

b) Reported values shall be:
   − Name of test (Work Opening Edge Retention Test); and
   − Pass or fail.

F.4.3.4 Sash/window seal test

a) Smoke shall be passed up the inside of the window 2 in (5 cm) from the sides and along the top of the work area.

b) Reported values shall be:
   − Name of test (Sash/Window Seal Test); and
   − Pass or fail.

F.4.4 Acceptance

F.4.4.1 Downflow test

The smoke shall show smooth downward flow with no dead spots or reflux (upward flow).
F.4.4.2 View screen retention test

The smoke shall show smooth downward flow with no dead spots or reflux. No smoke shall escape from the cabinet.

F.4.4.3 Work opening edge retention test

No smoke shall be refluxed out of the cabinet once drawn in, nor shall smoke billow over the work surface or penetrate onto it.

F.4.4.4 Sash/window seal test

There shall be no escape of smoke from the cabinet.

F.5 HEPA filter leak test

F.5.1 Purpose

This test determines the integrity of downflow and exhaust HEPA filters, filter housings, and filter mounting frames. The cabinet shall be operated within ± 5 ft/min (0.025 m/s) of the nominal set point, with the exception of the downflow HEPA filters on B1 cabinets.

F.5.2 Apparatus

F.5.2.1 An aerosol photometer with linear or expanded logarithmic scale shall be used. The instrument shall be capable of indicating 100% upstream concentration with a minimum aerosol concentration of 10 μg/L of polydisperse dioctylphthalate (DOP) particles, or an equivalent fluid that provides the same particle size distribution (e.g., polyalpha olefin [PAO] di[2-ethylhexyl] sebacate, polyethylene glycol, and medicinal-grade light mineral oil) produced by the generator described in annex A, section A.3.2.2 or equivalent. It shall also be capable of detecting an aerosol concentration in the downstream equal to 10⁻⁵ of the upstream concentration of the same particles. The sampling rate of air shall be 1 ft³/min (5 x 10⁻⁴ m³/s) ± 10%. Probe area shall have a maximum open area of 1.7 in² (11 cm²) and a minimum dimension of 0.50 in (1.3 cm). The photometer shall be set up in accordance with the photometer manufacturer’s instructions or IEST-RP-CC-013 if instructions are not provided.

F.5.2.2 An aerosol generator of the Laskin Nozzle type conforming to annex A, Figure A2 or equivalent shall be used to create an aerosol by flowing air through liquid DOP or equivalent substitute. When a Laskin nozzle generator is used, the compressed air supplied to the generator should be adjusted to a minimum of 20 psi (140 kPa), measured at the generator manufacturer’s recommended location. The nozzles shall be covered with liquid to a depth not to exceed 1.0 in (2.5 cm).

F.5.2.3 A pressure gauge for the generator having a maximum range of 0 to 80 psi (0 to 550 kPa) with resolution and accuracy of 1 psi (7 kPa) calibrated by the manufacturer or in accordance with the manufacturer’s instructions shall be used.

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F.5.3 Method of testing HEPA filters

F.5.3.1 Filters that can be scanned

a) Turn on the cabinet blower and lights (types A1 and A2 and B2 downflow filter test). Remove the filter diffusers and protective covers if any are present. Place the generator so the aerosol is introduced into each cabinet fan upstream of the HEPA filter(s). When the manufacturer has not identified the aerosol introduction point(s), introduce the aerosol in a manner to ensure thorough mixing in the cabinet airflow. For example, a T-connection can be fitted to the aerosol generator output to enable distribution of challenge into both entrances of a single blower or entrances of multiple blowers. The manufacturer shall determine the aerosol introduction point that provides the most uniform distribution.

b) Turn on the photometer and adjust it in accordance with the manufacturer's instructions.

c) Determine the aerosol concentration upstream of the HEPA filter.

- When the challenged airflow is not contaminated, sample the aerosol concentration upstream of the HEPA filter.

- When the challenged airflow is contaminated or when measuring the upstream concentration is not practical, the upstream concentration can be calculated. For example, when DOP is used as the challenge aerosol with a Laskin nozzle aerosol generator at 20 psi (140 kPa), the following formula applies:

\[ \mu g/L = 13,500 \times \text{number of nozzles} / \text{ft}^3/\text{min of challenged air} \]

NOTE – Use of DOP substitutes will require modification of this formula, unless the photometer is calibrated with the substitutes to yield results equivalent to those of DOP. Use of DOP substitutes will also require pressures different from 20 psig.

- Use an aerosol concentration that is at least equal to the photometric equivalent of 10 \( \mu g/L \) of DOP.

d) Set up the photometer to the upstream challenge in accordance with the photometer manufacturer’s instructions to detect leaks greater than or equal to 0.01% of the upstream concentration.

e) With the nozzle of the probe held not more than 1.0 in (2.5 cm) from the area being tested, scan the entire downstream side of the HEPA filter(s) and the perimeter of each filter pack by passing the photometer probe in slightly overlapping strokes at a traverse rate of not more than 2 in/s (5 cm/s). Separate passes shall be made around the entire periphery of the filter, along the bond between the filter pack and frame, and around the seal between the filter and the device.

f) Reported values shall be:

- Upstream aerosol challenge concentration;
- Method used to report concentration (measured or calculated);
- Maximum leak penetration in percent;
- Method used (scanned or probe tested); and
- Name of Test (HEPA filter leak test).

F.5.3.2 Filters that cannot be scanned

a) When a cabinet is ducted so that the exhaust filter cannot be scanned, it may be leak tested by drilling a hole approximately 0.3 in (1 cm) in diameter in the duct at a downstream location that
will produce a well-mixed aerosol and inserting the photometer sampling probe with rigid extension tubing through the hole.

b) Reported values shall be:

- Upstream aerosol challenge concentration;
- Method used to report concentration (measured or calculated);
- Maximum leak penetration in percent;
- Method used (scanned or probe tested); and
- Name of Test (HEPA filter leak test).

F.5.4 Acceptance

F.5.4.1 Filters that can be scanned

Sustained aerosol penetration shall not exceed 0.01% of the upstream concentration at any point.

F.5.4.2 Filters that cannot be scanned

Sustained aerosol penetration shall not exceed 0.005% of the upstream concentration.

F.6 Pressure decay / soap bubble

F.6.1 Pressure decay or soap bubble test

F.6.1.1 Purpose

The pressure decay or soap bubble test is performed to determine whether exterior surfaces of all plenums, welds, gaskets, and plenum penetrations or seals are free of leaks.

F.6.1.2 Apparatus

- manometer, pressure gauge, or pressure transducer system with a minimum range of 0 – 2 in w.g. (0 – 500 Pa) and accurate to ± 0.02 in w.g. (± 5 Pa);
- liquid leak detector;
- plastic sheet (0.02 in extruded high-impact styrene); and
- duct tape.

F.6.1.3 Method (pressure decay)

a) Prepare the cabinet as a closed system, i.e., seal the front window and exhaust port.

b) Remove decorative panels and other access obstructions, wherever necessary, to expose the plenums to be tested.

c) Attach a manometer, pressure gauge, or pressure transducer system to the test area to indicate the interior pressure (see annex A, figure A1a).

d) Pressurize the cabinet with air to a reading of 2 in w.g. (500 Pa), turn off the pressurizing air, and measure the pressure after 30 min. A leakage of 10% of the original pressure is allowable. If a cabinet does not hold 2 in w.g. (500 Pa), use the soap bubble method to locate leaks.
e) Reported values shall be:
   - Pressure range maintained during test;
   - Pass or fail; and
   - Name of Test (Pressure Decay Test).

F.6.1.4 Method (soap bubble)

a) Prepare the cabinet as a closed system, i.e., seal the front window and exhaust port.

b) Remove decorative panels and other access obstructions, wherever necessary, to expose the plenums to be tested.

c) Attach a manometer, pressure gauge, or pressure transducer system to the test area to indicate the interior pressure (see annex A, figure A1a).

d) Pressurize the cabinet with air to ensure a continuous reading of 2 in. w.g. (500 Pa) ± 10%.

e) Spray or brush the liquid leak detector along all welds, gaskets, penetrations, and seals on exterior surfaces of cabinet plenums. Small leaks will be indicated by bubbles. Large leaks will occur that blow the detection fluid from the hold without forming bubbles and may be detected by slight feel of airflow or sound.

f) Reported values shall be:
   - Pressure range maintained during test;
   - Pass or fail;
   - Description of leak location(s); and
   - Name of Test (Soap Bubble Leak Test).

F.6.1.5 Acceptance

A cabinet qualifies for field certification when the cabinet holds 2 in. w.g. (500 Pa) within ± 10% for 30 min or when all welds, gaskets, penetrations, and seals on exterior surfaces of air plenums are free of soap bubbles when at 2 in. w.g. (500 Pa) pressure above atmospheric.

F.7 Site installation assessment tests

F.7.1 Purpose

These tests are performed to verify that the biosafety cabinet is integrated properly into the facility.

F.7.2 Apparatus

   - owner’s manual; and
   - a visible source of cold smoke such as titanium tetrachloride.

F.7.3 Method

F.7.3.1 Alarm functions
F.7.3.1.1 Airflow alarms (excluding building automation systems)

a) Whenever an alarm is present to monitor the performance of airflow, it must be performance verified. The alarms shall be performance verified at every certification.

b) The procedures outlined in the owner’s manual shall be followed. Once the cabinet is set or certified in its acceptable airflow range, audible and visual alarms shall be required to indicate a 20% loss of exhaust volume within 15 sec.

F.7.3.1.2 Sash alarms

The sash shall be raised 1.0 in (2.5 cm) above the manufacturer’s recommended height. Signaling of an audible alarm shall be verified.

F.7.3.2 Interlocks

Supply fan interlock on B cabinets:

a) Shall be tested at time of alarm verification.

b) Reduce exhaust volume 20% once the cabinet is set or certified in its acceptable airflow range, and verify that audible and visual alarms indicate a 20% loss of exhaust volume within 15 sec. The internal cabinet fan(s) shall be interlocked to shut off at the same time the alarms are activated.

F.7.3.3 Exhaust system performance – canopy connection

Using a visible smoke source, verify negative pressure at the gap. No smoke shall escape into the room once it enters the exhaust system.

NOTE – For hard ducted hoods, measure the static pressure in the duct-work between the hood and duct-mounted balancing dampers.

F.8 Electrical leakage and ground circuit resistance and polarity tests

All new cabinets shall conform to UL 61010A-1. Older cabinets may refer to NSF 49 – 1992 for Electrical Leakage, Ground Circuit Resistance, and Polarity tests if necessary.

F.9 Lighting intensity test

F.9.1 Purpose

This test is performed to measure the light intensity on the work surface of the cabinet in foot-candles (lux) as an aid in minimizing cabinet operator’s fatigue.

F.9.2 Apparatus

A portable photoelectric illumination meter approved for field measurements in accordance with the current edition of the *Illuminating Engineering Society Lighting Handbook* and accurate to ± 10%.

---

40 IES, 120 Wall Street, Floor 17, New York, NY 10005 www.iesna.org
F.9.3 Method

a) Measure the background lighting intensity along the side-to-side centerline of the work tray on a uniform linear pattern close to but no greater than 12 in (30 cm) starting 6.0 in (15 cm) from the sidewalls (annex A, figure A4).

b) Turn on the lights and blower, and take readings at the same points again.

c) Reported values shall be:
   - Individual background readings;
   - Individual lighting Intensity readings;
   - Average background intensity;
   - Average lighting intensity;
   - Acceptance criteria;
   - Pass or fail; and
   - Name of Test (Lighting intensity test).

F.9.4 Acceptance

A cabinet qualifies for field certification when average lighting intensities are no less than 45 ft-candles (480 lux) greater than background levels, where background light levels average a maximum of 15 ft-candles (160 lux).

F.10 Vibration test

F.10.1 Purpose

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.

F.10.2 Apparatus

A vibration analyzer with a minimum reliable reading of $1 \times 10^{-4}$ in (2.5 μm) rms amplitude, or the ability to detect differences of this magnitude, in accordance with manufacturer’s instructions.

F.10.3 Method

a) Operate the cabinet with lights on within 5.0 ft/min (0.025 m/s) of the nominal set point velocities.

b) To determine the vibration displacement on the vertical axis, affix the sensing element of the vibration pickup unit firmly onto the geometric center of the work surface(s) by:
   - a clamp;
   - a bolt; or
   - an integral magnet with petroleum jelly film, or a double-faced adhesive tape.

   The test position is shown in annex A, figure A5.

c) Determine the gross vibration amplitude with the cabinet operating.
d) Determine the background vibration amplitude with cabinet blower(s) off, and if applicable, the exhaust blower on.

e) Subtract the background from the gross vibration amplitude to determine the net vibration amplitude attributable to the cabinet.

f) Reported values shall be:
   - Unit "On" vibration reading;
   - Background vibration reading;
   - Net vibration;
   - Pass or fail; and
   - Name of Test (Vibration Test).

F.10.4 Acceptance

A cabinet qualifies for field certification when net displacement does not exceed 0.002 in (50 μm) rms amplitude at 10 Hz to 10 kHz in the center of the work surface(s) when the cabinet is operating at the manufacturer's recommended airflow velocities.

F.11 Noise level tests

F.11.1 Purpose

This test is performed to measure the noise levels produced by the cabinet as a guide to satisfactory mechanical performance and an aid in minimizing cabinet operator's fatigue. The procedures can be performed in most acoustically ordinary rooms, such as a factory, where walls are neither sound absorbing nor completely sound reflecting.

F.11.2 Apparatus

A sound level meter having a minimum accuracy of ± 1 db and resolution of 1 db with a minimum range of at least 50 to 100 db and an "A" weighting scale set up in accordance with the manufacturer's instructions.

F.11.3 Method

a) Operate the cabinet within 5 ft/min (0.025 m/s) of the nominal set point with lights on.

b) Set the instrument to the "A" weighting mode.

c) Measure the noise level 12 in (30 cm) in front of the cabinet (leading front edge of the access opening) and 15 in (38 cm) above the plane of the work surface, in line with the vertical centerline of the cabinet (annex A, figure A3).

d) To measure the ambient noise level, turn the cabinet blower and lights off, and if applicable, leave the remote exhaust blower on and measure as in c) above.

e) Reported values shall be:
   - Unit "On" sound level reading;
   - Background sound level reading;
   - Net sound level;
F.11.4 Acceptance

A cabinet passes the Sound Level Measurement test when the overall noise level in front of the cabinet does not exceed 70 dbA when measured where the maximum ambient sound level is no greater than 60 dbA. When the ambient sound level is greater than 60 dbA, the reading obtained in annex F, section F.11.3c) shall be corrected in accordance with curves or tables provided in the instrument operator’s manual. If this information is not available, standard correction curves or tables shall be used (see below).

Correction chart for sound level readings

<table>
<thead>
<tr>
<th>Difference between total and background sound readings in dbA</th>
<th>Number to subtract from total to yield corrected noise level</th>
</tr>
</thead>
<tbody>
<tr>
<td>0-2</td>
<td>reduce background levels</td>
</tr>
<tr>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>4-5</td>
<td>2</td>
</tr>
<tr>
<td>6-10</td>
<td>1</td>
</tr>
<tr>
<td>&gt;10</td>
<td>0</td>
</tr>
</tbody>
</table>

F.12 Record of field certification

A cabinet that has met all the field test criteria listed in annex F shall have the following information posted on the front of the cabinet in a location readily visible to the user, unless otherwise specified by the user:

F.12.1 Certification Label

Biosafety cabinets field tested to this standard shall include the following information:

- date of certification;
- date cabinet should be recertified: no later than ____________;
- certifier’s report number (reference document showing tests performed and results);
- name, address, and telephone number of certifying company; and
- signature of the person who performed the field certification tests.

F.12.2 Certification Report

A certification report that will carry the language “certified in accordance with NSF annex F” or any similar language shall, at a minimum, include the following:

1. BSC model number
2. BSC serial number
3. BSC location
4. BSC venting information
   a. (Ducted or not ducted)
      i. Type of connection (canopy, direct or none)
4. Type of BSC
5. Test equipment used for each test:
   a. Manufacturer
   b. model
   c. serial number
   d. calibration date
7. Specific test data as detailed in annex F
8. Acceptance criteria for each test
9. Printed name of certification technician
10. Test date
11. Retest date
Annex G\textsuperscript{41}  
(informative)

**G.1 Biosafety Consultation Prior to Biosafety Cabinet (BSC) Purchase**

A biosafety officer or qualified safety professional should be consulted prior to a BSC purchase. Some institutions have biosafety cabinet purchases approved by the biosafety officer or qualified safety professional after consultation with the user, architect, and engineer. Biosafety officers or qualified safety professionals that perform this function should have training and field experience that includes methods used to control biohazards and knowledge of the design, application, and testing of biosafety cabinets.

Issues that may be considered include:
- Risk assessment;
- Selecting which kind of BSC is required and if it should be exhausted; and
- Assessment of the laboratory environment and the proper location of BSCs within it.

**G.2 Risk Assessment Procedure**

**G.2.1 Risk assessments encompass four main elements:**
- Hazard identification;
- Exposure assessment;
- Dose-response assessment; and
- Risk characterization, and Risk management (job analysis)\textsuperscript{42}.

**G.2.2 Risk assessment team members may include:**
- Investigator/Scientist;
- Laboratory staff;
- Animal care staff when appropriate;
- Animal veterinarian when appropriate; and
- Occupational health & biosafety professionals.

**G.2.3 Risk assessment hazards considered:**
- Animal hazards;
- Agent/pathogen/recombinant hazards;
- Chemical hazards; and
- Radiological hazards.

\textsuperscript{41} The information contained in this Annex is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI's requirements for an ANS. Therefore, this Annex may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.

G.2.4 Agent/pathogen/recombinant’s factors associated with risk of disease or injury:

- Virulence;
- Infectious dose;
- Route of infection (portal of entry);
- Toxigenicity;
- Agent’s host range;
- Availability of effective preventive measures; and
- Availability of effective treatment.

G.2.5 Factors associated with worker’s risk of exposure:

- Worker’s work activity; diagnostic, research or production scale;
- Worker’s proficiency, attitude and safety awareness; and
- Worker’s age, sex, pregnancy, race, immune status, and medications.

G.2.6 Risk management plan includes:

- Biosafety containment level assignment to the facility and microbiological practices;
- Safety equipment;
- Engineering controls;
- Personal protective equipment;
- Work practices – Standard Operating Procedures (SOPs);
- Emergency procedures;
- Work schedule – calendar; and
- Investigation protocols that include all risk management plans.

G.2.7 Investigation protocol review includes:

- Committee (IBC/IRB/IACUC) review, as appropriate;
- Meetings with workers to discuss approved protocols;
- Training;
- Dry runs without agent/pathogen/recombinant; and
- Regular audits.
### G.2.8 Risk Management Analysis Table:

<table>
<thead>
<tr>
<th>Risk Factor Assessment</th>
<th>Data collection</th>
<th>Increase or decrease</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Agent Identity</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Known, classified</td>
<td>Agent summary statement</td>
<td>Same</td>
</tr>
<tr>
<td>Suspected, classified</td>
<td>Agent summary statement</td>
<td>Same</td>
</tr>
<tr>
<td>Known, unclassified</td>
<td>Disease Information</td>
<td>Increase</td>
</tr>
<tr>
<td>Unknown</td>
<td>Disease Information</td>
<td>Increase</td>
</tr>
<tr>
<td><strong>Agent Transmission</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Aerosol potential (Sample/activity)</td>
<td>Tissue procedure</td>
<td>Decrease</td>
</tr>
<tr>
<td></td>
<td>Culture procedure</td>
<td>Increase</td>
</tr>
<tr>
<td></td>
<td>Concentration procedure</td>
<td>Moderate Increase</td>
</tr>
<tr>
<td></td>
<td>Animal/non-shedder</td>
<td>Decrease</td>
</tr>
<tr>
<td></td>
<td>Animal/shedder</td>
<td>Moderate Increase</td>
</tr>
<tr>
<td><strong>Infectious route</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Respiratory</td>
<td>Moderate Increase</td>
<td></td>
</tr>
<tr>
<td>Mucous membrane</td>
<td>Increase</td>
<td></td>
</tr>
<tr>
<td>Parenteral</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td><strong>Disease Severity</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Moderate</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Severe</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Life threatening/lethal</td>
<td>Severe Increase</td>
<td></td>
</tr>
<tr>
<td><strong>Prophylaxis</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>None</td>
<td>Severe Increase</td>
<td></td>
</tr>
<tr>
<td>Vaccine</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Immune globulin</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Antibiotics</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td>Antiviral</td>
<td>Decrease</td>
<td></td>
</tr>
<tr>
<td><strong>Treatment</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>None</td>
<td>Severe Increase</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>decrease</td>
<td></td>
</tr>
<tr>
<td><strong>Other Factors</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Livestock pathogen</td>
<td>USDA regulations</td>
<td>Increase</td>
</tr>
<tr>
<td>Poultry pathogen</td>
<td>USDA regulations</td>
<td>Increase</td>
</tr>
</tbody>
</table>

### G.3 Selection of a BSC cabinet

**G.3.1** Selecting the proper BSC should be done in two stages; first, select the proper class and type of unit required, then decide on the size of the unit and options that are needed. Deciding which class and type is appropriate can be accomplished by answering the following four questions:
G.3.1.1. What needs to be protected?

- Only the material being worked on (Product protection)?
- Only the technician and the laboratory (Personnel and environmental protection)?
- Alternatively, to protect all three (personnel, product, and environmental protection)?

If all that is needed is product protection, then a Clean Bench may be the unit of choice. Clean Benches use High Efficiency Particulate Air (HEPA) filter(s) to remove particulates from room air. This filtered, particulate-free air then flows through an enclosed work area, in a horizontal or vertical direction. These devices bathe the materials inside in filtered air, and then the air is typically discharged into the laboratory. While these devices protect the product from airborne contaminants, any aerosol generated in the work area will be discharged into the laboratory. As such, they cannot be used with toxic or biohazardous materials. Most microbiological work includes at least the potential for future work at BSL2 or higher, so it is relatively infrequent for a Clean Bench to be selected for microbiological work.

For personnel and environmental protection only, the Class I enclosure offers a simple and economical solution. Room air sweeps around the operator and through the work area. This contaminated air is then HEPA filtered and discharged either into the laboratory or out via an exhaust system. The Class I will protect the operator and the lab, however, because room air constantly washes over the work area, the product is exposed to airborne contaminants. Most microbiological work includes at least some concern to limit the potential for contamination of the work, so it is relatively infrequent for a Class I BSC to be selected for microbiological work.

Personnel, environmental, and product protection can be had most efficiently by a Class II Biohazard Cabinet. The inflow of air around the operator provides personnel protection. HEPA filtered air flowing downward through the work area provides product protection, and HEPA filtered exhaust protects the laboratory from biohazardous particulates.

G.3.1.2. What are all of the different types of work to be done in the cabinet?

One of the most difficult tasks in selecting a BSC is trying to foresee all the different types of work that will be taking place in it. It is critical to decide what things need protection, both now and in the future. All too often users purchase a Clean Bench or Class I device for current applications, only to find these devices are unsuitable as their work requirements change.

G.3.1.3. What types and quantities of volatile toxic chemicals will be used in the BSC?

As important as the preceding question, the user must also foresee the types and quantities of volatile toxic chemicals that will be used in the cabinet. Because volatile chemicals and other gases freely pass through HEPA filters, both Class I and Class II BSCs should be exhausted out of the laboratory when used with these types of toxic chemicals. For the Class II BSCs, Types B1 and B2 must be hard ducted to an external exhaust system in order to operate properly; Types A1 and A2 can be converted to operate in either a ducted (canopy connected) or recirculating mode. When properly ducted through a canopy connection, a Class II, Type A2 BSC can be used with minute quantities of volatile toxic chemicals.
### G.3.1.4 Characteristics of Type A and Type B BSCs

<table>
<thead>
<tr>
<th></th>
<th>Type A</th>
<th>Type B</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Intended Purpose</strong></td>
<td>Routine microbiological work. Biologic work with volatile toxic chemicals and/or radionuclides if canopy-connected.</td>
<td>Biological work using volatile toxic chemicals or radionuclides.</td>
</tr>
<tr>
<td><strong>Biological Containment</strong></td>
<td>Equal to Type B.</td>
<td>Equal to Type A.</td>
</tr>
<tr>
<td><strong>Exhaust System</strong></td>
<td>Optional as needed</td>
<td>Required</td>
</tr>
<tr>
<td><strong>Type</strong></td>
<td>May be ganged into multiple-cabinet exhaust system, if all BSCs are balanced properly? Is this the recommendation?</td>
<td>Should have dedicated ductwork and exhaust blower for each BSC.</td>
</tr>
<tr>
<td><strong>Function</strong></td>
<td>Only pulls exhaust air through the ductwork.</td>
<td>Must pull exhaust air through the Cabinet’s Exhaust HEPA filter and then through ductwork.</td>
</tr>
<tr>
<td><strong>Volume</strong></td>
<td>Less than Type B. (less than B2 but not less than B1)</td>
<td>More than corresponding Type A; Type B2 exhausts 30-50% more than a Type B1.</td>
</tr>
<tr>
<td><strong>Vacuum</strong></td>
<td>Typically 0.5 – 1.0 inches H₂O.</td>
<td>Typically, 2.0 inches H₂O minimum; Maximum may exceed 4.0 inches H₂O.</td>
</tr>
<tr>
<td><strong>Reserve Capacity</strong></td>
<td>Vacuum requirements will not change as the cabinet filters load.</td>
<td>Vacuum requirements may increase up to 2.0 inches H₂O as exhaust HEPA filter loads.</td>
</tr>
<tr>
<td><strong>Cabinet Flexibility</strong></td>
<td>Can be connected or disconnected from exhaust system as needed change.</td>
<td>Must be permanently connected to an exhaust system to function.</td>
</tr>
<tr>
<td><strong>Cabinet Cost</strong></td>
<td>Less than Type B</td>
<td>More expensive than Type A</td>
</tr>
<tr>
<td><strong>Installation Cost</strong></td>
<td>Less than Type B if recirculating; similar to Type B if canopy-connected.</td>
<td>More expensive than Type A; higher volumes may require larger ductwork.</td>
</tr>
<tr>
<td><strong>Operation Cost</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Electrical (BSC Only)</strong></td>
<td>Equal to Type B</td>
<td>Equal to Type A</td>
</tr>
<tr>
<td><strong>Tempered air loss</strong></td>
<td>If recirculating in lab; none. If canopy-connected, typically 100 CFM/foot of BSC width or less.</td>
<td>More expensive than any Type A configuration. Typically 150 CFM/foot of BSC width. (Again B1 is a little less)</td>
</tr>
</tbody>
</table>

#### G.3.1.5. If the unit requires an exhaust system, is there an appropriate location for the cabinet and its ductwork?

If a BSC is going to recirculate its HEPA-filtered air back into the laboratory, then the user has some freedom as to where the unit can be installed, provided it is out of major traffic areas, and there are no other air handling devices in the area.

When connected to a hard-ducted exhaust system, however, the location of the cabinet becomes dependent on the location of the exhaust system. The exhaust duct must be placed so it can penetrate ceilings and floors without disturbing other ventilation or plumbing systems. The exhaust system must also be designed to minimize excessive lengths and elbows.
G.3.2. BSC Size

Having decided which class and type of BSC is the best, the user should now decide on the size of the unit and its options. In deciding which size is best, the user should mark out an area of benchtop equal to the inside (work area) dimensions of the model they are interested in. The user(s) should perform several "dry runs" of their procedures in this area. If the user can work in this defined space, than the cabinet is the proper size, if not, the user may want to try working in the dimensions of the next larger model. If the user does decide on a larger model, however, be sure that the BSC can be transported to and installed in the laboratory through the existing freight elevators, hallways, and doors. It is important to remember that BSC widths typically refer to the internal work area. The external width of the BSC may be significantly wider.

G.3.3 BSC Options

G.3.3.1 Service Valves

Service valves allow inert gases, air, or vacuum lines to be plumbed into the BSC. Although many users connect natural gas to a service valve in the cabinet, this practice should be avoided if possible, because open flames in a Class II BSC disrupts the airflow, and there is the possibility of a buildup of flammable gas in BSCs that recirculate their air. Many models allow for the easy installation of these valves in the field, however, it is generally less expensive and easier to have the required number of valves installed when the unit is ordered.

G.3.3.2 Electrical Outlets

Most BSCs have electrical outlets installed in the work area as standard equipment. Specialized fittings, such as Ground Fault Interrupter Circuits (GFICs) should be installed and tested by the cabinet manufacturer.

G.3.3.3 Ultraviolet Lighting

Germicidal (or UV) Lamps are often installed as an adjunct to surface disinfection. UV lighting is not recommended in Class II (laminar flow) Biosafety cabinetry. While their usefulness is a subject for debate among users and manufacturers, they should be installed and tested by the manufacturer during assembly of the unit.

G.3.3.4 IV Bar

Because intravenous (IV) bars or rods have a significant impact on the airflows in the work area, always use the IV bar recommended by the manufacturer.

G.3.3.5 Base Stands

Base Stands or supports should also be considered at the time of specification. Some models of cabinets can weigh up to 900 pounds. The BSC must be attached to a manufacturer recommended base stand or a structure rated to support the unit's weight.

G.4 Prior to the Purchase

G.4.1 Investigators should consult with a biosafety officer or qualified safety professional request a risk assessment of the proposed investigation to ensure that an appropriate BSC is used for the work. Purchase of NSF/ANSI 49 listed Class II biosafety cabinets is recommended, but alternative containment equipment may be suggested for special tasks.
G.4.2 The investigator should notify building management to arrange for a feasibility assessment of laboratory alterations and BSC location can be completed. The investigator and biosafety officer or qualified safety professional should discuss the following points about the BSC and its delivery:

- Make sure all arrangements are planned in advance of the BSCs arrival,
- Get a written price quote for the entire package, including the BSC Model number, optional equipment, canopy exhaust connection, etc. Work out the details about shipping and delivery with the manufacturer's representative at the time of purchase,
- Determine the costs for shipping and delivery because there may be additional costs depending on delivery location and level of difficulty,
- Make sure that the sales representative clarifies in writing what "shipping and delivery" includes; does delivery include moving the BSC from the receiving dock of the building to the laboratory and does delivery also include BSC set-up in the work area?
- There are options for moving BSCs from a loading dock to a laboratory, such as hiring moving contractors to uncrate and move the BSC,
  - Make sure the corridor pathways are clear for delivery to the laboratory,
  - Will the BSC fit through door jams?
  - Will the BSC travel around sharp, narrow corridors and corners?
  - Will the elevators in the building accommodate the BSC?
  - Does the BSC have to be brought up steps?
  - The moving contractor should be advised that the BSC should be lifted onto its stand or leg extensions (working position), because a hydraulic lift may be needed.

G.5 Inspection

G.5.1 When the BSC arrives, inspect it carefully. Compare the invoice with the delivered equipment. Check for any damage or missing materials and report them immediately to the proper carrier and the BSC supplier regardless of how insignificant they may first appear. Be careful of sharp crating material and let the loading dock personnel help check for damage.

G.5.2 Arrange for certification after the BSC is installed. Building operations personnel may be needed to connect the BSC to laboratory plumbing, electrical, and supply/exhaust air ventilation systems.

G.6 Moving a Biosafety Cabinet

G.6.1 It is a common practice to move BSCs to other locations within a laboratory or to other laboratories. Despite the apparent simplicity of the job, there are certain conditions that must be met prior to moving this equipment. BSCs should not be moved without consultation with a biosafety officer or qualified safety professional.

G.6.2 Existing BSCs and ancillary equipment, such as canopy connection exhaust ducting, gas, electric and vacuum connections, should be cleared for maintenance by a biosafety officer or qualified safety
professional prior to disassembly. Prior to a move, BSCs should be space decontaminated. After a BSC is moved, it should be certified according to applicable performance standards.

G.7 Recommended microbiological decontamination procedure

G.7.1 Microbiological decontamination

Space decontamination is mandatory when maintenance work, filter changes, and performance tests require access to any contaminated portion of the cabinet. All work surfaces and exposed surfaces should be decontaminated with a suitable surface disinfectant before certification tests are performed and before gaseous decontamination takes place. In addition, it may be desirable to perform gaseous decontamination of the entire cabinet before performing certification tests when the cabinet has been used with agents assigned to Biosafety Level 2, and is recommended when the cabinet has been used with an agent assigned to Biosafety Level 3. A qualified safety and risk assessment of cabinets potentially contaminated with biological agents should be performed by a biosafety officer or qualified safety professional. Appropriate decontamination (space and/or surface) should be performed before BSCs are moved to another location. Additionally, after spills and splashes of research agents, contaminated surfaces should be suitably decontaminated.

G.7.2 Certification of Cabinet Decontamination

BSCs must be decontaminated prior to decommissioning and salvage, before physically moving the cabinet and whenever maintenance work or filter changes or performance tests require access to any contaminated portion of the cabinet.

G.7.2.1 Biological Decontamination

Surface decontaminate accessible work surfaces with either chlorine dioxide or formaldehyde. Rinse work surfaces with water and then wipe dry. Use formaldehyde gas or an acceptable alternative space decontamination procedure to decontaminate the HEPA filters and cabinet interior spaces. Remove and discard all HEPA filters and any prefilters. Rinse work surfaces with water and wipe dry.

G.7.2.2 Chemical, Radiological, Oil, or Heavy Metal Decontamination

Surface decontaminate accessible work surfaces with an appropriate disinfectant and/or cleaning agent wipe down. Use formaldehyde gas or an acceptable alternative space decontamination procedure if biological agents may be present. Rinse work surfaces with water and wipe dry. Remove and discard all HEPA filters and any prefilters.

DECONTAMINATION FORM (Sample)

BSC MODEL Number _________ Serial Number ________________

1. Check each type of hazardous material that has been used or is contained in this equipment. If there has been no contamination, check "NONE" for each hazard.

2. List decontamination procedure and product used for decontamination

3. Indicate biosafety level of facility where cabinet was used:

   BSL-1  ____  BSL-2  ____  BSL-3  ____  BSL-4  ____  Not applicable  ____

4. Complete and sign the certification below,

<table>
<thead>
<tr>
<th>CONTAINED HAZARD (v')</th>
<th>DECONTAMINATION PROCEDURE</th>
<th>NONE</th>
<th>HAZARD TYPE</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>BIOLOGICAL</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>CHEMICAL</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>RADIOLOGICAL</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>OIL, HEAVY METAL (e.g. lead, mercury, or other hazardous material)</td>
</tr>
</tbody>
</table>

I hereby certify that this equipment has been decontaminated and thoroughly cleaned in accordance with the appropriate procedures (or that the equipment has not been used with any of the materials listed above).

__________________________________________  ____________
Signature of last user or biosafety officer              Date

__________________________________________  ____________
Name (PLEASE PRINT)                                      Title

__________________________________________  ____________
Room Number                                             Phone Number
G.7.3 Decontamination Methods

In most instances where space decontamination is necessary, one of the procedures described below utilizing either depolymerized paraformaldehyde or chlorine dioxide gas is used. Prior to decontamination with an alternative method (such as vaporous hydrogen peroxide [VHP]), cycle parameters and validation of those parameters must be developed for each model and size of BSC. Material compatibility in terms of degradation and absorption of an alternative decontaminant are critical for maintaining cabinet integrity and the time required for decontamination, respectively. Alternate methods are required in certain instances, e.g., slow disease viruses. The decontamination method should be determined by consultation between user and certification agency. When paraformaldehyde is used for gas decontamination, follow OSHA Regulations Code of Federal Regulations, Title 29, Formaldehyde-1910-1048, which addresses monitoring; posting of regulated areas; respirator selection, protection and fit testing; medical surveillance; hazard communication and training; and recordkeeping. Automatic formaldehyde gas decontamination/neutralization may be used as a substitute to the formaldehyde procedure given below if the manufacturer's instructions have been followed. When using chlorine dioxide gas, similar precautions as used for formaldehyde should be followed. Similarly, automated chlorine dioxide gas systems are available which may be used if the manufacturer's instructions are followed.

G.7.3.1 Paraformaldehyde

CAUTION – All sources of hydrogen chloride must be removed from the cabinet before decontamination. Hydrogen chloride in the presence of formaldehyde, at ambient air conditions, will form the carcinogen Bis(chloromethyl)ether (BCME)\(^{44}\).

a) Calculate the total volume of the cabinet by multiplying the height, width, and depth.

b) Multiply the total volume of the cabinet by 0.30 g/ft\(^3\) (11 g/m\(^3\)) of space to determine the gram weight of paraformaldehyde required [CHECK CONCENTRATION]. Determine the stoichiometric amount of \(\text{NH}_4\text{HCO}_3\) or alternative to neutralize the resulting formaldehyde gas with ammonia gas. The ammonium bicarbonate should be weighed out so that it is 10% greater than the weight of paraformaldehyde used for the decontamination to ensure completion of the reaction.

c) If the cabinet is equipped with an exhaust duct, this duct must be gas tight. This may be accomplished at the terminal end of the duct, or if present, at the damper located near the cabinet. If the exhaust duct is more than 10 ft (3 m) long, additional paraformaldehyde may be needed to compensate for the increased volume. If the cabinet exhausts into a recirculating building exhaust system, disconnect the cabinet from the building system and form a gas tight seal (plastic film and tape may be used).

d) If the cabinet exhaust air is discharged into the room, tape a plastic cover over and completely seal the exhaust port.

e) To provide for emergency evacuation of the formaldehyde and to allow removal of the neutralized formaldehyde following the decontamination and neutralization, a flexible hose can be pre-positioned close to the cabinet. This hose must be attached to a chemical fume hood or other exhaust suitable for the evacuation of toxic fumes.

f) Place a heating device, such as a commercially available electric frying pan or a remote formaldehyde generator/neutralizer, with the thermostat set at 450 to 475 °F (232 to 246 °C), on the work tray. The paraformaldehyde is spread evenly over the heating surface of the heating device.

\(^{44}\) NIOSH, Department of Health and Human Services (DHHS), *Hazard Review of Bis(chloromethyl)ether (BCME)*
CAUTION – The auto-ignition temperature of paraformaldehyde is 572 °F (300 °C).

g) Place an additional heating device on the work tray for the neutralizing agent. The neutralizing agent (NH₄HCO₃ or equivalent) should be separated from the air in the cabinet until needed. Below are two examples of how this separation could be achieved.

– Example 1: The NH₄HCO₃ or equivalent alternative is spread evenly over the heating surface of the heating device. The top of the device is covered with aluminum foil in such a way as to prevent the NH₄HCO₃ or alternative from reacting with the formaldehyde during the decontamination. The aluminum foil can be placed to allow the escape of ammonia gas when heated, or provision can be made to remove the aluminum foil remotely at the start of the neutralization phase. The removal technique must not allow unsafe levels of formaldehyde to escape the cabinet.

– Example 2: The cabinet is sealed using plastic with gloves as an integral part of the sheet of plastic. The NH₄HCO₃ or equivalent alternative is placed in a sealed container inside the cabinet. At the neutralization phase, the person performing the decontamination reaches into the cabinet without breaking the seal by using the gloves. The NH₄HCO₃ or equivalent alternative is removed from the sealed container and spread evenly over the heating surface of the heating device. The heating device is energized and the NH₄HCO₃ or equivalent alternative is heated and releases ammonia.

h) Place a hot plate, a beaker of water, and temperature and humidity indicators on the cabinet work tray. Do not connect electrical cords to the internal cabinet electric supply.

i) Close the opening to the work area with heavy gauge plastic film and tape. Close all possible leak areas, such as the exit of electrical cords, around the window and the junction of the plastic film and cabinet.

j) Determine the temperature and humidity inside the cabinet.

k) The temperature should be 70 °F (21 °C) or higher, and humidity should be 60 to 85%. Use the hot plate to heat the beaker of water until the desired temperature and humidity are achieved.

l) Prior to depolymerizing the formaldehyde, access to the area or room around the cabinet must be restricted in accordance with applicable federal and state regulation and prudent safety practice. OSHA’s Standard on Occupational Exposure to Formaldehyde requires that areas where the airborne concentration of formaldehyde exceeds the Permissible Exposure Limits be established as a regulated area with signs and labels marking the area and access restricted to properly trained personnel. Applicable regulations must be reviewed and complied with.

m) Plug the cord of the heating device into an outlet not installed on the cabinet.

n) After 25% of the paraformaldehyde has depolymerized, turn on the cabinet blower(s) for 10 to 15 s. Repeat after 50%, 75%, and 100% of the paraformaldehyde has depolymerized. In cases where the cabinet blower is inoperative, circulation of air within the cabinet should be promoted with additional blowers or fans, or the time of decontamination should be extended beyond the times suggested in p) below.

o) Disconnect the hot plate and heating device used for the paraformaldehyde from the electrical outlets.

p) Allow the cabinet to stand for a minimum of 6 h, preferably overnight (12 h).\textsuperscript{46}

q) Prepare the neutralizing agent as previously established in step g) and energize the heating device containing the NH\textsubscript{4}HCO\textsubscript{3} and the cabinet blower until the NH\textsubscript{4}HCO\textsubscript{3} has dissipated. As with the paraformaldehyde, after 25% of the NH\textsubscript{4}HCO\textsubscript{3} has depolymerized, turn on the cabinet blower(s) for 10 to 15 s. In cases where the cabinet blower is inoperative, circulation of air within the cabinet should be promoted with additional blowers or fans or the time of neutralization should be extended to a minimum of 6 h.

r) Let the cabinet stand for at least 1 h before opening seals.

s) If a flexible hose has been provided for the evacuation of the neutralized formaldehyde, slit the plastic covering the exhaust opening of the cabinet and seal the flexible hose to the opening. If the hose is working properly, the plastic covering the front opening of the cabinet should be sucked in. One or two small openings (approximately 6 x 6 in [15 x 15 cm]) are cut into the plastic covering the front opening of the cabinet to allow fresh air to enter the cabinet while the neutralized formaldehyde is being drawn out of the hose at the exhaust opening of the cabinet.

NOTE – Alternate removal procedures are acceptable if they allow for safe and effective removal of the formaldehyde gas.

G.7.3.2 Chlorine Dioxide (CD)

G.7.3.2.1 Method 1 – Fixed amount of CD

a) Calculate the total volume (in ft\textsuperscript{3} or m\textsuperscript{3}) of the cabinet by multiplying the height, width, and depth.

b) Calculate the amount of CD-generating chemical required for the decontamination. Multiply the total volume of the cabinet by 0.13 g/ft\textsuperscript{3} (4.7 g/m\textsuperscript{3}) to determine the mass of CD required to be generated. Multiply this value by the value of mass of CD per unit mass of generating chemicals, as given by the supplier of the generating chemicals.

c) If the cabinet is equipped with an external duct, fully close the exhaust decontamination damper, while leaving balancing, backdraft, EVAV or other dampers in their original position. This duct and the exhaust decontamination damper must be of a "gas tight" design. Sealing may also be accomplished at the terminal end of the duct. If the exhaust duct is more than 10 ft (3 m) long, additional CD-generating chemical may be needed to compensate for the increased volume. If in the unlikely event the cabinet exhausts into a recirculating building exhaust system or does not have a fully functioning gas-tight decontamination damper, disconnect the cabinet from the building system and form a gas tight seal (plastic film and tape may be used).

d) If the cabinet exhaust air is discharged into the room, tape a plastic cover over and completely seal the exhaust HEPA filter or port.

e) Place the chlorine dioxide generator within the BSC, or attach the external CD gas generator delivery system to the BSC. In either case, a means of recirculation to ensure adequate distribution of CD and humidity within the BSC, including above the exhaust filter, will be provided. (The recirculation loop may include the CD generator within the loop.) The inlet tube will preferably be connected into or beneath the workspace and the return tube shall be connected to a location above the exhaust HEPA filter.

f) Provide a means, either within or external to the BSC, by which the air within the BSC may be humidified and the relative humidity (RH) monitored and maintained within a range of 60 - 85% RH throughout the decontamination process. A hot plate, beaker of water, and temperature and humidity indicators on the cabinet work tray may be used. If using a hot plate within the cabinet, do not connect its electrical cords to the internal cabinet electric supply, as these devices do not generally provide adequate current.

g) Either provide a means, within or external to the BSC, by which the CD gas within the cabinet may be subsequently removed. Such a system might involve either the use of activated carbon granules or pellets or a chemical scrubbing system, through which the air within the cabinet can be circulated.

h) Close the opening to the work area with heavy gauge plastic film and tape. Seal all possible leak areas, such as the exit of electrical cords, around inlet and outlet hoses for the CD gas and/or its recirculation, around the window, and at the junction of the plastic film and cabinet.

i) Determine the temperature and humidity inside the cabinet.

j) The temperature should be 60 °F (15 °C) or higher, and the humidity should be 60 – 85 %RH. Use the hot plate with beaker of water or other means of humidity generation until the desired humidity level is attained. The cabinet blower and/or recirculation blower shall be operating during the entire humidification process.

k) Prior to the generation of CD gas, access to the area or room around the cabinet should be restricted in accordance with applicable federal and state regulation and prudent safety practice. It is recommended that a regulated area of radius of 20 ft be established about the cabinet to be decontaminated with CD, to be so indicated with signs and labels marking the area and access restricted to properly trained personnel. It is recommended that the room or area surrounding the cabinet be under negative relative pressure to prevent gas drifting in the event of leakage.

l) Begin generation and injection of CD gas into cabinet. Use the amount of CD – generating chemical as determined in step (b) above.

m) The cabinet blower (if available) and CD recirculation blower shall be operating during the entire CD gas generation period. Following the completion of CD gas generation, the cabinet blower and/or CD recirculation blower should be energized for at least 1 min during every 15 min of contact time.

n) Allow the cabinet to stand a minimum of 85 min from the initiation of CD gas generation with the assumption that the duration until peak concentration will be under 10 minutes.

o) Activate the system (scrubber) for removal of CD gas from the cabinet. Have the cabinet blower (if available) and CD recirculation blower energized during this period.

p) Allow sufficient time for the CD level within the cabinet to decrease to its STEL, the Short-Term Permissible Exposure Limit (0.3 ppm). This time depends upon the scrubbing system, but will generally require at least 30 min.

G.7.3.2.2 Method 2 – Fixed concentration of CD

a) If the cabinet is equipped with an external duct, fully close the exhaust decontamination damper, while leaving balancing, backdraft, EVAV, or other dampers in their original position. This duct and the exhaust decontamination damper must be of a “gas tight” design. Sealing may also be accomplished at the terminal end of the duct. If the exhaust duct is more than 10 ft (3 m) long, additional CD-generating chemical may be needed to compensate for the increased
volume. If in the unlikely event the cabinet exhausts into a recirculating building exhaust system or does not have a fully functioning gas-tight decontamination damper, disconnect the cabinet from the building system and form a gas tight seal (plastic film and tape may be used).

b) If the cabinet exhaust air is discharged into the room, tape a plastic cover over and completely seal the exhaust HEPA filter or port.

c) Place the chlorine dioxide generator within the BSC, or attach the external chlorine dioxide gas (CD) generator delivery system to the BSC. For an external generator, the inlet and outlet tubes/hoses to the BSC, may be connected to or beneath the workspace. For all B-type cabinets and for A-type cabinets with an inoperable internal blower, a means of recirculation to ensure adequate distribution of CD and relative humidity within the BSC, including above the exhaust filter, will be provided. (The recirculation loop may include the CD generator within the loop.) The inlet tube will preferably be connected into or beneath the workspace and the return tube will preferably be connected to a location above the exhaust HEPA filter.

d) Provide a means, either within or external to the BSC, by which the air within the BSC may be humidified and the relative humidity (RH) monitored and maintained within a range of 60 - 85% RH throughout the decontamination process. A hot plate, beaker of water, and temperature and humidity indicators on the cabinet work tray may be used. If using a hot plate within the cabinet, do not connect its electrical cords to the internal cabinet electric supply, as these devices do not generally provide adequate current.

e) Either provide a means, within or external to the BSC, by which the CD gas within the cabinet may be subsequently removed. Such a system might involve either the use of activated carbon granules or pellets or a chemical scrubbing system, through which the air within the cabinet can be circulated.

f) Provide a means to monitor the concentration of CD gas during the decontamination. Gas sampling is to be extracted from within the BSC at a distance of at least 1 ft from the CD gas inlet.

g) Close the opening to the work area with heavy gauge plastic film and tape. Seal all possible leak areas, such as the exit of electrical cords, around inlet and outlet hoses for the CD gas and/or its recirculation, around the window, and at the junction of the plastic film and cabinet.

h) Determine the temperature and humidity inside the cabinet.

i) The temperature should be 60 °F (15 °C) or higher, and the humidity should be 60 – 75 %RH. Use the hot plate with beaker of water or other means of humidity generation until the desired humidity level is attained. The cabinet blower and/or recirculation blower shall be operating during the entire humidification process.

j) Prior to the generation of CD gas, access to the area or room around the cabinet should be restricted in accordance with applicable federal and state regulation and prudent safety practice. It is recommended that a regulated area of radius of 20 ft be established about the cabinet to be decontaminated with CD, to be so indicated with signs and labels marking the area and access restricted to properly trained personnel. It is recommended that the room or area surrounding the cabinet be under negative relative pressure to prevent gas drifting in the event of leakage.

k) Begin generation and injection of CD gas into cabinet. Monitoring the CD concentration within the cabinet, cease generation when the concentration has at least achieved the targeted CD concentration (3.0 or 5.0 mg/L.).

l) The cabinet blower (if available) and CD recirculation blower (if present) shall be operating during the entire CD gas generation period. Following the completion of CD gas generation, the
cabinet blower and/or CD recirculation blower should be energized for at least 1 min during every 15 min of contact time.

m) Continuously monitor the CD gas concentration during decontamination. Whenever the CD concentration decreases below the targeted concentration level, (3.0 or 5.0 mg/L) generate and inject more CD gas until the CD concentration has at least attained the targeted concentration level.

n) Continue the decontamination for a duration of 60 min for a targeted concentration of 3.0 mg/L or 45 min for a targeted concentration of 5.0 mg/L, measured from the time that the targeted concentration was first achieved.

o) Activate the system (scrubber) for removal of CD gas from the cabinet. Have the cabinet blower (if available) and CD recirculation blower energized during this period.

p) Allow sufficient time for the CD level within the cabinet to decrease to its STEL, the Short-Term Permissible Exposure Limit (0.3 ppm). This time depends upon the scrubbing system, but will generally require at least 30 min.

G.8 Recommended HEPA Filter Disposal Procedures

G.8.1 HEPA filters that have been decontaminated are often burned in an incinerator. This disposal method is also effective for HEPA filters containing toxic chemicals. Factors to be considered when incinerating filters include, but are not limited to, composition of the waste to be burned, feed rate, combustion temperature and dwell time in the primary chamber.

G.8.2 HEPA filters may be placed in heavy plastic bags, such as those used to bag-out filters from contaminated filter housings. The bagged filters can be chemically decontaminated in situ by cutting small holes in the bag and delivering disinfectant by inserting a garden-type spray through the hole and spraying the filter media. The holes can be sealed with duct tape and shipped to an incinerator or sanitary landfill. This chemical method may be appropriate for filters containing agents (i.e. toxic chemicals or prions) that cannot be inactivated by the usual space decontamination procedures.

G.8.3 Decontaminated HEPA filters may be safety buried in a sanitary landfill because they no longer pose a hazard.

G.9 Lifespan of BSCs

The current lifespan of a Biosafety Cabinet is approximately 15 years. Use of modern day Biosafety cabinets (BSCs) began in the early 1970’s with BSCs that were manufactured to the NIH-03-112C Standard and subsequently the NSF Standard 49. BSCs manufactured in the 70’s, 80’s and early 90’s have provided over 15 years of service. Several considerations should be made of BSCs in this age group.

− Will the BSC need extensive service? (i.e. HEPA filter replacement, blower/motor replacement, will the electrical wire harnesses need replacement? etc.).

− Can an older BSC be commissioned after it has been in storage or purchased as a resale?

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47 WHEN SHOULD WE REPLACE OUR BIOLOGICAL SAFETY CABINETS, NuAire Technical Publication, GTB0155 REV 2 5/06 NuAire Corporation, Plymouth, MN
Will original test reports be available or will the BSC be commissioned to current NSF Standards?

After 15 years, replacement parts may or may not be available due to electrical or mechanical changes at the factory or industrial part suppliers. For example, magnetic ballasts and T12 fluorescent bulbs will not be available after the year 2010.

In addition, today’s BSCs have evolved through the years with many improvements in containment, ergonomics, serviceability, and energy efficiency that should be considered in a repair versus replacement decision.

G.10 Decommissioning process

G.10.1 No biosafety cabinet shall be sent to a landfill as a BSC, it shall be disassembled per requirements contained in this section.

G.10.2 Decontamination and PPE

G.10.2.1 After a review of the BSC hazard use, the cabinet may be considered chemically contaminated and requiring special decontamination procedures, not the standard gaseous sterilization. Follow paragraph G.10.2.3.

G.10.2.2 All decommissioned BSCs used with pathogens shall be gas sterilized.

G.10.2.3 BSCs to be decommissioned that were used with chemical agents shall have a hazard review made to determine whether special decontamination practices and PPE should be followed.

G.10.2.4 For those BSCs used with biological agents that may not be inactivated via formaldehyde or chlorine dioxide, the filters shall be incinerated and 10% bleach applied to all remaining contaminated surfaces. Obtain prior approval of the Facility Safety Officer.

G.10.2.5 PPE shall be used as directed by the Facility Safety Officer or the biosafety safety officer at CDC.

G.10.3 Metal Parts

G.10.3.1 All metal parts of less than 30 pounds (13 Kg) per item shall be removed from the lab and taken to an appropriate metal recycling container.

G.10.3.2 Metal parts in excess of 30 pounds (13 Kg), including the unit chassis, shall be taken to a designated area in the facility to be picked up by a commercial recycling vendor.

G.10.4 Glass Windows

All glass safety windows shall be taken to the designated glass container. Remove all metal parts that are not press fit to the glass edges.

G.10.5 Wiring

All accessible wiring shall be taken to a wiring recycling container.

G.10.6 Electrical Ballasts

All lamp ballasts shall be taken to the ballast collection center in the building.
G.10.7 Lamps

G.10.7.1 All fluorescent lamps shall be taken to the lamp container area in the building.

G.10.7.2 All ultraviolet lamps shall be handled as mercury-containing waste.

G.10.8 Labels

All warning labels, manufacturer's identification labels, certification labels shall be removed and destroyed.

G.10.9 Used HEPA filters

See G.8 Recommended HEPA Filter Disposal Procedures.
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Annex H

(informative)

Recommended materials, finishes, and construction

H.1 Sheet metal and finishes

H.1.1 All cabinet interior work surfaces, including the drain pan assembly, should be fabricated with corrosion-resistant steel conforming to Federal Specification QQ-S-766 (Class 304, Number 3 Finish).

H.1.2 If carbon steel sheet is used in cabinet fabrication, it should be prime grade, stretcher, or roller leveled, conforming to Federal Specification QQ-S-698 (Cold Rolled Sheets, Condition Number 3 Regular Finish).

H.1.3 Before painting, carbon steel surfaces should be free of dirt, oil, and grease. The carbon steel should be given a phosphate coating treatment in accordance with Federal Specifications TT-C-490. Prime and finish coats can be applied by spraying or dipping and should be baked after each coat for a minimum of 15 min at 300 °F (148.9 °C). The finish should be uniform, with a minimum thickness of 1 mm. Concealed surfaces or hollow metal sections should be protected by the finish, applied by a suitable method after welding and before assembly. Epoxy coatings may be used to coat all carbon steel surfaces and should conform to Federal Specification TT-C-001224. The finish should be uniform. Polyurethane coating may be used to coat all carbon steel surfaces and should conform to Federal Specification TT-C-001227. The finish should be uniform.

H.2 Glass

H.2.1 If safety glass is used for the window, it should be nominally 0.25 in (6.3 mm) laminated safety plate glass.

H.2.2 If tempered glass is used for the window, it should be nominally 0.25 in (6.3 mm) tempered glass conforming to American Society for Testing and Materials C 1048 or equivalent.

H.3 HEPA filter gasket materials

HEPA filter gasket materials should be cellular sheet or molded rubber or closed cell expanded neoprene gasket materials. Unless otherwise specified, the gasket should be fastened to the influent face of the filter frame. The gasket should be 0.25 ± 0.031 in (6.3 ± 0.8 mm) thick by 0.75 ± 0.031 in (19 ± 0.8 mm) wide and flush with the outer edges of the frame. The gasket should be either molded in continuous, unbroken form, or made from four strips joined at the corners by interlocking means, so that no gaps are visible, and the joint should be airtight. The gasket should be continuously cemented to the face of the filter frame to prevent any air leakage between the gasket and frame.49

48 The information contained in this Annex is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI's requirements for an ANS. Therefore, this Annex may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.

H.4 HEPA filter case – Type IC

HEPA Filter Case – Type IC (wood type fire retardant treated particle board) is acceptable for the case of the HEPA filter. Military Specification MIL-F-51068<sup>50</sup> lists other acceptable materials.

H.5 Specifications

The specifications require filter-mounting tolerances for openings up to 20 in (51 cm), ± 0.063 in (± 1.6 mm); and openings over 20 in (51 cm), to be ± 0.13 in (± 3.2 mm). The squareness of filter mountings should have diagonals within 0.063 in (1.6 mm) total allowance. Flatness at the filter gasket seal surface should be ± 0.015 in (± 0.4 mm) within any 10 in (25 cm) run.51

H.6 Sealants

H.6.1 Two part accelerated synthetic rubber (polysulfide type), temperature resistance, high adhesion aircraft specification grade, SAE AMS-S-8802, or equivalent, is acceptable.

H.6.2 One part silicon base sealant compound, such as Dow Corning RTV 732 Adhesive Sealant, Dow Corning RTV 781 Building Sealant, Dow Corning RTV 734 or RTV 112 Self-leveling Sealants, <sup>52</sup> or equivalent, is acceptable when used in accordance with the manufacturer's recommendations.

H.7 Fans

Fan(s) should be direct connected, centrifugal fans conforming to Air Movement and Control Association (AMCA)<sup>53</sup> standards. The performance curve for the specific fan furnished should be provided with each cabinet. Curves should display cubic feet/minute (cubic meters/second) vs. static pressure and voltage (and/or frequency) vs. cubic feet/minute (cubic meters/second).

H.8 Components and wiring

All electrical components and wiring should conform to the latest edition of the National Electrical Code, National Electrical Manufacturer's Association (NEMA)<sup>54</sup>, or Underwriters Laboratories (UL), whichever is applicable and provides the highest standard.


<sup>51</sup> Specifications taken from Military specification MIL-F-51068 (cancelled) www.defenselink.mil/pubs/

<sup>52</sup> The Dow Chemical Company, 2030 Dow Center, Midland, MI 48642 www.dow.com

<sup>53</sup> Air Movement and Control Association (AMCA), 30 West University Dr., Arlington Heights, IL 60004 www.amca.org

<sup>54</sup> NEMA, 1300 North 17<sup>th</sup> St., Suite 1847, Rosslyn, VA 22209 www.nema.org
Annex I

Reference standards and specifications pertinent to Class II biosafety cabinetry

I.1 Miscellaneous publications

I.1.1 Air Moving and Conditioning Association (AMCA)
- AMCA 99 – Standards Handbook
- AMCA 210-67 – Test Code for Air Moving Devices
- AMCA AS 2406 – Fans, Designation of Direction of Rotation and Discharge
- AMCA 211 – Fans, Labeling Requirements

I.1.2 American National Standards Institute, Inc. (ANSI)
- S1.4 – 1984 – Specification for Acoustical Calibrators
- Z97.1 – 1984 – Performance Specifications and Methods of Test for Safety Glazing Material Used in Buildings

I.1.3 Illuminating Engineering Society (IES)
- IES Lighting Handbook

I.1.4 National Electrical Code

I.1.5 National Electrical Manufacturers’ Association (NEMA)

I.1.6 Underwriters Laboratories, Inc.
- UL-62-1965 – Flexible Cord and Fixture Wire
- UL-94-1985 – Test for Flammability of Plastic Materials for Parts in Devices and Appliances
- UL-181 – Factory-Made Air Duct Materials and Air Duct Connectors
- UL-586-1985 – Test Performance of High Efficiency Particulate Air Filter Units

The information contained in this Annex is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI’s requirements for an ANS. Therefore, this Annex may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.

Latest edition in effect at the time of manufacture.
11.1.7 U. S. Department of Energy
   – ERDA 76-11 – Nuclear Air Cleaning Handbook (March 1976)

1.1.8 U. S. Department of Labor
   – Occupational Safety and Health Administration (OSHA) Safety and Health Standards for Respiratory Protection – 29CFR* 1910.134

1.1.9 U. S. Department of Health and Human Services
   – Centers for Disease Control, National Institute of Occupational Safety and Health, Requirements for Respirator, 30CFR* Part II
   *Code of Federal Regulations

1.1.10 American Conference of Governmental Industrial Hygienists

1.1.11 U. S. Naval Research Laboratory
   – Report 5959 (July 1963)

1.1.12 American Society for Testing and Materials (ASTM)
   – C 1048 – Specification for Heat Treated Flat Glass, Kind HS, Kind FT Coated and Uncoated Glass

I.2 Federal specifications
   – J#C-145 – Cable, Power, Electrical and Wire, Electrical; (Weather Resistant)
   – W-C-00596 – Connector, Plug, Electrical; Connector Receptacle, Electrical
   – W-S-00896 – Switch, Toggle
   – W-S-893 – Switch, Toggle, and Mounting Strap (Interchangeable)
   – CC-M-636 – Motor, Alternating-Current (Fractional Horsepower)
   – QQ-S-698 – Steel, Sheet and Strip, Low-Carbon
   – QQ-S-776 – Steel Plates, Sheets, and Strip-Corrosion Resisting
   – TT-C-490 – Cleaning Methods and Pretreatment of Ferrous Surfaces for Organic Coatings
   – TT-C-535 – Coating, Epoxy, Two-Component, for Interior and Exterior Use of Metal, Concrete and Masonry
– TT-C-001224 – Coating System, Epoxy, Glaze for Interior Surfaces
– TT-C-001227 – Coating System, Polyurethane Glaze for Interior Surfaces
– PPP-B-601 – Boxes, Wood, Cleated-Plywood
– PPP-B-621 – Boxes, Wood, Nailed and Lock-Corner
– PPP-B-640 – Boxes, Fiberboard, Corrugated, Triple-Wall
– PPP-C-650 – Crates, Wood, Open and Covered
– PPP-C-843 – Cushioning Material, Cellulosic
– PPP-T-60 – Tape, Packaging, Waterproof

I.3 Federal standards

– Federal Standard No. 102 – Preservation, Packaging and Packing Levels
– Federal Standard No. 123 – Marking for Domestic Shipment

I.4 Military specifications

– MIL-C-104 – Motor, Alternating Current (Fractional Horsepower)
– MIL-C-132 – Crates, Wood, Open; Maximum Capacity 2,500 pounds
– MIL-C-3774 – Crates Wood, Open; 12,000 and 16,000 Pound Capacity
– MIL-L-10547 – Liners, Case and Sheet Overwrap, Water-Vaporproof or Waterproof, Flexible
– MIL-P-116 – Preservation, Methods of
– MIL-R-3065 – Rubber, Fabricated Products-Gaskets, Synthetic Rubber
– MIL-S-8802 – Sealing Compound, Temperature-Resistant Aircraft High Adhesion
– MIL-F-51079B – Filters, Particulate, High Efficiency, Fire Resistant, Biological Use
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Annex J  
(informative)

J.1  Helium leak test

J.1.1  Purpose

This test on all biologically contaminated air plenums under positive pressure to the room determines whether exterior joints made by welding, gasketing, or sealing with sealants are free of leaks that might release potentially hazardous materials into the atmosphere.

J.1.2  Apparatus

The helium leak detector shall be calibrated in accordance with the manufacturer's instructions using a calibrated leak standard.

J.1.3  Method

a)  The room where testing will be performed shall be free of test gases, and air movements shall be kept to a minimum. Where levels are detected, they shall be below the acceptable leak rate for the test or, alternatively, corrected for by the leak detector instrument. No smoking should take place in the test area.

b)  Prepare the cabinet as a sealed system (see Annex A, section A.1.1).

c)  Pressurize the cabinet with air to 2 in w.g. (500 Pa). If the cabinet holds this pressure without more than ± 10% loss for 30 min, release pressure. If the cabinet does not hold this pressure, examine for gross leaks with liquid leak detector (see Annex A, section A.1.1), repair, and retest.

d)  Helium leak: Flow pure helium through the cabinet until the well-mixed helium concentration at the exhaust point reads 15% helium, and then pressurize the cabinet to 2 in w.g. (500 Pa). Alternatively, use an inflated bladder inside the cabinet to displace 15% of the internal gas volume and inject helium into the cabinet volume while venting the bladder outside the cabinet volume. Then pressurize to 2 in w.g. (500 Pa).

e)  Turn on the cabinet blower for 30 s to circulate gas.

f)  Adjust the helium leak detector to a sensitivity setting of $1 \times 10^{-5}$ cc/s, in accordance with the manufacturer's instructions.

g)  Move the detector probe over seams, joints, utility penetrations, panel gaskets, and other areas of possible leakage. Hold the detector probe at the surface of cabinet, being careful not to jar the instrument. Move the detector probe over the surface at a rate of approximately 1.0 in/s (2.5 cm/s), keeping the probe 0.25 to 0.50 in (6.3 to 13 mm) away from the surface (see Annex A, figure A1b).

57 The information contained in this Annex is not part of this American National Standard (ANS) and has not been processed in accordance with ANSI's requirements for an ANS. Therefore, this Annex may contain material that has not been subjected to public review or a consensus process. In addition, it does not contain requirements necessary for conformance to the Standard.
J.1.4 Acceptance

Measured leakage from any point in the cabinet shall not exceed a leak rate of \(1 \times 10^{-5}\) cc/s when pressurized to 2 in w.g. (500 Pa) with at least 15% concentration of helium.

J.2 Sulfur hexafluoride (SF\(_6\)) leak test

J.2.1 Purpose

This test on all biologically contaminated air plenums under positive pressure to the room determines whether exterior joints made by welding, gasketing, or sealing with sealants are free of leaks that might release potentially hazardous materials into the atmosphere.

J.2.2 Apparatus

- an industrial-type SF\(_6\) leak detector (Ion Track Inc. [ITI] Leakmeter, or equivalent capable of detecting a halide leak of \(1 \times 10^{-7}\) cc/s); and

- the SF\(_6\) leak detector (shall be calibrated in accordance with the manufacturer’s instructions using a calibrated leak standard).

J.2.3 Method

a) The room where testing will be performed shall be free of test gases, and air movements shall be kept to a minimum. Where levels are detected, they shall be below the acceptable leak rate for the test or, alternatively, corrected for by the leak detector instrument. No smoking should take place in the test area.

b) Prepare the cabinet as a sealed system (see Annex A, section A.1.1).

c) Pressurize the cabinet with air to 2 in w.g. (500 Pa). If the cabinet holds this pressure without more than \(\pm 10\%\) loss for 30 min, release pressure. If the cabinet does not hold this pressure, examine for gross leaks with liquid leak detector (see Annex A, section A.1.1), repair, and retest.

d) Pressurize the air filled cabinet at atmospheric pressure to 2 in w.g. (500 Pa) with SF\(_6\) gas.

e) Turn on the cabinet blower for 30 s to circulate gas.

f) Adjust the SF\(_6\) leak detector to a sensitivity setting of \(5 \times 10^{-7}\) cc/s, in accordance with the manufacturer’s instructions.

  g) Move the detector probe over seams, joints, utility penetrations, panel gaskets, and other areas of possible leakage. Hold the detector probe at the surface of cabinet, being careful not to jar the instrument. Move the detector probe over the surface at a rate of approximately 1 in/s (2.5 cm/s), keeping the probe 0.25 to 0.50 in (6.3 to 13 mm) away from the surface (Annex A, figure A1b).

J.2.4 Acceptance

Measured leakage from any point in the cabinet shall not exceed a leak rate of \(5 \times 10^{-7}\) cc/s to compensate for the dilution of halide gas.
Standards and Criteria

The following standards and criteria established and adopted by NSF as minimum voluntary consensus standards are used internationally:

2 Food equipment
3 Commercial warewashing equipment
4 Commercial cooking, rethermalization, and powered hot food holding and transport equipment
5 Water heaters, hot water supply boilers, and heat recovery equipment
6 Dispensing freezers
7 Commercial refrigerators and freezers
8 Commercial powered food preparation equipment
12 Automatic ice making equipment
13 Refuse processors and processing systems
14 Plastics piping system components and related materials
18 Manual food and beverage dispensing equipment
20 Commercial bulk milk dispensing equipment
21 Thermoplastic refuse containers
24 Plumbing system components for recreational vehicles
25 Vending machines for food and beverages
29 Detergent and chemical feeders for commercial spray-type dishwashing machines
35 High pressure decorative laminates (HPDL) for surfacing food service equipment
36 Dinnerware
37 Air curtains for entranceways in food and food service establishments
40 Residential wastewater treatment systems
41 Non-liquid saturated treatment systems
42 Drinking water treatment units – Aesthetic effects
44 Residential cation exchange water softeners
46 Evaluation of components and devices used in wastewater treatment systems
49 Biosafety cabinetry: Design, construction, performance and field certification
50 Equipment for swimming pools, spas, hot tubs and other recreational water facilities
51 Food equipment materials
52 Supplemental flooring
53 Drinking water treatment units – Health effects
55 Ultraviolet microbiological water treatment systems
58 Reverse osmosis drinking water treatment systems
59 Mobile food carts
60 Drinking water treatment chemicals – Health effects
61 Drinking water system components – Health effects
62 Drinking water distillation systems
140 Sustainable carpet assessment
143 Environmentally preferable products – Hard surface cleaners
169 Special purpose food equipment and devices
170 Glossary of food equipment terminology
173 Dietary supplements
177 Shower filtration systems – Aesthetic effects
184 Residential dishwashers
222 Ozone generators
245 Wastewater treatment systems – Nitrogen reduction
14159-1 Hygiene requirements for the design of meat and poultry processing equipment
14159-2 Hygiene requirements for the design of hand held tools used in meat and poultry processing equipment
14159-3 Hygiene requirements for the design of mechanical belt conveyors used in meat and poultry processing equipment

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THE HOPE OF MANKIND rests in the ability of man to define and seek out the environment which will permit him to live with fellow creatures of the earth, in health, in peace, and in mutual respect.