



Standard Test Method for Quantitative Sporocidal Three-Step Method (TSM) to Determine Sporocidal Efficacy of Liquids, Liquid Sprays, and Vapor or Gases on Contaminated Carrier Surfaces¹

This standard is issued under the fixed designation E 2414; 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 determines the efficacy of sporocidal agents on microorganisms dried on the surface of solid carriers.

1.2 This test method can be used to evaluate sporocidal products (or decontaminant, disinfectant, and so forth), suspected, claimed, or assumed to have sporocidal activity. This test method allows:

1.2.1 Establishing the sporocidal efficacy of different disinfectants;

1.2.2 Identifying the effect, if any, of the surface materials on sporocidal efficacy; and

1.2.3 Comparing the relative resistance to disinfection of different microbial species or strains.

1.3 The values stated in SI units are to be regarded as the standard.

1.4 Strict adherence to the protocol is necessary for the validity of the test results.

1.5 *Follow all the safety guidance of the institution in which the testing is being conducted. 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 and health practices and determine the applicability of regulatory limitations prior to use.*

2. Referenced Documents

2.1 *ASTM Standards*:²

E 1054 Test Methods for Evaluation of Inactivators of Antimicrobial Agents

¹ This test method is under the jurisdiction of ASTM Committee E54 on Homeland Security Applications and is the direct responsibility of Subcommittee E54.03 on Decontamination.

Current edition approved June 15, 2005. Published July 2005.

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. Summary of Test Method

3.1 This test method determines the efficacy of disinfectants on spores of *Bacillus subtilis* dried on carriers according to a general technique first described by J. L. Sagripanti and A. Bonifacino (**1, 2**)³.

3.1.1 Spores of *B. subtilis* can be and have been replaced by similar amounts of spores of *Bacillus anthracis* to substantiate claims as may be needed in biodefense.

3.1.2 The material of the carrier is selected according to claims or intended use, or both, of the disinfectant product. General claims made to decontaminate metallic and polymeric materials are tested on carriers made of stainless steel and silicone medical rubber, respectively. Flat coupons (0.5 by 0.5 cm) are preferable. As may be required by claims or intended use, the test method can be accurately used also on a variety of carriers with diverse geometrical characteristics (additional examples of materials were reported by Sagripanti and Bonifacino (**1, 2**)).

3.1.3 Although the test method described herein refers specifically to liquid disinfectants, the same procedure can be used to assess sporocidal activity of vapors and gaseous sporocidal agents, provided adequate containment is accomplished (see **10.4.5**).

4. Significance and Use

4.1 The quantitative micromethod described herein was designed to fulfill the following specifications:

4.1.1 To be quantitative (the number of viable spores loaded into carriers is determined by the spores quantitatively recovered in the controls),

4.1.2 Sensitive (sporocidal activity can be accurately measured up to 7 Log₁₀ inactivation, see Section **12**),

4.1.3 Reproducible (standard deviation of spore killing is smaller than 1 Log and usually closer to 1/10 of a Log, see Section **12**),

³ The boldface numbers in parentheses refer to the list of references at the end of this standard.