



Standard Test Method for Size-Differentiated Counting of Particles and Fibers Released from Cleanroom Wipers Using Optical and Scanning Electron Microscopy¹

This standard is issued under the fixed designation E 2090; 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.

INTRODUCTION

Techniques for determining the number of particles and fibers that can potentially be released from wiping materials consist of two steps. The first step is to separate the particles and fibers from the wiper and capture them in a suitable medium for counting, and the second step is to quantify the number and size of the released particles and fibers.

The procedure used in this test method to separate particles and fibers from the body of the wiper is designed to simulate conditions that the wiper would experience during typical use. Therefore, the wiper is immersed in a standard low-surface-tension cleaning liquid (such as a surfactant/water solution or isopropyl alcohol/water solution) and then subjected to mechanical agitation in that liquid. The application of moderate mechanical energy to a wiper immersed in a cleaning solution is effective in removing most of the particles that would be released from a wiper during typical cleanroom wiping. This test method assumes the wiper is not damaged by chemical or mechanical activity during the test.

Once the particles have been released from the wiper into the cleaning solution, they can be collected and counted. The collection of the particles is accomplished through filtration of the particle-laden test liquid onto a microporous membrane filter. The filter is then examined using both optical and scanning electron microscopy where particles are analyzed and counted. Microscopy was chosen over automated liquid particle counters for greater accuracy in counting as well as for morphological identification of the particles.

The comprehensive nature of this technique involves the use of a scanning electron microscope (SEM) to count particles distributed on a microporous membrane filter and a stereo-binocular optical microscope to count large fibers. Computer-based image analysis and counting is used for fields where the particle density is too great to be accurately determined by manual counting.

Instead of sampling aliquots, the entire amount of liquid containing the particles and fibers in suspension is filtered through a microporous membrane filter. The filtering technique is crucial to the procedure for counting particles. Because only a small portion of the filter will actually be counted, the filtration must produce a random and uniform distribution of particles on the filter. After filtration, the filter is mounted on an SEM stub and examined using the optical microscope for uniformity of distribution. Large fibers are also counted during this step. Once uniformity is determined and large fibers are counted, the sample stub is transferred to the SEM and examined for particles. A statistically valid procedure for counting is described in this test method. The accuracy and precision of the resultant count can likewise be measured.

This test method offers the advantage of a single sample preparation for the counting of both particles and fibers. It also adds the capability of computerized image analysis, which provides accurate recognition and sizing of particles and fibers. Using different magnifications, particles from 0.5 to 1000 μm or larger can be counted and classified by size. This procedure categorizes three classes of particles and fibers: small particles between 0.5 and 5 μm ; large particles greater than 5 μm but smaller than 100 μm ; and large particles and fibers equal to or greater than 100 μm . The technique as described in this test method uses optical microscopy to count large particles and fibers greater than 100 μm and SEM to count the other two classes of particles. However, optical microscopy can be employed as a substitute for SEM to count the large particles between 5 and 100 μm^2 .

1. Scope

1.1 This test method covers testing all wipers used in cleanrooms and other controlled environments for characteristics related to particulate cleanliness.

1.2 This test method includes the use of computer-based image analysis and counting hardware and software for the counting of densely particle-laden filters (see 7.7-7.9). While the use of this equipment is not absolutely necessary, it is strongly recommended to enhance the accuracy, speed, and consistency of counting.

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

1.4 *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:*³

D 1193 Specification for Reagent Water

F 25 Test Method for Sizing and Counting Airborne Particulate Contamination in Cleanrooms and Other Dust-Controlled Areas

F 312 Test Methods for Microscopical Sizing and Counting Particles from Aerospace Fluids on Membrane Filters

2.2 *Other Documents:*

ISO 14644-1 Cleanrooms and Associated Controlled Environments – Classification of Air Cleanliness⁴

ISO 14644-2 Cleanrooms and Associated Controlled Environments – Part 2: Specifications for testing and monitoring to prove continued compliance with ISO 14644-1⁴

Fed. Std. 209E Airborne Particulate Cleanliness Classes in Cleanrooms and Clean Zones⁵

3. Terminology

3.1 *Definitions of Terms Specific to This Standard:*

3.1.1 *automatic counting, n*—counting and sizing performed using computerized image analysis software.

3.1.2 *cleanroom wiper, n*—a piece of absorbent knit, woven, nonwoven, or foam material used in a cleanroom for wiping, spill pickup, or applying a liquid to a surface.

¹ This test method is under the jurisdiction of ASTM Committee E21 on Space Simulation and Applications of Space Technology and is the direct responsibility of Subcommittee E21.05 on Contamination.

Current edition approved Nov. 1, 2006. Published December 2006. Originally approved in 2000. Last previous edition approved in 2000 as E 2090 - 00.

² The counting of particles 5 to 100 µm by optical microscopy is not described in this test method. However, procedures for counting particles in this size range are described in the Test Methods **F 25** and **F 312**.

³ 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 Available from American National Standards Institute, 11 W. 42nd St., 13th Floor, New York, NY 10036.

⁵ Cancelled Nov. 29, 2001 and replaced with **ISO 14644-1** and **ISO 14644-2**, FED-STD-209E may be used by mutual agreement between buyer and seller. Available from US Government Printing Office, Superintendent of Documents, 732 N. Capitol St., NW, Mail Stop: SDE, Washington, DC 20401.

3.1.2.1 *Discussion*—Characteristically, these wipers possess very small amounts of particulate and ionic contaminants and are primarily used in cleanrooms in the semiconductor, data storage, pharmaceutical, biotechnology, aerospace, and automotive industries.

3.1.3 *effective filter area, n*—the area of the membrane which entraps the particles to be counted.

3.1.4 *fiber, n*—a particle having a length to diameter ratio of 10 or greater.

3.1.5 *illuminance, n*—luminous flux incident per unit of area.

3.1.6 *particle, n*—a unit of matter with observable length, width, and thickness.

3.1.7 *particle size, n*—the size of a particle as defined by its longest dimension on any axis.

4. Summary of Test Method

4.1 *Summary of Counting Methods*—See the following:

Counting Technique	Particle Size Range		
	>100 µm	5–100 µm ^A	0.5–5 µm
Stereobinocular optical microscope	20× manual		NA ^B
Scanning electron microscope	NA	200× auto	3000× manual or automatic ^B

^ASee Footnote 2.

^BNA = not applicable.

5. Significance and Use

5.1 This test method provides for accurate and reproducible enumeration of particles and fibers released from a wiper immersed in a cleaning solution with moderate mechanical stress applied. When performed correctly, this counting test method is sensitive enough to quantify very low levels of total particle and fiber burden. The results are accurate and not influenced by artifact or particle size limitations. A further advantage to this technique is that it allows for morphological as well as X-ray analysis of individual particles.

6. Apparatus

6.1 *Scanning Electron Microscope*, with high-quality imaging and computerized stage/specimen mapping capability.

6.2 *Stereo-Binocular Optical Microscope*, with at least 40×-magnification capability equipped with a two-arm, adjustable-angle variable-intensity light source and a specimen holding plate.

6.3 *Orbital Shaker*, that provides 20-mm (¾-in.) diameter circular motion in a horizontal plane at 150 r/min.

6.4 *Microanalytical Stainless Steel Screen-Supported Membrane Filtration Apparatus*, with stainless steel funnel, TFE-fluorocarbon gasket and spring clamp.

6.5 *Vacuum Pump*, capable of providing a pressure of 6.5 kPa (65 mb) (49 torr) or lower.

6.6 *Cold Sputter/Etch Unit*, with gold or gold/palladium foils.

6.7 *Video Camera (3-CCD preferable)*, that can be attached to the stereo-binocular microscope and a monitor to provide video microscopy capability.

6.8 *Personal Computer (486-Type Processor or Better) and Monitor*.

6.9 *Frame-Grabbing Hardware and Image Analysis Software*, compatible with the personal computer.⁶

6.10 *Hand-Operated Tally Counter*.

6.11 *Stage Micrometer*, with 0.1- and 0.01-mm subdivisions.

6.12 *Horizontal, Unidirectional Flow Workstation*, with ISO Class 5 (**Fed. Std. 209** Class 100) or cleaner air.

7. Materials

7.1 *Deionized Water*, in accordance with Specification **D 1193**, Type III, $4.0 \times 10^{-6} (\Omega\text{-cm})^{-1}$ or better.

7.2 *Cleanroom Gloves* (for example, unpowdered latex gloves).

7.3 *Fine-Point, Duckbill Tweezers*.

7.4 *Forceps*, two pairs, with flat gripping surface tips.

7.5 *Glass Beakers*, 1.5 L, cleaned in accordance with **10.2.1**.

7.6 *Polyethylene Photographic Tray*, approximately 250 by 340 by 45 mm cleaned in accordance with **10.2.1**.

7.7 *Polycarbonate Membrane Filters* (typically 0.1- to 0.4- μm pore size), white, and 25-mm diameter.

7.8 *Petri Slide*, 47 mm.

7.9 *SEM Aluminum Specimen Stubs*, typically 32-mm diameter by 10-mm height.

7.10 *Polystyrene Latex Microspheres* (sizes 0.5 and 5 μm) for use in calibration (see Section 9).

7.11 *Carbon Paint*, for SEM stub preparation.

7.12 *Low-Surface-Tension Cleaning Liquid*—Any 8- to 10-mole ethoxylated-octyl- or nonyl-phenol-type surfactant⁷ prepared as a 0.1 % stock solution in deionized water. This solution will facilitate the release of both nonpolar and polar contaminants and can serve as a general test standard across industries. However, this test method is not limited to a specific cleaning solution and only requires that the cleaning liquid used be relatively free of particles and fibers. It is recommended that the cleaning liquid most relevant to the product end use be considered for this test method.

8. Preparation of Apparatus

8.1 *Setting Up Stereo-Binocular Optical Microscope*—See Section 10.

8.2 *Fiber Counting by Optical Microscopy*—See Section 10.

8.3 *Setting Up Scanning Electron Microscope (SEM)*—See Section 10.

8.4 *Particle Counting by SEM*—See Section 10.

⁶ “Image-Pro Plus,” Version 3.0, available from Media Cybernetics, has been found to be satisfactory for this test method.

The sole source of supply of the apparatus known to the committee at this time is Media Cybernetics. If you are aware of alternative suppliers, please provide this information to ASTM International Headquarters. Your comments will receive careful consideration at a meeting of the responsible technical committee,¹ which you may attend.

⁷ Triton® X-100 manufactured by Rohm and Haas Co. has been found to be satisfactory for this test method.

The sole source of supply of the apparatus known to the committee at this time is Rohm and Haas Co. If you are aware of alternative suppliers, please provide this information to ASTM International Headquarters. Your comments will receive careful consideration at a meeting of the responsible technical committee,¹ which you may attend.

9. Calibration and Standardization

9.1 For the fiber counting by optical microscopy, the size calibration at 20 $\times$ magnification can be done by comparing the fiber sizes, as visualized in the video monitor, with the rulings on the stage micrometer (with 0.1- and 0.01-mm subdivisions). For the equipment described above, a linear dimension of 8 mm in the video screen equaled 100 μm . The conversion factors are equipment-dependent and users of this test method shall establish the relation between screen size and object size.

9.2 In the SEM study, to determine the values of the start and the end areas for the computer-assisted automatic particle counting, it is necessary to perform the size calibration study by experimenting with standard-sized particles such as polystyrene microspheres or actual particles of known dimensions which can be ascertained by using the micrometre bar measurement tool available on most SEMs.

9.3 To prepare a stub with 0.5- and 5- μm spheres, add 10 μL of each of the 0.5- and 5- μm sphere suspensions to a beaker containing 500 mL of deionized water.

9.4 Filter the solution using a new membrane filter.

9.5 Prepare the SEM stub. Save the stub in a clean container as a standard size reference for the automatic particle counting at 200 and at 3000 $\times$.

9.6 For the manual procedure at 3000 $\times$, avoid counting particles having approximate linear lengths of 25 mm and up, as those will have sizes larger than 5 μm as determined from measurements done against the micrometre bars at various magnifications in the SEM.

10. Procedure

10.1 The procedure consists of two parts: preparing the sample and counting the fibers and particles. Fibers and particles greater than 100 μm are counted using an optical microscope at 20 $\times$ magnification; large (between 5 and 100 μm) and small (between 0.5 and 5 μm) particles are counted using an SEM at 200 and 3000 $\times$ magnifications respectively. Both manual and computer-aided automatic counting methods are used in this procedure.

10.1.1 *Sample Preparation*—Sample preparation consists of two steps:

10.1.1.1 Preparation of a background filter stub and

10.1.1.2 Preparation of the sample filter stub containing particles released from a cleanroom wiper.

10.2 *Preparation of a Background Filter Stub*—To measure the background level of particles from the glassware, polyethylene tray, and filtration system, it is necessary to prepare an experimental blank.

10.2.1 The cleaning of the photographic tray, glassware, and the filtration apparatus should be accomplished in the following manner:

10.2.1.1 Clean the photographic tray thoroughly by rinsing the inner surface at least five times with deionized water.

10.2.1.2 Ultrasonically clean the glassware, storage containers, and filtration assembly then thoroughly rinse using deionized water.

10.2.1.3 Allow all containers and assemblies to drain dry in the unidirectional flow workstation.

10.2.1.4 Store all containers and assemblies, including the photographic tray, in the clean workstation to prevent environmental contamination.

10.2.2 The choice of the cleaning solution should reflect the liquid that the wiper will come in contact with during actual use. A typical example of a cleaning solution would be a low-concentration surfactant/deionized water mixture (see 7.12). This mixture serves well as a standard for general comparative purposes since it facilitates the release of both nonpolar and polar contaminants. However, this test method is not limited to a specific cleaning solution and only requires that the solution be relatively free of particles and fibers. The specific cleaning solution used must be reported in accordance with 12.1.2. A low-concentration surfactant/deionized water mixture as described in 7.12 is used in the test method example.

10.2.3 *Stock 0.1 % Surfactant Solution Preparation:*

10.2.3.1 Place 300 mL of deionized water in a 1.5-L beaker.

10.2.3.2 Place the beaker on a hot plate and raise the water temperature to 40°C.

10.2.3.3 Slowly add 1 g (35 drops) of the concentrated surfactant into the hot water.

10.2.3.4 Mix well to make the solution homogeneous.

10.2.3.5 Add more deionized water to raise the volume to 1 L.

10.2.3.6 Aliquots from this stock solution will be used for the test procedure.

10.2.3.7 The stock solution shall be prepared daily to prevent any biological growth.

10.2.4 *Blank Preparation:*

10.2.4.1 Place 500 mL of deionized water into the clean photographic tray.

10.2.4.2 Place the tray on the platform of an orbital shaker (Fig. 1) stationed inside the hood of a clean workstation having of an ISO Class 5 (Fed. Std. 209 Class 100) or cleaner environment.

10.2.4.3 Add a 25-mL aliquot from the stock 0.1 % surfactant cleaning solution to the water in the tray.

10.2.4.4 Run the orbital shaker at 150 r/min for 5 min. Some equipment may require somewhat lower rotation rates, for example, 130 r/min, to avoid liquid spills.

10.2.5 Insert the base of the filtration assembly into the stopper. Place the TFE-fluorocarbon gasket onto the base, then

place the stainless steel screen on top of the gasket.⁸ Insert the stopper holding the base, gasket and screen into the filtration flask.

10.2.6 Connect the filtration flask to the vacuum pump but do not turn the pump on at this point.

10.2.7 Transfer a 25-mm diameter polycarbonate membrane filter to a petri slide with the filter shiny (coated) side facing up using a fine-point duckbill tweezer.

10.2.8 Rinse the filter gently under running deionized water.

10.2.9 Using the tweezers, slide the filter from the petri slide onto the stainless steel screen of the filtration apparatus with the shiny (coated) side of the filter facing up.

10.2.10 Place the stainless steel funnel on top of the filter and clamp the assembly together. Fig. 2 shows the fully assembled vacuum filtration apparatus.

10.2.11 Pour the water from the tray into a clean 1.5-L beaker.

10.2.12 Add approximately 25 mL of clean deionized water to rinse the tray and pour this water into the beaker as well.

10.2.13 Slowly pour the water from the beaker into the filtration funnel until the funnel is approximately two thirds full.

10.2.14 Turn on the vacuum pump and adjust the vacuum so that the filtration rate is approximately 50 mL/min.

10.2.15 Continue to transfer the water from the beaker to the funnel until the entire contents of the baker are emptied into the filter funnel.

10.2.16 Add approximately 25 mL of clean deionized water to rinse the beaker and pour this water into the filter funnel as well. Ensure that the filter funnel remains filled with solution from the beaker until the filtration is complete.

10.2.17 Remove the funnel and carefully transfer the filter onto a clean SEM specimen stub, using fine-point duckbill tweezers.

10.2.18 Allow the filter to air dry in an ISO Class 5 (Fed. Std. 209 Class 100) or cleaner environment.

10.2.19 Affix the perimeter of the filter to the specimen stub by applying several (at least four) spots of conductive carbon paint.

⁸ For example, see the assembly diagram in <http://www.millipore.com/catalogue.nsf/docs/C804>. Permission to reference this copyrighted image is provided as a courtesy by the Millipore Corporation.

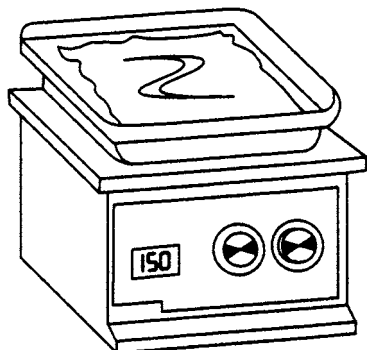


FIG. 1 Orbital Shaker

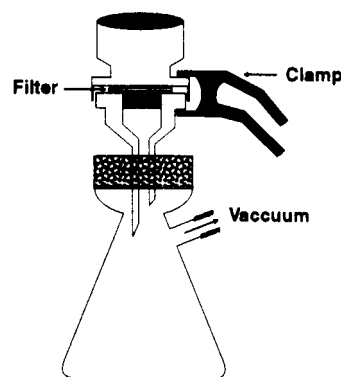


FIG. 2 Filtration Assembly

10.2.20 Transfer the stub to the vacuum sputtering unit and apply a gold coating to the filter. Typically, 20 s – 40 s sputter time will provide adequate gold coverage, depending on the equipment used.⁹

10.2.21 Label the sample as the background count for this particular experiment and place it in a clean, covered container. Set it aside for subsequent particle enumeration. The counting procedures are described in 10.4 and 10.5. If there is concern that the background may exhibit excessive contamination, then the operator may wish to reverse the order of counting described in 10.5.14, that is, count the background first and delay preparation of the sample stub (10.3) until there is confidence that there are no contamination issues in the set up.

10.2.22 After preparing the blank, the wiper sample is prepared using the same glassware and filtration system.

10.2.23 Accurate counts in the test wiper sample require subtracting background counts from the sample counts. The value of the background count should be less than 15 % of the sample count. If this is not the case, reclean the apparatus and perform the experiment again. For very clean wipers which may exhibit very low counts in the >100 μm range, this requirement may be lifted.

10.3 Preparation of Sample Stub:

10.3.1 Place 500 mL of deionized water into the same photographic tray that was used in preparation of the background sample.

10.3.2 Place the tray on the platform of the orbital shaker (Fig. 1).

10.3.3 Add a 25-mL aliquot from the stock surfactant cleaning solution (see 10.2.2 and 10.2.3) to the water in the tray.

10.3.4 Shake the tray for 1 min to facilitate the mixing of surfactant and water.

10.3.5 Using cleanroom gloves, open the bag of wipers to be tested.

10.3.6 Using two pairs of clean forceps, carefully lift a wiper from the bag and gently drape the wiper onto the surface of the water in the tray.

10.3.7 Run the shaker at 150 r/min for 5 min.

10.3.8 Using the forceps, lift the wiper from the tray slowly by holding two adjacent corners, allowing the excess water to drip into the tray.

10.3.9 Measure the dimensions of the wiper to two significant figures and set the wiper aside.

10.3.10 Pour the water from the tray into the beaker previously used for the background sample preparation.

10.3.11 Add approximately 25 mL of clean deionized water to rinse the tray and pour this water into the beaker as well.

10.3.12 Complete the sample preparation for the test specimen by repeating 10.2.7-10.2.21, using a new membrane filter from the same package. Some wipers may have excessive numbers of particles that can overload the filter, making it impossible to obtain accurate counts. In these cases, one samples a representative portion of the water of the beaker and

filters only that portion. As an example, if 25 mL of the total 550 mL were sampled for filtration, this would represent only 25/550th of the available particles. The actual particle count on the wiper would be calculated by multiplying the particles counted in the representative portion by 550/25, then subtracting the blank value.

10.3.13 Label the sample as the test specimen for the particular experiment.

10.4 Manual counting of >100- μm fibers and particles.

10.4.1 Place the wiper sample stub in the specimen-holding mount plate and then place the mount plate on the x-y stage of the optical microscope (see Fig. 3).

10.4.2 Set the microscope at the lowest magnification and its circular iris dial in the middle of its range.

10.4.3 Turn on the illuminator and adjust the knobs to have adequate and uniform illumination on the stub.

10.4.3.1 To obtain the uniformity, set one of the arms of the light guide so the light grazes the surface of the membrane filter (approximately 15 to 30° angle between the light beam and the surface of the filter).

10.4.3.2 Set the other arm from the other side of the filter, again with the light grazing the filter surface.

10.4.3.3 The illuminance can be varied by adjusting the iris dial and by slightly adjusting the knobs of the illuminator back and forth.

10.4.4 Bring the particles/fibers on the filter surface to focus by adjusting the focus knob while observing the field through the microscope eyepieces.

10.5 Viewing Fields From the Optical Microscope:

10.5.1 To make the counting process more convenient, the images from the sample stub can be viewed in the video monitor using a video camera and a computer with frame-grabbing hardware and software. Connect the R, G, B leads from the computer to the corresponding R, G, B leads from the video camera, allowing the image to be displayed on the video monitor.

10.5.2 Focus the microscope so that the particles and fibers on the filter are seen brightly and clearly in the monitor. Readjust the arms of the light guide to make the distribution of light on the filter as uniform as possible, as viewed in the monitor. Direct the light to the filter stub at a grazing angle of approximately 15 to 30° between the light beam and the surface of the filter.

10.5.3 Change the magnification to 40 $\times$ and readjust the light so that the field is completely illuminated and refocus the

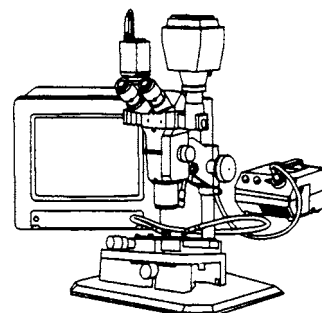


FIG. 3 Optical Microscope

⁹ The application of a vacuum in the sputtering unit will not remove or disturb particles on the surface of the filter. Recovery studies are documented in Footnote 15.

microscope. The purpose of focusing at high magnification is to ensure accurate viewing at the lower magnifications.

10.5.4 Change the magnification to 20× and readjust the lighting if necessary for the best viewing maintaining a grazing angle of approximately 15 to 30° between the light beam and the surface of the filter.

10.5.5 Scan the entire surface of the filter by moving it in the x and y directions and check for the uniformity of distribution of fibers and particles throughout the filter. Discard the sample and prepare a new sample if the inspection discloses a nonuniform distribution of particles on the filter.

10.5.6 If the distribution of fibers and particles looks uniform and random, readjust x and y to position the filter to the lower left-most viewing field.

10.5.7 Starting at the lower left-most field, scan the filter by moving the stage horizontally along the x axis from left to right.

10.5.8 While scanning, manually count all the large particles and fibers (>100 μm) as seen along the scanning path. For the equipment described any fiber or particle whose largest dimension on any axis is 8 mm or greater at 20× magnification as viewed in the monitor, is actually 100 μm and greater in size and should be counted. The conversion factors are equipment dependent and users of this test method shall establish the relation between screen size and object size.

10.5.9 Record the counts by indexing the tally counter each time large particles and fibers are seen on the monitor screen.

10.5.10 After each lateral scan, move the filter vertically along the y axis until a new area of the filter comes into view.

10.5.11 Perform the counting as the filter is moved laterally, this time from right to left.

10.5.12 Continue vertical and lateral movements until the filter is completely scanned and all the particles and fibers that are 100 μm and larger on the filter are counted.

10.5.13 Record the total count in the data sheet as N (see Appendix X2 for example).

10.5.14 Replace the sample stub with the background stub and count all the particles and fibers that are 100 μm and larger by following the same procedure as previously described and record the total count in the data sheet as N_{blank} .

10.5.15 Subtract the blank average N_{blank} from the sample average N to obtain the corrected counts of particles and fibers that are 100 μm and larger in the wiper sample.

10.5.16 Denote the difference as F .

$$F = N - N_{\text{blank}} \quad (1)$$

10.5.17 Divide F by the area of the wiper in square metres and perform the calculations for the total number of 100-μm and larger particles and fibers per square metre of the wiper material as described in Section 11.

10.6 SEM Counting Procedure at 200×:

10.6.1 After counting particles and fibers 100 μm and larger using the optical microscope, proceed to count the rest of the particles on the same filter using the SEM. Particles in two different size categories are counted using the SEM at two different magnifications, 200 and 3000×. Counts at 200× include all particles between 5 and 100 μm; counts at 3000×

include smaller particles ranging from 0.5 to 5 μm. This test method includes the use of computerized image analysis and counting techniques.

10.6.2 It is assumed that the operator is well-versed in the operational procedures of the SEM. It is advisable to be familiarized with the filtering, viewing, and particle enumeration techniques by running simulation experiments to measure known quantities of submicrometre to 5-μm polystyrene latex microspheres.

10.6.3 Transfer the sample stub used in the optical microscope to the SEM sample holder.

10.6.4 Slide the holder inside the sample chamber of the SEM.

10.6.5 Evacuate the chamber, turn on the filament, and prepare the SEM for viewing at 200×. For proper viewing of a sample in the SEM and for making the field appropriate for computer-assisted counting, adjust the magnification, focus, contrast and brightness, and tilt angle.

10.6.6 Focus the field initially at 5000× and then reduce the magnification to 200×. The purpose of focusing at a higher magnification than that which will be used is to bring extreme clarity to the image of the particles on the filter, so the computer software can unambiguously recognize and accurately categorize particles by number and size.

10.6.7 Inspect the filter by manually scanning the entire surface at 200× magnification for the uniformity of distribution of particles. If the inspection discloses a nonuniform distribution of particles on the filter, the sample should be discarded and a new sample should be prepared for an accurate counting of all particles and fibers.

10.6.8 A visual field is defined as the total area seen on the SEM video display monitor. Since it is very time-consuming to count all the particles present on a filter surface, a statistical sampling of random locations covering the entire filter area is used for this test method. If the SEM has a computer-driven automated stage, the preselected counting locations can be stored in the SEM computer. The locations can then be accessed automatically for counting the particles present in those fields.

10.6.9 For this test method, preselecting 16 such visual fields (Fig. 4) for counting at 200× and 32 fields (Fig. 5) for

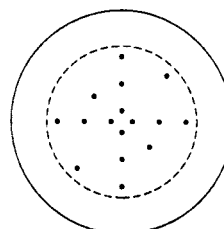


FIG. 4 Layout of 16 Preselected Points for Counting at 200×

counting at 3000× will be sufficient to ensure statistical validity, assuming a goal of ±10 % accuracy at a 95 % confidence level (see Appendix X1 for the statistical analysis). The locations for the counts are selected to cover the central area of the filter, the area at approximately half of the radius of

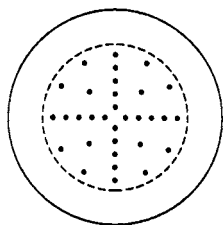


FIG. 5 Layout of 32 Preselected Points for Counting at 3000×

the effective filter area, and the area proximal to the perimeter but not touching the edge of the effective filter area.

10.6.10 Identify the 16 fields that will be counted in this procedure in accordance with the example in Fig. 4.

10.6.11 For the computer-assisted counting of the number of particles in each of the 16 preselected fields at a magnification of 200×, follow the procedure as outlined in 10.6.12-10.6.18.

10.6.12 Move the stage to one of the preselected fields on the filter.

10.6.13 Set conditions for the automatic measurements in the computer. Restrict particle sizes to 5 through 100 μm in the image analysis software.¹⁰ The sizing algorithm should be verified experimentally with known calibration samples (see Section 9).

10.6.14 Focus the field at 5000× and reduce the magnification back to 200×.

10.6.15 Adjust brightness and contrast in the SEM until proper illumination is achieved. The illumination parameter setting is specific for individual particle counting software and can be predetermined through experimentation with known amounts of standard-sized particles such as polystyrene microspheres (see Section 11).¹¹

10.6.16 Obtain a computer count the particles and record the computer count of the 5- to 100-μm particles in this field under *W* in the data sheet (see Appendix X2 for example).

10.6.17 Move to a new field and repeat 10.6.14-10.6.16.

10.6.18 For all subsequent fields repeat 10.6.17.

10.6.19 Total the 16 counts, calculate the average, W_{av} , and record this number in the data sheet.

10.6.20 Replace the test specimen in the SEM with the background specimen and complete the counting of particles at the same coordinates by repeating the procedure previously outlined for the test sample.

10.6.21 Total the 16 counts, calculate the average, $W_{av}^{(blank)}$, and record this number in the data sheet.

10.6.22 Subtract the blank average $W_{av}^{(blank)}$ from the sample average W_{av} to obtain the corrected count of 5- to 100-μm particles per field. Denote the difference as *T*.

$$T = W_{av} - W_{av}^{(blank)} \quad (2)$$

10.6.23 Perform the calculations for the total number of 5- to 100-μm particles per square metre of the wiper material as described in Section 11.

10.7 SEM Counting Procedure at 3000×

10.7.1 Count the smaller particles (0.5 to 5 μm) on the same test specimen filter using the SEM at a higher magnification of 3000×. In this procedure, when particle counts are low (for example, less than 25 particles per field), counting can be done manually. However, the computer-assisted automatic counting procedure, similar to that used at the 200× study, should be utilized for samples having more than 25 particles per field.

10.8 SEM Manual Counting Procedure at 3000× (for samples with less than 25 particles per field):

10.8.1 Use the test specimen stub already in the SEM.

10.8.2 Manually count the number of particles in the test sample in each of the 32 preselected fields (Fig. 5) at a magnification of 3000× and record the results in the data sheet under *P* (see Appendix X2 for example).

10.8.3 Total the 32 counts, calculate the average, P_{av} , and record this number in the data sheet.

10.8.4 Replace the test specimen in the SEM with the background specimen stub and complete the counting of particles at the same coordinates by repeating the procedure previously outlined for the test sample.

10.8.5 Total the 32 counts, calculate the average, $P_{av}^{(blank)}$, and record this number in the data sheet.

10.8.6 Subtract the blank average $P_{av}^{(blank)}$ from the sample average P_{av} to obtain the corrected count of 0.5- to 5-μm particles per field. Denote the difference as *V*.

$$V = P_{av} - P_{av}^{(blank)} \quad (3)$$

10.8.7 Perform the calculations for the total number of 0.5- to 5-μm particles per square metre of the wiper material as described in Section 11.

10.9 SEM Computer-Assisted Counting Procedure at 3000× (for samples with more than 25 particles per field):

10.9.1 For the computer-assisted counting at 3000×, follow the procedure as follows:

10.9.2 Move the stage to one of the 32 preselected fields on the filter (see Fig. 5).

10.9.3 Set conditions for the automatic measurements in the computer. Restrict particle sizes to 0.5 through 5 μm in the image analysis software. The sizing algorithm should be verified experimentally with known calibration samples (see Section 9).¹²

10.9.4 Focus the field at 5000× and reduce the magnification back to 3000×.

10.9.5 Adjust brightness and contrast in the SEM until proper illumination is achieved. The illumination parameter setting is specific for individual particle counting software and can be predetermined through experimentation with known amounts of standard-sized particles such as polystyrene microspheres (see Section 9).¹³

10.9.6 Obtain a computer count the particles and record the 0.5 to 5-μm particles in this field as *P* in the data sheet (see Appendix X2 for example).

10.9.7 Move to a new field and repeat 10.9.4-10.9.6.

¹⁰ In the Image-Pro Plus 3.0 Program, presetting the area values for Start = 10 and End = 250 will select particles between 5 and 100 μm at 200×.

¹¹ In the Image-Pro Plus 3.0 Program, the appropriate illumination setting or density range for 200× is from 8.0 to 8.5.

¹² In the Image-Pro Plus 3.0 Program, presetting the area values for Start = 5 and End = 250 will select particles between 0.5 and 5 μm at 3000×.

¹³ In the Image-Pro Plus 3.0 Program, the appropriate illumination setting or density range for 3000× is from 0.1 to 1.0.

10.9.8 For all subsequent fields repeat 10.9.7.

10.9.9 Total the 32 counts, calculate the average, P_{av} , and record this number in the data sheet.

10.9.10 Replace the test specimen in the SEM with the background specimen stub and complete the counting of particles at the same coordinates by repeating the procedure previously outlined for the test specimen.

10.9.11 Total the 32 counts, calculate the average, $P_{av(blank)}$, and record this number in the data sheet.

10.9.12 Subtract the blank average $P_{av(blank)}$ from the sample average P_{av} to obtain the corrected count of 0.5- to 5- μm particles per field. Denote the difference as V .

$$V = P_{av} - P_{av(blank)} \quad (4)$$

10.9.13 Perform the calculations for the total number of 0.5- to 5- μm particles per square metre of the wiper material as described in Section 11.

11. Calculation

11.1 Sample Calculations:

11.1.1 Sample Calculation at 20 $\times$ —Particles and Fibers >100 μm

11.1.1.1 Reference 10.5.13.

Total number of wiper particles and fibers, N , counted on the entire effective filter area = 253

11.1.1.2 Reference 10.5.14.

Total number of particles and fibers, N_{blank} , counted on the background filter = 7

11.1.1.3 Reference 10.5.15. Calculate the total number of blank-corrected particles and fibers:

$$F = 253 - 7 = 246 \quad (7)$$

where:

F = total number of particles and fibers >100 μm contributed by the wiper.

11.1.1.4 Reference 10.5.17. Normalize the particle and fiber count to the wiper area in square metres. This is done by dividing F by the wiper area.

$$\text{Wiper area} = 0.23 \times 0.23 \text{ m} = 0.0529 \text{ m}^2 \quad (8)$$

$$\begin{aligned} \text{Particles/fibers/m}^2 \text{ of wiper} &= F/\text{wiper area} \\ &= 246/0.0529 = 4650 \text{ particles and fibers/m}^2 \end{aligned} \quad (9)$$

Report this number to 2 significant digits as 4700 particles/fibers > 100 $\mu\text{m}/\text{m}^2$.

11.1.2 Sample Calculation at 200 $\times$ —Particles 5 to 100 μm :

11.1.2.1 Reference 10.6.19.

$$\text{Fields counted} = 16 \quad (10)$$

$$\text{Total wiper particles counted in 16 fields} = 536 \quad (11)$$

$$\text{Average particles per field, } W_{av} = 536/16 = 33.5 \quad (12)$$

11.1.2.2 Reference 10.6.21.

$$\text{Fields counted} = 16 \quad (13)$$

$$\text{Total blank particles counted in 16 fields} = 42 \quad (14)$$

$$\text{Average blank particles per field, } W_{av(blank)} = 42/16 = 2.63 \quad (15)$$

11.1.2.3 Reference 10.6.22. Average blank-corrected particles per field:

$$T = 33.5 - 2.63 = 30.87 \quad (16)$$

11.1.2.4 Calculate the total effective filter area:

$$\text{Diameter of active filter} = 19 \text{ mm} \quad (17)$$

$$\begin{aligned} \text{Total effective filter area} &= \pi \times (19/2)^2 \text{ mm}^2 \\ &= 284 \times 10^6 \mu\text{m}^2 = 2.84 \times 10^8 \mu\text{m}^2 \end{aligned} \quad (18)$$

11.1.2.5 Calculate the area of a single field as viewed through the SEM. It is necessary to know the area in square micrometres that a single field of view represents. For this example, assume a linear dimension of 1.73 mm represents 5 μm as viewed on the SEM monitor screen at 200 $\times$ magnification (this can be determined using the SEM micrometre bar measurement tool). Also, assume the monitor measures 237.5 by 174.5 mm. This corresponds to an area of a single field of view of $(237.5 \times 5/1.73) \times (174.5 \times 5/1.73) = 346\,184 \mu\text{m}^2$. The field of view is equipment dependent and users of this test method shall calculate the field of view for their own equipment.

11.1.2.6 Calculate the number of fields, N_1 , that can be viewed in the effective filter area. Divide the effective filter area by the area of a single field:

$$N_1 = (2.84 \times 10^8 \mu\text{m}^2)/(346\,184 \mu\text{m}^2) = 820 \text{ fields} \quad (19)$$

11.1.2.7 Calculate the total number of particles, S , on the filter. Multiply the average blank-corrected particles per field, T , by the total number of fields on the filter, N_1 :

$$S = T \times N_1 = 30.87 \times 820 = 25\,313 \quad (20)$$

where:

S = total number of 5- to 100- μm particles contributed by the wiper.

11.1.2.8 Normalize the particle count to the wiper area in square metres. Divide S by the wiper area:

$$\text{Wiper area} = 0.23 \times 0.23 \text{ m} = 0.0529 \text{ m}^2 \quad (21)$$

$$\begin{aligned} \text{Particles per m}^2 \text{ of wiper} &= S/\text{wiper area} \\ &= 25\,313/0.0529 = 478\,507 \text{ particles/m}^2 \end{aligned} \quad (22)$$

Report this number to two significant digits as 4.8×10^5 particles/m².

11.1.3 Sample Calculation at 3000 $\times$ —Particles 0.5 to 5 μm :

11.1.3.1 Reference 10.9.9.

$$\text{Fields counted} = 32 \quad (23)$$

$$\text{Total wiper particles counted in 32 fields} = 380 \quad (24)$$

$$\text{Average particles per field, } P_{av} = 380/32 = 11.88 \quad (25)$$

11.1.3.2 Reference 10.9.11.

$$\text{Fields counted} = 32 \quad (26)$$

$$\text{Total blank particles counted in 32 fields} = 46 \quad (27)$$

Average blank particles per field, $P_{av(blank)} = 46/32 = 1.44$ (28)

11.1.3.3 Reference 10.9.12. Calculate the average blank-corrected particles per field:

$$V = 11.88 - 1.44 = 10.44 \quad (29)$$

11.1.3.4 Calculate the total effective filter area:

$$\text{Diameter of active filter} = 19 \text{ mm} \quad (30)$$

$$\begin{aligned} \text{Total effective filter area} &= \pi \times (19/2)^2 \text{ mm}^2 \\ &= 2.84 \times 10^8 \mu\text{m}^2 \end{aligned} \quad (31)$$

11.1.3.5 Calculate the area of a single field as viewed through the SEM. It is necessary to know the area in square micrometres that a single field of view represents. For this example, assume a linear dimension of 26 mm represents 5 μm as viewed on the SEM monitor screen at 3000 $\times$ magnification (this can be determined using the SEM micrometre bar measurement tool). Also, assume the monitor measures 237.5 by 174.5 mm. This corresponds to an area of a single field of view of $(237.5 \times 5/26) \times (174.5 \times 5/26) = 1533 \mu\text{m}^2$.

11.1.3.6 Calculate the number of fields, N_1 , that can be viewed in the effective filter area. Divide the effective filter area by the area of a single field:

$$N_1 = 2.84 \times 10^8 \mu\text{m}^2 / 1533 \mu\text{m}^2 = 185\,258 \text{ fields} \quad (32)$$

11.1.3.7 Calculate the total number of particles, S , on the filter. Multiply the average blank-corrected particles per field, V , by the total number of fields on the filter, N_1 :

$$S = V \times N = 10.44 \times 185\,258 = 1\,934\,094 \text{ particles} \quad (33)$$

where:

S = total number of 0.5- to 5- μm particles contributed by the wiper.

11.1.3.8 Normalize the particle count to the wiper area in square metres. Divide S by the wiper area:

$$\text{Wiper area} = 0.23 \times 0.23 \text{ m} = 0.0529 \text{ m}^2 \quad (34)$$

$$\begin{aligned} \text{Particles/m}^2 \text{ of wiper} &= S/\text{wiper area} = \\ &1\,934\,094/0.0529 = 36.56 \times 10^6 \text{ particles/m}^2 \end{aligned} \quad (35)$$

Report this number to two significant digits as 37×10^6 particles/ m^2 .

12. Report

12.1 Report the following information:

12.1.1 General information including the following: ASTM test method number, date and time of test, sample identification, author of report, and other personnel involved in testing.

12.1.2 Sample preparation information including the following: filter type and diameter, filter pore size, effective filter area, description of test liquid used (low-surface-tension cleaning solution), total volume of liquid filtered, any deviation from standard procedure as written.

12.1.3 Experimental setup of instrumentation including the following: filter coating instrument type, instrument parameters (time and power setting), and type of coating. Optical microscope type, SEM type, SEM voltage, stage tilt angle, settings used in image analysis software, and any deviation from the standard procedure as written.

12.1.4 *Results Including the Following*—Description of tested wiper (material, construction, size), calculations for 20, 200, and 3000 $\times$ measurements (see Section 11), indication whether counting was computer-assisted or manual for each magnification, any unusual observations based on particle/fiber morphology, particle identification (if possible through morphology or EDX), statistical analysis (mean, standard deviation, CV) if multiple wipers of a single type are tested, other comments based on operator observations of experimental conditions or results.

13. Precision and Bias

13.1 Because less than 10 000 particles or fibers will actually be counted, the final result will always have a statistical uncertainty of at least 1 %. In performing the calculations, it is recommended that the rounding error be kept to 0.1 % or less, which means using numbers with four or more significant digits, where available.

13.2 In reporting the final result, it is recommended that rounding be done to two significant digits if the first digit of the number is 3 or greater, and three significant digits if the first digit is less than 3 to avoid indicating precision that is not present. Thus, 0.306 becomes 0.31 but 0.2986 becomes 0.299. This will keep rounding error to less than 0.5/30 or 1.7 %.

13.3 *Precision*—A statistically valid procedure for counting is described in this test method. The accuracy and precision of the resultant count can likewise be measured statistically (see Appendix X1).

13.4 *Bias*—No justifiable statement can be made on the bias of this test method for the particle counting of the wipers since the true value of the property (except counting of standard known amounts of polystyrene microspheres) cannot be established by an accepted referee test method.

14. Keywords

14.1 cleanroom wipers; contamination control; membrane filter; optical microscopy; particle counting and sizing; particles and fibers; scanning electron microscopy

APPENDIXES

(Nonmandatory Information)

X1. STATISTICAL REQUIREMENTS FOR SEM PARTICLE COUNTING FOR WIPER SUBSTRATES

X1.1 The statistical accuracy goal for this test method is an accuracy level within $\pm 10\%$ of the true population mean when described at a confidence level of 95 %. Classical statistical theory can be used to define standard deviation, standard error, confidence intervals, and accuracy.

X1.2 The following is a demonstration of how to use statistics to determine the confidence interval and accuracy level for a particular set of data:

Standard Deviation: $s.d. = E(X - \bar{X})^2_{n-1}$ Standard Error: $s.e. = \frac{(s.d.)}{\sqrt{n}}$

Confidence Interval: $\bar{X} - z(\alpha_{12} \times (s.e.)) < \mu < \bar{X} + z(\alpha_{12} \times (s.e.))$

$$\text{Accuracy: } \pm \frac{z(\alpha_{12} \times (s.e.))}{\bar{X}} \times 100$$

where: X = single experimental measured value (particles/field),
 $n \bar{X}$ = arithmetic mean of all measured values (particles/field),
 n = number of fields counted,
 $z\alpha_{12}$ = standardized normal deviate, and
 μ = true population average.

Example: Number of fields counted: $n = 32$
 Arithmetic mean of particles/field: $\bar{X} = 10.19$
 Sample standard deviation: $s.d. = 2.36$
 Standard error of mean: $s.e. = 0.42$
 Standardized normal deviate
 for $(1-\alpha)$ confidence interval: $z\alpha_{12} = 1.96$

95 % confidence level = $10.19 \pm 1.96 \times 0.42 = 10.19 \pm 0.82$
 Hence, μ is estimated to be between 9.37 and 11.01

$$\text{and accuracy} = \pm \frac{1.96 \times 0.42}{10.19} \times 100 = \pm 8.08 \%$$

Please note that counting 32 fields covers approximately 0.02 % of the area of the entire filter. The calculation is as follows:

$$\text{Total fields} = \frac{\text{Area of active filter}}{\text{Field area in SEM at } 3000\times} = \frac{238 \times 10^6}{1533} = 155\,251$$

$$\text{Hence, fraction of field counted: } \frac{32}{155\,251} \times 100 = 0.021 \%$$

X1.3 There is precedence in the literature^{14, 15, 16, 17} which claims that enumeration of as low as 0.01 % of the total fields by SEM can be sufficient to obtain statistically acceptable particle count.

¹⁴ Bhattacharjee, H. R., et al, "The Use of Scanning Electron Microscopy to Quantify the Burden of Particles Released from Cleanroom Wiping Materials," *Scanning*, Vol 15, 1993, pp. 301-308.

¹⁵ Mayette, D. C., et al, "A Reconsideration of the Scanning Electron Microscope as a Particle Enumerating Tool in UPW," *ICCCS Proceedings*, 1992, pp. 51-57.

¹⁶ Bhattacharjee, H., and Paley, S., "Evaluating Sample Preparation Techniques for Cleanroom Wiper Testing," *MICRO*, February 1997, pp. 39-45.

¹⁷ Bhattacharjee, H. R., and Paley, S. J., "Comprehensive Particle and Fiber Testing for Cleanroom Wipers," *Journal of the IEST*, Vol 41, No. 6, November/December 1998, pp. 19-25.

X2. PARTICLE/FIBER COUNTING DATA SHEET



Date: _____ Wiper ID: _____

Operator: _____ Lot #: _____

O.M. at 20X		SEM at 200X			SEM at 3,000X					
N Total Fibers (≥ 100µ)		W Particles/Fibers (5µ–100µ)			P Particles (0.5µ–5µ)			P Particles (0.5µ–5µ)		
File #		File #			File #			File #		
Sample N	Background N _{back}	Field #	Sample W	Background W _{back}	Field #	Sample P	Background P _{back}	Field #	Sample P	Background P _{back}
		1.			1.			17.		
		2.			2.			18.		
		3.			3.			19.		
		4.			4.			20.		
		5.			5.			21.		
		6.			6.			22.		
		7.			7.			23.		
		8.			8.			24.		
		9.			9.			25.		
		10.			10.			26.		
		11.			11.			27.		
		12.			12.			28.		
		13.			13.			29.		
		14.			14.			30.		
		15.			15.			31.		
		16.			16.			32.		
		Total (15 field)		Total (15 field)				Total (32 field)		Total (32 field)
		W _{av}		W _{av,back}				P _{av}		P _{av,back}
		T = W _{av} – W _{av,back}						V = P _{av} – P _{av,back}		

F = N – N_{back}

T = W_{av} – W_{av,back}

V = P_{av} – P_{av,back}

Comments (if any):

FIG. X2.1 Particle/Fiber Counting Data Sheet

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