



Designation: D7855/D7855M – 13 (Reapproved 2021)

Standard Test Method for Determination of Mold Growth on Coated Building Products Designed for Interior Applications Using an Environmental Chamber and Indirect Inoculation¹

This standard is issued under the fixed designation D7855/D7855M; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ϵ) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This test method covers an environmental chamber and the conditions of operation to evaluate in a 4-week period the relative resistance to mold growth and microbial surface defacement on coated building products designed for interior application using an indirect inoculation method. The apparatus is designed so it can be easily built or obtained by any interested party.

1.2 This test method can be used to evaluate the comparative resistance of coated building products to accelerated mold growth. Ratings do not imply a specific time period that the coated building product will be free of fungal growth during installation in an interior environment.

1.3 This test method is not intended for use in the evaluation of public health claims.

1.4 The test method is intended for the accelerated evaluation of mold growth on a coated building product designed for interior use. This method is not intended for evaluation of surfaces designed for exterior applications or uncoated surfaces. Use of this test method for evaluating exterior performance has not been validated, nor have the limitations for such use been determined.

1.5 The values stated in either SI units or inch-pound units are to be regarded separately as standard. The values stated in each system may not be exact equivalents; therefore, each system shall be used independently of the other. Combining values from the two systems may result in non-conformance with the standard.

1.6 *This standard does not purport to address all of the safety concerns, if any, 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.*

¹ This test method is under the jurisdiction of ASTM Committee D01 on Paint and Related Coatings, Materials, and Applications and is the direct responsibility of Subcommittee D01.28 on Biodeterioration.

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1.7 This international standard was developed in accordance with internationally recognized principles on standardization established in the Decision on Principles for the Development of International Standards, Guides and Recommendations issued by the World Trade Organization Technical Barriers to Trade (TBT) Committee.

2. Referenced Documents

2.1 *ASTM Standards:*²

D16 Terminology for Paint, Related Coatings, Materials, and Applications

D1193 Specification for Reagent Water

D6329 Guide for Developing Methodology for Evaluating the Ability of Indoor Materials to Support Microbial Growth Using Static Environmental Chambers

E177 Practice for Use of the Terms Precision and Bias in ASTM Test Methods

E691 Practice for Conducting an Interlaboratory Study to Determine the Precision of a Test Method

3. Terminology

3.1 *Definitions*—For definitions of terms refer to Terminology D16.

3.2 *Definitions of Terms Specific to This Standard:*

3.2.1 *chamber control, n*—open Petri dish containing appropriate agar to demonstrate viability of fungal organisms within the environmental chamber.

3.2.2 *coated building product, n*—a building material having a liquid, liquefiable or mastic composition that is converted to a solid protective, decorative, or functional adherent film after application as a thin layer onto a building fabric.

3.2.3 *interior, n*—any surface not exposed to exterior environments in end use.

3.2.4 *interior finish, n*—interior wall and ceiling finish and interior floor finish.

² For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

3.2.5 *material control, n*—untreated representative substrate.

3.2.6 *sample, n*—a portion of material taken from a larger quantity for the purpose of estimating properties or composition of the larger quantity.

3.2.7 *sample tests, n*—a group of samples (one or more).

3.2.8 *test run, n*—the evaluation of coated building products in accordance with the procedure outlined in this test method.

3.2.9 *test specimen, n*—a portion of a test unit needed to obtain a single test determination.

4. Summary of Test Method

4.1 This test method is an indirect inoculation to a coated interior building product of two fungal organisms, *Aspergillus niger* and *Penicillium citrinum*. Test specimens are placed in an environmental chamber maintained at 30 ± 2 °C [86 ± 3.6 °F] and at greater than 90 % relative humidity for four weeks. Humidity is maintained by adding sufficient sterile DI water to the bottom of the covered test chamber. A continuous fungal inoculation is provided by open Petri dishes supporting seven day cultures of the two test organisms placed on a rack below the test pieces. Test specimens are removed from the chamber after four weeks exposure and examined for fungal growth. The evaluation is a macroscopic inspection of the test pieces with indirect lighting.

5. Significance and Use

5.1 An accelerated test for determining the resistance of interior coated building products to mold growth is useful in estimating the relative performance for use in interior environments under conditions favorable to fungal growth.

5.2 Static or environmental chambers provide controlled laboratory micro-environment conditions. These chambers are not intended to duplicate room conditions, and care must be taken when interpreting the results. Static chambers are not a substitute for dynamic chambers or field studies.

6. Interferences

6.1 Proper lab ventilation, hygiene, and aseptic technique must be followed to ensure fungal cultures are pure and no cross contamination of fungal strains or growth media occurs.

6.2 The exposure of test specimens to environmental conditions including temperature, humidity, and light can impact test results. To minimize variability of test results consistent handling and storage of test specimens is important.

7. Apparatus

7.1 *Environmental Chamber*—A non-corrosive covered box containing standing water placed in an incubator at 30 ± 2 °C [86 ± 3.6 °F] will expose the test specimens to a controlled environment of temperature and humidity. Containers found suitable include glass, polycarbonate³ or other plastic storage containers which are generally available. For example a

³ The 5.0 Gallon Rectangular Food Storage Containers from various suppliers have been found to work well.

nineteen liter container measuring approximately 460 mm long by 300 mm wide by 230 mm high [18 in. long by 12 in. wide by 9 in. high] can accommodate fifteen 75 by 100 mm [3 by 4 in.] test specimens suspended from rods using cable ties. Opaque chambers shall have a viewing port that permits observation of chamber controls. Chamber shall permit temperature and humidity monitoring without opening the chamber lid. Examples would include wireless or wired probes.

7.1.1 The test chamber shall be designed so that no condensate forming on the top interior surface will drip onto the test specimens. For stand-alone chambers, this can be accomplished by designing the top so that the interior surface is at an angle of at least 30 degrees, relative to the plane of the bottom of the chamber. A sheet of polycarbonate secured at an angle, or hinged or joined sheets of polycarbonate attached to the underside of the lid will direct the condensation away from the test samples. Condensation inside the test chamber is not a concern as it indicates that humidity is being maintained.

7.1.1.1 A non-corrosive open grid is placed at the bottom of the test chamber above the water level supporting 100 by 15 mm [4 by 5/8 in.] sterile Petri dishes alternating the two fungal organisms. See Fig. 1 for non-corroding open grid example. Place sufficient Petri dishes on the rack to fill the grid. The plastic grid designed to cover recessed ceiling fixtures or similar works well.

7.1.1.2 Position test specimens by suspending them from rods using plastic cable ties. Samples must be 50 to 100 mm [2 to 4 in.] above the inoculated Petri dishes. The minimum distance between adjacent specimens and between test specimens and chamber walls shall be at least 25 mm [1 in.]. Materials used as the rods or mounting racks shall be non-corroding and of sufficient strength to support specimens throughout the duration of the test. Use of engineered plastics such as polycarbonate has been found suitable. Fig. 2 shows a photo of typical chamber construction. Fig. 3 illustrates use of cable ties to hang test specimens from rods.

7.2 *Measurement Instruments*, capable of accurate and precise measures of temperature and humidity. See Section 12.

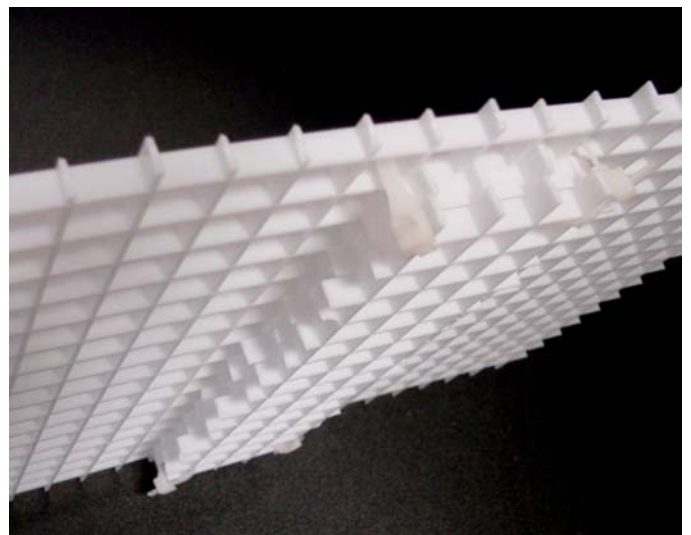


FIG. 1 Non-corroding Open Grid for Placement in Bottom of the Environmental Chamber



FIG. 2 Typical Environment Chamber Set Up

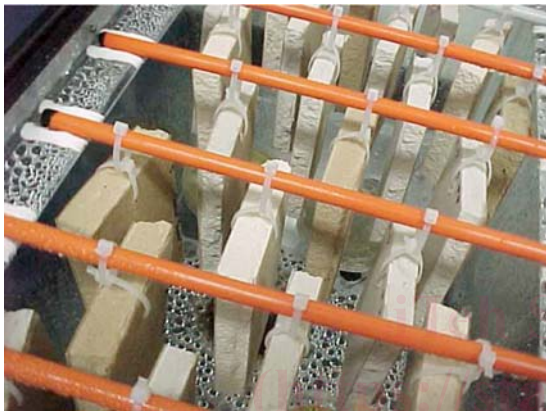


FIG. 3 Cable Ties used to Hang Test Specimens from Rods Inside of the Environmental Chamber

7.3 Incubator or Controlled Temperature Room maintained at 30 ± 2 °C [86 ± 3.6 °F].

8. Reagents and Materials

8.1 Cultures—*Aspergillus niger*, ATCC⁴ 6275 or IMI/CABI Bioscience⁵ 45551, *Penicillium citrinum* ATCC 9849 or IMI/CABI Bioscience 321326.

8.1.1 Selection of the appropriate test organisms is extremely important and must be representative of the types of organisms found or likely to be found on the interior coated building products being tested. The organisms named in 8.1 are not representative of all potential fungal organisms that may be found growing on interior coated building products. Other fungal organisms may also be used in separate evaluations, but the specified organisms in 8.1 shall be used and reported. The potential for interferences between non-specified fungal test organisms shall be considered when using organisms other than those named in 8.1.

NOTE 1—Subcommittee D01.28 reviewed the published study listed in the Reference section of this document and determined the organisms in 8.1 as appropriate.

⁴ Cultures can be obtained from American Type Culture Collection, P.O. Box 1549, Manassass, VA 20108 or Mycological Services, P.O. Box 1056, Crawfordsville, IN 47933.

⁵ Cultures can be obtained from IMI/CABI Bioscience, Nosworthy Way, Wallingford, Oxfordshire, OX108DE UK.

8.2 Chamber Controls—Open PDA plates placed on the bottom of the chamber at opposite corners and near the center.

8.3 Material Controls, if available, see 3.2.

8.4 Purity of Reagents—Water shall be distilled water or higher purity. See Specification D1193.

8.5 Sabouraud Dextrose Agar or media appropriate for fungi selected.

8.6 Sterile Disposable Cotton-tipped Swabs.

8.7 Sterile 100 by 15 mm (4 by 5/8 in.) Petri Dishes.

9. Hazards

9.1 This test must be performed by trained individuals in laboratories specially equipped for conducting microbiological tests.

10. Sampling and Test Specimens

10.1 Sampling shall be representative of the product being evaluated.

10.2 Test Specimens:

10.2.1 A minimum of three test specimens shall be cut from each sampled coated interior building product to be evaluated. The number of test specimens will be reported in the results.

10.2.2 Additional replicates should be available to rerun the test if necessary.

11. Preparation of Apparatus and Inoculum

11.1 Clean and sanitize the environmental chambers prior to use. Add approximately 25 mm [1 in.] of water to the bottom of the container. Ensure the water level is sufficient to provide humidity through the duration of the test. If the test specimens absorb the water or water is lost through the seal of the lid, additional water must be added to ensure the relative humidity remains at 90 % or greater for the duration of the test. The water level should be at least 25 mm [1 in.] below the rack supporting the Petri dishes of the fungal test organisms.

NOTE 2—Petroleum jelly or similar product may be used between the lid and the container to improve the seal.

11.2 Insert the temperature/humidity sensor or data logger into the environmental chamber and set in an incubator or other temperature controlled chamber set at 30 ± 2 °C [86 ± 3.6 °F] to equilibrate for 24 h before starting the test. Record the temperature and humidity not less than every 7 days. If temperature and humidity readings are outside the parameters set in 4.1, results shall be discarded and testing restarted with new test pieces. Temperature and humidity measurements shall be included in the final report.

11.3 Prepare spore suspensions of each test fungi from 7 to 14 day old well sporulating cultures. Maturation of the fungal organisms designated in 8.1 may not occur at the same rate. *Penicillium citrinum* typically takes longer to sporulate than *Aspergillus niger* so cultivation should begin earlier assuring both organisms are sporulating when placed in the environmental chamber. Stock cultures may be kept for no more than four months at 3 to 10 °C [37 to 50 °F].

11.3.1 To prepare the inoculum, dislodge fungal spores from agar by rolling a sterile cotton-tipped swab moistened with



FIG. 4 Example of Sporulating Fungal Plates Arranged in an Alternating Pattern in the Environmental Chamber

sterile distilled water across the sporulating fungi. Transfer the spores from the cotton tipped swab to a test tube containing 5 mL of sterile distilled water and a nontoxic wetting agent for each test organism. Blend the fungal spore suspension on the vortex mixer for 10 s to liberate spores from hyphae and to break up spore clumps. Repeat this procedure for each fungal organism used.

11.4 Pour 25 mL [1.0 oz.] of Sabouraud Dextrose or appropriate agar for test organisms named in 8.1 into 100 by 15 mm [4 by 5/8 in.] sterile Petri dishes and allow the agar to solidify. Prepare sufficient Petri dishes to fill the rack placed at the bottom of each environmental chamber. For the chamber size referenced in 7.1, twelve 100 by 15 mm [4 by 5/8 in.] Petri dishes are appropriate.

11.5 Transfer 200 to 300 μ L (microlitres) of a single spore suspension to each Petri dish. Spread the inoculum across the surface of the agar with a cell spreader or equivalent, cover and incubate at 28 to 30 °C [82 to 86 °F] for 7 to 14 days or until dense sporulating fungal growth is present. Prepare an equal number of Petri dishes for both test organisms in sufficient quantity to cover the rack placed at the bottom of the environmental chamber.

11.6 If growth is sparse or absent after 14 days, prepare fresh inoculums as discussed in 11.3 and repeat inoculation of new agar plates.

11.7 Place the sporulating agar plates on the rack of the equilibrated environmental chamber referenced in 11.2 and remove covers. Place Petri dishes alternating between the two test organisms to ensure equal exposure of the test specimens to both fungal strains. Fig. 4 shows the alternating pattern of agar plates on the rack at the bottom of the environmental chamber.

12. Calibration and Standardization

12.1 Instruments for temperature measurement shall measure with a precision of ± 2 °C [± 3.6 °F] over the full range of instrument capability.

12.2 Instruments for humidity measurement shall measure with a precision of ± 5 % RH at 95 % RH.

12.3 Both types of measurement instruments shall be calibrated no less than annually by a laboratory using standards that are documented traceable to NIST standards.

13. Conditioning

13.1 Equilibrate triplicate test specimens of each sample and condition to room temperature before starting test.

14. Procedure

14.1 *Sample Preparation*—Wear disposable powder-free plastic gloves or equivalent when handling test pieces. Approximate test specimen size shall be approximately 75 by 100 mm [3 by 4 in.] or approximately 75 cm² [12 in.²].

NOTE 3—A 25 mm [1 in.] grid may be drawn with a pencil on the surface of the test specimen to help standardize the evaluation of the sample for fungal growth.

14.2 Add approximately 25 mm [1 in.] of distilled water to the bottom of the test chamber. Remove the lids of the 7 to 14 day old sporulating culture plates and place them on the tray in the bottom of the chamber above the water level.

14.3 *Exposure*—Place test specimens and chamber controls randomly in the environmental chamber allowing free air circulation. Test specimens shall be placed so there is no contact between them, the inside walls or the lid of the chamber. Required minimum spacing between test specimens is 25 mm [1 in.]. Place environmental chamber in the incubator or temperature room maintained at 30 \pm 2 °C [86 \pm 3.6 °F]. Record twelve regularly spaced temperature and humidity observations over the 4 week incubation period.

14.3.1 After four weeks exposure remove the test specimens from the environmental chambers and determine the percent fungal growth across the surface. Rate the test specimen in accordance with the scale in 15.3 and record the temperature and humidity at the time of test completion.