**Designation: ~~D5589 – 09 (Reapproved 2013)~~ D5589 – 19**

# Standard Test Method for Determining the Resistance of Paint Films and Related Coatings to Algal Defacement[1]

This standard is issued under the fixed designation D5589; 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 accelerated method for determining the relative resistance of a paint or coating film to algal growth.

NOTE 1—It is hoped that a ranking of relative performance would be similar to that ranked from outdoor exposures. However, this test method should not be used as a replacement for exterior exposure since many other factors, only a few of which are listed will affect those results.

NOTE 2—ASTM weathering standards are no longer referenced in this document, but Practices D822, D4141, D4587, D5031, and D6695 are commonly used.

1.2 The values stated in SI units are to be regarded as the standard. The values given in parentheses are for information only.

1.3 *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~~ safety, health, and ~~health~~environmental practices and determine the applicability of regulatory limitations prior to use.*

1.4 *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]

D822 Practice for Filtered Open-Flame Carbon-Arc Exposures of Paint and Related Coatings
D4141 Practice for Conducting Black Box and Solar Concentrating Exposures of Coatings
D4587 Practice for Fluorescent UV-Condensation Exposures of Paint and Related Coatings
D5031 Practice for Enclosed Carbon-Arc Exposure Tests of Paint and Related Coatings
D6695 Practice for Xenon-Arc Exposures of Paint and Related Coatings



## 3. Summary of Test Method

3.1 This test method outlines a procedure to (*1*) prepare a suitable specimen for testing, (*2*) inoculate the specimen with a mixture of the proper algal species, (*3*) expose the inoculated samples under the appropriate conditions for growth, and (*4*) provide a schedule and guidelines for visual growth ratings. This test method is not designed to include all the necessary procedures to maintain the proper microbiological techniques required to provide the most accurate results.

## 4. Significance and Use

4.1 Defacement of paint and coating films by algal growth is a common phenomenon under certain conditions. It is generally known that differences in the environment, lighting, temperature, substrate, and other factors in addition to the coating composition affect the susceptibility of a given painted surface. This test method attempts to provide a means to comparatively evaluate different coating formulations for their relative performance under a given set of conditions. It does not imply that a coating that resists growth under these conditions will necessarily resist growth in the actual ~~application (see~~ application.~~Note 1~~).

[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.

Current edition approved ~~Oct. 1, 2013~~Dec. 1, 2019. Published ~~October 2013~~December 2019. Originally approved in 1994. Last previous edition approved in ~~2009~~2013 as ~~D5589 – 09.~~D5589 – 09 (2013). DOI: ~~10.1520/D5589-09R13.~~10.1520/D5589-19.

[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.

4.2 Familiarity with microbiological techniques is required. This test method should not be used by persons without at least basic microbiological training.

## 5. Apparatus and Materials

5.1 *Balance,* capable of weighing to 0.10 g.

5.2 *Incubator,* or other device capable of maintaining a constant temperature between 25 ± 2°C, relative humidity of ≥85 %, and having a constant ~~fluorescent~~ full spectrum (see Note 3) light source.

5.3 *Refrigerator.*

5.4 *Petri Dishes,* 100 by 15 mm (3.9 by 0.6 in.).

5.5 *Autoclave.*

5.6 *Paint Brush,* coarse bristle, 12 to 19 mm (½ to ¾ in.).

5.7 *Test Substrate,* filter paper, either regular paper or glass fiber, approximately 4.2 cm (1.65 in.) in diameter, or drawdown paper (~~unlaquered~~(unlacquered chart paper) approximately 21.6 by 28.0 cm (8.5 by 11 in.), cut into ten strips, approximately 21.6 by 2.8-cm (8.5 by ~~1.1-in.) strips may be used.~~1.1-in.).

5.8 *Tissue Grinder.*

5.9 *Atomizer or Chromatography Sprayer.*

5.10 *Sterile Glass Rods, Forceps, 250-mL Glass Erlenmeyer Flask,* and other routine microbiological equipment.

5.11 ~~*Allen's Medium*~~ *BG-11 Medium with Trace Metals Mixture.* [3] ~~or Bold's Basal Medium ingredients (see 6.3).~~

5.12 *Distilled Water.*

NOTE 3—Fluorescent or LED D65 bulbs, 12 hours on, 12 off. Follow manufacturers' recommendations regarding light bulb service life and when to replace them.

## 6. Reagents and Materials

6.1 *Purity of Reagents*—Reagent grade chemicals should be used in all tests. Unless otherwise indicated, it is intended that all reagents should conform to the specifications of the Committee on Analytical Reagents of the American Chemical Society, where such specifications are available.[4] Other grades may be used, provided they are first ascertained to be of sufficiently high purity to permit use without decreasing the accuracy of the determination.

6.2 *Purity of Water*—Unless otherwise indicated, references to water are understood to mean distilled water or water of equal or higher purity.

~~6.3 *Allen's Medium*—Prepare liquid medium by dissolving in 1000 mL of water the following reagents in the designated amounts:~~

| ~~Reagent~~ | ~~Amount, g/1000 mL~~ |
|---|---|
| ~~$NaNO_3$~~ | ~~1.5~~ |
| ~~$K_2HPO_4$~~ | ~~0.039~~ |
| ~~$MgSO_4 \cdot 7H_2O$~~ | ~~0.075~~ |
| ~~$CaCl_2 \cdot 2H_2O$~~ | ~~0.027~~ |
| ~~$Na_2CO_3$~~ | ~~0.020~~ |
| ~~$Na_2SiO_3 \cdot 9H_2O$~~ | ~~0.058~~ |
| ~~Citric acid~~ | ~~0.006~~ |
| ~~EDTA[A]~~ | ~~0.006~~ |
| ~~Allen's trace element solution~~ | ~~1.0 mL[B]~~ |
| ~~Distilled water~~ | ~~to 1000 m~~ |
| ~~Ferric citrate (see Note 2)~~ | ~~0.006 (see Note 2)~~ |

~~[A] Ethylenediaminetetraacetic acid, disodium salt~~
~~[B] *Allen's Trace-Element Solution:*~~

~~Dissolve in 500 mL of distilled water:~~

[3] ~~Bold, H. C., Wynne, M. J., "Introduction to the Algae," Prentiss-Hall, Englewood Cliffs, NJ, 1978, pp. 574–5.~~BG-11 medium, trace metals mix are available through Sigma-Aldrich.

[4] ~~Kirsop B. E., and Snell J. J. S., "*Maintenance of Microorganisms*," Academic Press, Harcourt Brace Jovanovich, Orlando, FL, 1984, p. 158.~~

[4] ~~*Reagent Chemicals, American Chemical Society Specifications,*~~ *ACS Reagent Chemicals, Specifications and Procedures for Reagents and Standard-Grade Reference Materials,* American Chemical Society, Washington, DC. For suggestions on the testing of reagents not listed by the American Chemical Society, see *Analar Standards for Laboratory Chemicals*, BDH Ltd., Poole, Dorset, U.K., and the *United States Pharmacopeia and National Formulary*, U.S. Pharmacopeial Convention, Inc. (USPC), Rockville, MD.

| Reagent | Amount, g |
|---|---|
| $H_3BO_3$ | 2.86 |
| $MnCl_2·4H_2O$ | 1.81 |
| $ZnSO_4·7H_2O$ | 0.222 |
| $Na_2MoO_4·2H_2O$ | 0.391 |
| $CuSO_4·5H_2O$ | 0.079 |
| $Co(NO_3)_2·6H_2O$ | 0.0494 |

NOTE 2—The ferric citrate must be autoclaved separately. The ferric citrate should be added after the medium has cooled from being autoclaved.

6.3.1 Adjust the pH of the medium to 7.8 using 1.0 *M* NaOH/1.0 M HCl and autoclave at 121°C (without ferric citrate added) to 45 to 50°C before aseptically adding the ferric citrate (see Note 2).

6.3.2 *Allen's Agar*—Prepare by dissolving 15 g of agar in 1000 mL Allen's Medium before autoclaving. Cool to 45 to 50°C before aseptically adding the ferric citrate. After mixing, pour the media into petri dishes.

6.4 *Bold's Basal Medium*—Prepare ten individual stock solutions in distilled water as indicated:

| Stock Solutions | g/400 mL |
|---|---|
| 1. $NaNo_3$ | 10.0 |
| 2. $MgSO_4·7H_2O$ | 3.0 |
| 3. NaCl | 1.0 |
| 4. $K_2HPO_4$ | 3.0 |
| 5. $KH_2PO_4$ | 7.0 |
| 6. $CaCl_2·2H_2O$ | 1.0 |
| **Trace Element Solutions:** | **g/L** |
| 7. $ZnSO_4·7H_2O$ | 8.82 |
| $MnCl_2·4H_2O$ | 1.44 |
| $MoO_3$ | 0.71 |
| $CuSO_4·5H_2O$ | 1.57 |
| $Co(NO_3)_2·6H_2O$ | 0.49 |
| Distilled Water | to 1 L |
| Autoclave to dissolve. | |
| 8. $H_3BO_3$ | 11.42 |
| 9. EDTA-KOH solution: | |
| EDTA | 50.0 |
| KOH | 31.0 |
| 10. $FeSO_4·7H_2O$ | 4.98 |
| $H_2SO_4$ (concentrate) | 1.0 mL |

6.4.1 Combine 10 mL each of Stock Solutions 1 through 6 with 1 mL each of Stock Solutions 7 through 10 in 936 mL distilled water. Autoclave at 121°C.



6.3 A variety of algal cultures, including wild strains isolated from paint films, may be used in this protocol. Choose strains from the following list, use field isolates or use other strains found to grow satisfactorily under the protocol conditions. It is recommended to choose at least one culture from each type. The choice of strains should be agreed upon between the parties involved in the testing.[5]

| Algae<br>Algae | Collection/Strain[A]<br>Collection/Strain |
|---|---|
| Unicellular Green | |
| *Chlorella sp.* | ATCC 7516 |
| *Chlorella vulgaris* | ATCC 11468 |
| Filamentous Green | |
| *Ulothrix gigas* | ATCC 30443 |
| *Trentepohlia aurea* | UTEX 429 |
| *Trentepohlia odorata* | CCAP 483/4 |
| Colony-forming Green | |
| *Scenedesmus quadricauda* | ATCC 11460 |
| Filamentous Bluegreen | |
| *Oscillatoria sp.* | ATCC 29135 |
| *Calothrix sp.* | ATCC 27914 |

[5] Available from the following culture collections and found suitable for this test: American Type Culture Collection (ATCC), 12301 Parklawn Drive, Rockville, MD 20852; University of Texas (UTEX), Department of Botany, The University of Texas at Austin, Austin, TX 78713-7640; Culture Collection of Algae and Protozoa (CCAP), Institute of Freshwater Ecology, The Windermere Laboratory, Far Sawrey, Ambleside, Cumbria LA22 OLP, U.K. Grow purchased cultures in media and under incubation conditions recommended by culture collection.