BSR/ASHRAE Standard 185.4P

Public Review Draft

Method of Testing In-Room Ultraviolet Devices and Systems for Microbial Inactivation on Surfaces in a Test Room

First Public Review (February 2024)
(Complete Draft for Full Review)

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FOREWORD

Standards Project Committee (SPC) 185.4 was organized in 2022 to develop a method of test to determine inactivation efficacy of microorganisms on surfaces exposed to UV irradiance.

This is a test method standard, and its results are to be used to directly compare UVGI equipment on a standardized basis irrespective of their application. Results are also used to give the design engineer an easy-to-use basis for specifying UV devices or estimating the relative performance of UVGI equipment for a given application as it pertains to surface applications. It is entirely possible that an industry organization may use this test method as the basis for an application standard in which they might require testing at conditions different than those required in this standard.

This test method is to be performed by persons with training in microbiology in facilities designed and equipped for work with infectious agents at the appropriate biosafety level (Biosafety in Microbiological and Biomedical Laboratories, 6th ed). It is the responsibility of the investigator to determine whether Good Laboratory Practice Regulations (GLP) are required and to follow them when appropriate (40 CFR, Part 160 for EPA submissions and 21 CFR, Part 58 for FDA submissions).

This standard does not purport to address all of the safety concerns associated with its use. It is the responsibility of the user of this standard to establish appropriate safety, health, and environmental practices and determine the applicability of regulatory limitations prior to use.

NOTE: Informative notes are used throughout this standard to provide non-mandatory guidance for the user in addition to the non-mandatory guidance found in informative appendices. Informative notes are for information only and are not part of the standard.

1. PURPOSE

This standard establishes a test method for evaluating the efficacy of germicidal ultraviolet systems for microbial inactivation on multiple surface locations in a test room.

2. SCOPE

2.1 The standard applies to ultraviolet devices and systems using only germicidal ultraviolet energy for inactivation.

2.2 The method of test specifies selected indicator microorganisms and defines procedures for inoculating test carriers in a room-scale test chamber.

2.3 The method of test defines the test carrier quantity and positions in the test room.

2.4 This standard provides a method for counting the number of viable microorganisms on the test carriers before and after ultraviolet inactivation.

2.5 This standard establishes protocols and minimum requirements for the materials and equipment required to conduct the tests, defines methods of calculating and reporting results obtained from the test data, and establishes a reporting system to be applied to in-room devices and systems covered herein.
2.6 This standard does not address the health and safety effects of operating devices and systems in an occupied room.

3. DEFINITIONS and ACRONYMS

3.1 Some terms are defined below for the purposes of this standard. When definitions are not provided, common usage shall apply.

**Burn-in time:** a period of time that UV lamps are powered on prior to putting the lamps into service, typically 100 hours.

**Carrier:** substrate on which test microbes are deposited.

**Germicidal:** capable of killing or inactivating microorganisms.

**Germicidal UV (GUV):** Ultraviolet (UV) radiation with a wavelength between 280 nm and 200 nm.

**Irradiance:** the power of electromagnetic radiation incident on a surface per unit surface area, typically reported in microwatts per square centimeter ($\mu$W/cm$^2$).

**Reflectivity:** the fraction of incident radiation reflected by a surface.

**Ultraviolet Germicidal Irradiation (UVGI):** the use of ultraviolet C (UV-C) energy, through a system designed to deliver UV-C, to kill or inactivate microorganisms.

**UV-C:** UV radiation with a wavelength between 280 nm and 100 nm.

**UV-C Device:** a complete assembly consisting of lamp(s), power supply(s), and supporting fixtures.

**UV-C System:** one or more UV-C Devices or fixtures utilized for this method of test.

**Wavelength:** the distance between repeating units of a wave pattern commonly measured in nanometers in the UV spectrum.

3.2 Acronyms

- **BSL** biosafety level
- **CFU** colony forming units
- **COR** center of room
- **FBS** fetal bovine serum
- **LR** log reduction
- **SDS** sodium dodecyl sulfate
- **TSB** trypticase soy broth
- **TSA** trypticase soy agar
- **UV** ultraviolet

4. TEST ROOM SET UP

4.1 Test Room

The test room shall comply with the requirements as specified in this standard.
4.1.1 A means of viewing the lamps to verify operation shall be included and shall ensure that exposure to UV-C radiation does not occur during viewing.

4.1.2 Temperature within the test room throughout the test shall be 21°C +/- 2.2°C (70°F +/- 4ºF), relative humidity shall be between 30% to 60%.

4.1.3 Test Room Dimensions and layout: see Figure 1 and 2
   • Minimum Dimensions (per side): 4.3m +/- 0.5m (14ft +/- 1.6ft)
   • Maximum Dimensions (per side): 6.1m +/- 0.5m (20ft +/- 1.6ft)
   • Minimum Ceiling Height: 2.4m +/- 0.3m (8ft +/- 1ft)
   • Maximum Ceiling Height: 3.7m +/- 0.3m (12ft +/- 1ft)

4.1.4 Test Room Surface Reflectivity
   4.1.4.1 Test-room shall have total ultraviolet (UV) reflectance of <10% at the main operational wavelength of the UV-C system under test.
   4.1.4.2 For common surface materials, peer reviewed data of total reflectivity may be used to meet this requirement. For unknown materials, wavelength reflectivity shall be validated by calibrated reflectivity measurements. The test report shall list the reflectance value at the main operational wavelength.
   Note: It shall be assumed that all metal surfaces have a reflectivity of >15% in the wavelength range of 200-405nm, unless the surface is coated with a low reflectivity material. It must be assumed that the reflectivity varies depending on the wavelength.

4.1.5 Test Carrier Placement:
   4.1.5.1 Floor Samples: Test carriers shall be placed in triplicate at locations 0.91m (3ft) and 2.1m (7ft) from the center of the room (COR). See Figure 3.
   4.1.5.2 Pole Samples: 0.91m (3ft) and 2.1m (7ft) from COR.
      • Poles with 2 cubes per pole
        • Low cube: The top horizontal surface of the lowest cube shall be at a height of 86 cm +/- 0.6 cm (34 in +/- 0.2 in)
        • High cube: The top horizontal surface of the highest cube shall be at a height of 15 cm +/- 0.6 cm (60 in +/- 0.2 in)
      • Orientation Defined: Refer to Section 4.1.6.1 for further details
   • Test carriers shall be placed in triplicate on each face of each cube.

4.1.6 Cube Description:
   Each cube consists of six equal square sides no less than 15cm (6in) and no more than 23cm (9in) in any dimension (length, width, or height). See Figures 4 A and B.

4.1.6.1 Each pole shall have mounted to it two cubes (High & Low) with triplicate sample sites to be placed on all six faces of each cube (Top, Bottom, Left, Right, Front, and Back). See Figure 5.

Each cube shall be positioned and oriented so that the top and bottom horizontal surfaces are parallel to the floor and ceiling. All four of the outer vertical surfaces (Front, Back, Left, and Right) shall be parallel to the two walls in the same planes and perpendicular to the other two walls of the room. The center points of the cubes shall be in line with the center of the mounting pole. See Figure 4. NOTE: Mounting of the cubes shall minimize UV blockage and reflectivity to the cube surfaces.

4.2 Installation of UV-C System
4.2.1 Installation and configuration of the UV-C system within the test room shall be as designated by the manufacturer or equipment provider.

4.2.2 The burn-in time for lamps shall be per manufacturer instructions. The ultraviolet source burn-in time/warm-up cycle shall follow manufacturer recommendations.
4.2.3 Measurement of UV radiation from UV-C system. This test is optional.

4.2.3.1 A NIST traceable UV radiometer capable of measuring wavelengths and irradiance, appropriate to the UV-C system being tested, shall be used.

4.2.3.2 Where applicable, the UV-C system shall be configured and orientated so that its maximum total radiation output is directed towards the radiometer.

5. Microbial Culture and Testing Procedure

Required organism for this test: *Bacillus spizizenii* (ATCC 6633). This strain is formerly known as *Bacillus subtilis* subsp. *spizizenii*. In addition, as desired by the manufacturer, other organisms may be chosen.

5.1 Microorganism Propagation

5.1.1 Initial Growth of *B. spizizenii* (ATCC 6633)

5.1.1.1 From Liquid Stock: Inoculate 10 mL sterile trypticase soy broth (TSB) with 10 µL of reference microorganism stock. From Agar Plate: Using a 10 µl sterile loop, pick colony and inoculate into TSB.

5.1.1.2 Vortex suspension and incubate dynamically at approximately 150 rpm at 35 ± 1 °C for 24 ± 4 hours.

5.1.2 Spore Propagation

5.1.2.1 Obtain the incubated culture from Section 5.1.1.2.

5.1.2.2 Spread plate 100 µL from the initial growth culture onto a sterile trypticase soy agar (TSA) plate. Repeat with 2 additional sterile TSA plates.

5.1.2.3 Incubate plates upright at room temperature until the sample has absorbed into the media.

5.1.2.4 Incubate the sterile TSA plates inverted at 35 ± 1 °C for 14 days.

5.1.3 Harvest and Purification of Spores

5.1.3.1 Use a sterile inoculation loop, rubber policeman, or cell scraper to scrape the *B. spizizenii* (ATCC 6633) lawn off of the TSA plates and suspend in 10 mL sterile, distilled water. Take care not to damage the agar.

5.1.3.2 Vortex agitate the cell suspension for 10 seconds.

5.1.3.3 At this point, the suspension may be stored at (0 to 6) °C for up to 72 hours.

5.1.3.4 Heat the cell suspension in a 70 ± 5 °C water bath for 1 hour ± 5 minutes.

5.1.3.5 Centrifuge the cell suspension 3,200 +/- 200 rcf for 10 minutes.

5.1.3.6 Aspirate and discard the supernatant.

5.1.3.7 Prepare 10 ml of filter sterilized lysozyme solution (60 µg/mL) in 10 mM tris-HCl Buffer, pH 7.4 for each suspension. This solution shall be prepared each day of testing.

5.1.3.8 Add 10 mL 60 µg/mL lysozyme to the cell pellet. Vortex agitate for 10 seconds.

5.1.3.9 Incubate the cells at 36 ± 1 °C for 1 hour ± 5 minutes.

5.1.3.10 Centrifuge the cell suspension at 3,200 +/- 200 rcf for 10 minutes.

5.1.3.11 Discard supernatant and resuspend the pellet in 10 mL sterile distilled water.

5.1.3.12 Centrifuge the cell suspension at 3,200 +/- 200 rcf for 10 minutes.

5.1.3.12.1 Prepare a 10 ml filter-sterilized sodium dodecyl sulfate (SDS) solution (0.05% weight/volume) in distilled water for each suspension.

5.1.3.13 Discard supernatant and resuspend the pellet in 10 mL of 0.05% SDS.

5.1.3.14 Vortex agitate the cell suspension for 10 seconds.

5.1.3.15 Centrifuge the cell suspension at 3,200 +/- 200 rcf for 10 minutes.

5.1.3.16 Discard supernatant and resuspend the pellet in 10 mL of sterile distilled water.
5.1.3.17 Repeat the previous two steps 1 additional time.
5.1.3.18 At this point, the suspension may be stored at (2 to 8) °C for up to 2 months.

5.1.4 Quality check on spore purity
5.1.4.1 Enumerate the number of spores and the number of vegetative cells with an appropriate method such as phase contrast microscopy or validated equivalent method, such as flow cytometry. For microscopy, examine at least five fields-of-view (FOV) having at least 20 microbes per FOV. The minimum spore fraction must be achieved in each FOV. Calculate the fraction of spores in the total cell count. The minimum spore fraction should be 0.8. Spore suspension from multiple plates can be combined for uniformity.

5.2 Inoculation
5.2.1 Carrier shall:
5.2.1.1 Be Stainless Steel grade 304 (ASTM A240),
5.2.1.2 Be passivated before and between uses (EPA Interim Method for Evaluating the Efficacy of Antimicrobial Surface Coatings),
5.2.1.3 Have a number 4 finish (ASTM A480),
5.2.1.4 Be ≤ 1 x 0.9 inch so that it can fit into a standard 50 mL conical centrifuge tube, and
5.2.1.5 Be large enough that it can accommodate ten discrete 1-µL droplets.

5.2.2 Prepare a spore suspension at a concentration of (1 to 5) x 10⁸ CFU/ml. in sterile distilled water with 5% fetal bovine serum (FBS).

5.2.3 Inoculate the carriers with ten, discrete, evenly spaced, 1-µL drops of the B. spizizenii stock prepared above at a concentration ≥ 1.00 x 10⁸ CFU/mL for a concentration of ≥ 1.00 x 10⁶ CFU/carrier at the conclusion of the experiment based on positive control coupon results.

5.2.4 Inoculate three carriers per location plus three additional coupons to serve as positive controls.

5.2.5 For negative controls, reserve three carriers prepared as above but inoculate with 1µl of 5% FBS as above in 5.2.3 instead of the B. spizizenii stock.

5.2.6 Allow carriers to dry at room temperature under still air until dry in a biological safety cabinet (15-30 minutes).

5.2.7 Store the dried carriers in an airtight container at (2 to 8) °C for up to be 7 days before use.

5.3 Sample Treatment
5.3.1 Test carriers shall be placed according to Section 4.1.5. Negative and positive control carriers shall be placed in the testing room in a light impermeable container during the exposure of test carriers to the UV-C source.

5.3.2 Exposure of test carriers to the UV-C system shall be determined by the manufacturer’s instructions for use.

5.4 Sample Recovery
5.4.1 Label 50 mL conical bottom centrifuge tubes with carrier identifier.
5.4.2 Add 10 mL sterile, distilled water to each tube.
5.4.3 Remove carrier from mount, place into tube, and cap.
5.4.4 Immediately before plating, vortex tubes for 2 min.
5.4.5 Pipette the desired volume for CFU assay.
5.5 Sample Processing
5.5.1 Quantify microbiological growth on test carriers, positive control, and negative control carriers using a viable culture method. If more than one nontarget CFU is present on the negative controls, consider the entire set of positive controls and test samples to be contaminated. Redo the method, ensuring the soil solution and media does not have microbial contamination.

5.6 Data Collection and Analysis
5.6.1 After incubation, count the number of colonies matching the *B. spizizenii* morphology (compare to inoculum and positive controls) on plates from a dilution that yields (20 to 200) CFU. Less than 20 CFU can be counted on a plate from an undiluted sample. Record plates with no growth as <1 CFU. Count any CFUs that do not match the target morphology but do not include in the *B. spizizenii* count.

5.6.2 A sample shall be excluded if there is any non-target growth detected that inhibits or suppresses growth of *B. spizizenii*. An excluded sample shall be reported, and the results from that location flagged as such.

5.6.3 For the positive controls, determine the mean log CFU (\(\mu_{\text{control}}\)) for each test location using the following equation, where \(c = \text{CFU count per replicate}\):

\[
\mu_{\text{control}} = \frac{\log_{10} c_1 + \log_{10} c_2 + \log_{10} c_3}{3}
\]

5.6.4 For the test samples, determine the mean log transformed CFU (\(\mu_{\text{treatment}}\)) for each test location using the following equation, where \(x = \text{CFU count per replicate}\):

\[
\mu_{\text{treatment}} = \frac{\log_{10} x_1 + \log_{10} x_2 + \log_{10} x_3}{3}
\]

5.6.5 Calculate the log reduction (LR) per location by the following equation:

\[
LR = \mu_{\text{control}} - \mu_{\text{treatment}}
\]

For reporting, LR is also calculated using the above equation with a single replicate.

5.6.5.1 Note on calculations: change the number of samples and the sample size denominator if a deviation from the prescribed three samples is made.

5.6.5.2 Note: The reduction can also be presented as a percent reduction by the following equation:

\[
\text{Percent Reduction (\%)} = (1 - 10^{-LR}) \times 100
\]

5.7 Quality Control
5.7.1 Conditions for a Valid Test
5.7.1.1 The negative control samples must have no more than one nontarget CFU and no *B. spizizenii* across all three samples.
5.7.1.2 The positive control samples must have a mean concentration (1 to 5) \times 10^6 \text{ CFU/coupon} \ B. \ spizizenii.
5.7.1.3 Minimum spore concentration shall be at minimum 80% spore and at maximum 20% vegetative cells as per Section 5.1.4.

5.8 Reporting Requirements
5.8.1 Report the log reduction and/or percent reduction, mean log concentration, and each value for each location and control group.
5.8.2 Report any deviations, qualifiers, or anomalous results with interpretive comments for each location.
5.8.3 Report spore fraction.
5.8.4 Report recovery and quantification method by reference to established protocol and include any deviations.

6. REPORTING RESULTS

6.1 The summary section of the method of test report shall include the following information:
   6.1.1 Name and location of the test laboratory
   6.1.2 Date of the test
   6.1.3 Test operator’s name(s)
   6.1.4 Name of the UV system manufacturer (or test requester, if different from the manufacturer)

6.2 Room details
   6.2.1 Room dimensions in L (m/ft) x W (m/ft) x H (m/ft)
   6.2.2 Description of materials used for walls and ceiling
   6.2.3 Reflectivity of test room
      List specific NIST traceable UV-radiometer information (brand, accuracy range, and last calibration record) for all measuring instruments and sensors used for determining room reflectivity measurement to validate <10% test-room reflectivity.

6.3 Description of the UV-C system:
   6.3.1 UV-C System brand name, model number and serial number, and any identifying marks
   6.3.3 Photos of the entire UV-C system being evaluated.
   6.3.4 Technical specification of the entire UV-C system being evaluated as provided by the UV-C system provider, which should include the following minimum specifications. If the UV-C system comprises separate multiple sources (or emitters), technical description of each source (or emitter) should be included.
      6.3.4.1 Physical dimensions of the UV-C system
      6.3.4.2 UV wavelength(s) being emitted
      6.3.4.3 Input power requirements (battery information, if robotic)
      6.3.4.4 Operating current and voltage

6.4 Test Set-Up
   6.4.1 Plan and elevation view drawings showing location and designation of test carriers. Drawing to include:
   a. Pole samples:

<table>
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<tr>
<th>Distance from Center of Room</th>
<th>Location on Pole</th>
<th>Locations on Cube</th>
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<tbody>
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<td>3 ft</td>
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<td>Top, Bottom, Front, Back, Left, Right</td>
</tr>
<tr>
<td></td>
<td>High Cube</td>
<td>Top, Bottom, Front, Back, Left, Right</td>
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<tr>
<td>7 ft</td>
<td>Low Cube</td>
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b. Floor Samples

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</tr>
<tr>
<td>7 ft</td>
<td>7-F</td>
</tr>
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6.5. UV-C system Operating information
Operation of UV-C System to be followed as per manufacturer’s operational protocol and safety protocol. The following details are to be reported:
6.5.1 Plan and elevation view drawings showing location of UV-C system (all sources (emitters)) in the test room. Drawing to include:
   a. Number of all sources (emitters) used in the test
   b. Exact location of all sources (emitters) used in the test
   c. Distance between UV-C system (all sources) and test carriers
6.5.2 Photos of UV-C system located in the test room, including all sources (emitters) as part of the UV-C system at the start of the test
6.5.3 Total cycle time for the test
6.5.4 Number of individual cycles used and corresponding cycle times at each manufacturer designated location of the UV-C system

6.6 Test Parameters
6.6.1 Test carriers: Material and dimensions
6.6.2 Microorganism, ATCC strain designation & lot number
6.6.3 Inoculum Count (CFU/carrier)
6.6.4 Inoculum Volume (ml):
6.6.5 Carrier Surface Area (cm²):
6.6.6 Environmental Conditions (Temperature (°C), Relative Humidity):
6.6.7 Soiling Type (FBS %): Spore recovery buffer
6.6.8 Enumeration Media, Method (TSA, pour?)
6.6.9 Incubation period (hrs) and conditions (Temperature (°C), aerobic or anaerobic?)

6.7 Test Results
6.7.1 For each carrier report: test location, treatment time, replicate number, carrier number, percent reduction, log reduction, and log CFU
6.7.2 For each test location report: mean log CFU, percent reduction, and mean log reduction. Refer to Section 5.5 for calculation of results.
7. NORMATIVE REFERENCES


Biosafety in Microbiological and Biomedical Laboratories, 2020. 6th edition, NIH/CDC


8. BIBLIOGRAPHY


British Standard BS 8628:2022: Disinfection using ultraviolet radiation — Methods for quantitative testing of automated ultraviolet disinfection activities by direct illumination — Determination of bactericidal, mycobactericidal, sporicidal, yeasticidal, fungicidal, virucidal and phagocidal activities. Published by BSI Standards Limited 2022


**Figure 1**
LEGEND

A. Ceiling minimum height is 8 ft (2.44 m).
B. Ceiling maximum height is 12 ft (3.6 m).
C. Floor minimum dimension is 14 ft (4.27 m).
D. Floor maximum dimension is 20 ft (6.1 m).
E. Cube Dimensions: 6 in to 9 in. (152.4 mm to 228.6 mm) cube.
F. Pole Distances: 3 ft. and ft (0.91 m and 2.13 m) from center of room.
G. Floor Sample Distances: 3 ft. and ft (.91 m and 2.13 m) from center of room.
H. Center of Room.
Figure 3

ROOM DIMENSIONS: Maximum of 20 ft. (6.1m) & Minimum of 14 ft. (4.27m)

- CENTER OF ROOM
- Floor Sample Site (3 ft./.91m from Center of Room)
- Floor Sample Site (7 ft./2.13m from Center of Room)
- Cubes (High & Low; 3 ft./.91m from Center of Room)
- Cubes (High & Low; 7 ft./2.13m from Center of Room)
Figure 4

A. CUBE VIEW
(From approximate center of room)

B. Top View
Figure 5

LEGEND

I. Pole max width is 3 in (76.2 mm).
J. Cube minimum is 6 in (152.4 mm).
K. Cube maximum is 9 in (228.6 mm).
L. Top height of cube 1 is 5.0 ft (1.53 m) from floor.
M. Top height of cube 2 is 2.835 ft (0.864 m) from floor.
INFORMATIVE APPENDICES

INFORMATIVE APPENDIX A

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TEST ORGANISM

This standard is based on one test organisms (refer to Section 5 Microbiology). Other organisms may also be chosen and used with the methodology of the test.

Microorganisms vary in both their sizes and shapes. Therefore, there is the need to select microorganisms that reflect that natural diversity. Additionally, because the efficacy of UV-C radiation to inactivate microorganisms is being evaluated, it is necessary to select microorganisms that range from readily inactivated to more difficult to inactivate. Generally, vegetative bacteria are readily inactivated and bacterial spores, more difficult (Figure A, Informative Appendix E. Inactivated is typically understood to mean that the microorganism does not have the ability to replicate. The susceptibility of a microorganism to UV-C radiation is defined as the k-value for a single-pass flow reactor and is expressed as cm²/µJ. The magnitude of the k-value defines the susceptibility – large k-values indicate low susceptibility to, and small k-values indicate high susceptibility to, inactivation by UVC radiation.

INFORMATIVE APPENDIX B

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LIMITATIONS

This method of test is limited to a test room and does not reflect real-world use.

INFORMATIVE APPENDIX C

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SAFETY

The primary safety concern associated with this method is exposing test personnel to microorganisms. All microorganisms may be considered opportunistic pathogens depending upon the susceptibility and immunological condition of an individual. There are four biosafety levels (BSLs) for activities with microorganisms based on the potential risk posed by the organism and the intended activities in the laboratory. The guidelines for the appropriate practices, safety equipment, and facilities are described in the Biosafety in Microbiological and Biomedical Laboratories (BMBL) manual (Biosafety in Microbiological and Biomedical Laboratories, 6th Ed, 2020). Prior to initiating any tests, a risk assessment should be done to establish the BSL needed. BSL-1 safety practices and controls are appropriate for microorganisms not normally associated with human disease. BSL-2 is recommended for organisms associated with human disease that pose a moderate hazard to workers. BSL-3 is applicable to indigenous/exotic agents associated with human disease and with the potential for aerosol transmission. BSL-4 is needed for dangerous/exotic agents of a life-threatening nature. BSL-2, BSL-3, and BSL-4 work requires specialized containment that is not detailed in this test method.

In addition to the biosafety issues, other safety issues associated with this method may require safe transport (including lifting), and use of various heavy and awkward equipment at times.

No personnel should be subject to direct UV exposure, but if exposure is unavoidable, personnel should wear protective clothing (no exposed skin), protective eyewear, and gloves. Most eyewear, including prescription glasses, are sufficient to protect eyes from UV, but not all offer complete coverage; standard issue protective goggles may be the best alternative.

INFORMATIVE APPENDIX D

(This appendix is not part of this standard. It is merely informative and does not contain requirements necessary for conformance to the standard. It has not been processed according to the ANSI requirements for a standard and may contain material that has not been subject to public review or a consensus process. Unresolved objectors on informative material are not offered the right to appeal at ASHRAE or ANSI.)

ENVIRONMENTAL CONSIDERATIONS

The method described herein is a laboratory procedure, and release of microorganisms into the environment should not be permitted. Engineering controls as described in the BMBL and when used on the test systems are designed to prevent such a release. However, a risk analysis should be done to assess the possibility of an accidental release. If this analysis indicates a high likelihood of an accidental release, then the effect that the microorganisms may have on the environment needs to be assessed. Many microorganisms only requiring BSL-1 containment for human health, while not infectious to humans, may be plant pathogens and may have a detrimental impact on the environment. The American Type Culture Collection (ATCC), the Centers for Disease Control (CDC), and the United States Department of Agriculture (USDA) have published information on plant pathogens. These issues should be in laboratory environmental safety plans.

INFORMATIVE APPENDIX E

(This appendix is not part of this standard. It is merely informative and does not contain requirements necessary for conformance to the standard. It has not been processed according to the ANSI requirements for a standard and may contain material that has not been subject to public review or a consensus process. Unresolved objectors on informative material are not offered the right to appeal at ASHRAE or ANSI.)

MICROORGANISM SUSCEPTIBILITY TO UV-C RADIATION
### Table 2 Representative Members of Organism Groups

<table>
<thead>
<tr>
<th>Organism Group</th>
<th>Member of Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vegetative Bacteria</td>
<td><em>Staphylococcus aureus</em></td>
</tr>
<tr>
<td></td>
<td><em>Streptococcus pyogenes</em></td>
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<tr>
<td></td>
<td><em>Escherichia coli</em></td>
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<tr>
<td></td>
<td><em>Pseudomonas aeruginosa</em></td>
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<tr>
<td></td>
<td><em>Serratia marcescens</em></td>
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<tr>
<td>Mycobacteria</td>
<td><em>Mycobacterium tuberculosis</em></td>
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<tr>
<td></td>
<td><em>Mycobacterium bovis</em></td>
</tr>
<tr>
<td></td>
<td><em>Mycobacterium leprae</em></td>
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<tr>
<td>Bacterial Spore</td>
<td><em>Bacillus anthracis</em></td>
</tr>
<tr>
<td></td>
<td><em>Bacillus cereus</em></td>
</tr>
<tr>
<td></td>
<td><em>Bacillus subtilis</em></td>
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<tr>
<td>Fungal Spores</td>
<td><em>Aspergillus versicolor</em></td>
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<tr>
<td></td>
<td><em>Penicillium chrysogenum</em></td>
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<tr>
<td></td>
<td><em>Stachybotrys chartarum</em></td>
</tr>
<tr>
<td>Viruses</td>
<td>Influenza viruses</td>
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<tr>
<td></td>
<td>Measles</td>
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<tr>
<td></td>
<td>SARS</td>
</tr>
<tr>
<td></td>
<td>Smallpox</td>
</tr>
<tr>
<td></td>
<td>Coronavirus</td>
</tr>
</tbody>
</table>