

**INSTRUCTIONAL MANUAL
CAT. 65212-10**

**Via-Cell® Bioaerosol
Sampling Cassette**



Electron Microscopy Sciences

P.O. Box 550 • 1560 Industry Road • Hatfield PA 19440

Toll Free: 1-800-523-5874

www.emsdiasum.com

sgkcck@aol.com

Tel: 215-412-8400 • Fax: 215-412-8450

Introduction

Thank you for your purchase of the Via-Cell® Bioaerosol Sampling Cassette! This sampler is unique in that its design is suitable for collecting both viable and non-viable bioaerosols. In addition to its many applications, features, and benefits, the Via-Cell® Bioaerosol Sampling Cassette collects particles, maintaining the viability of spores during transport to the laboratory or any other site in which your application is conducted.

Applications

- Cleaning rooms and monitoring
- Flood restoration
- Indoor air quality ... and more!

Benefits and Other Advantages

- Compatible with multiple kinds of sampling pumps
- Sterile and pre-loaded
- Small, compact, and convenient size
- Easily disposable, eliminating cross-contamination issues
- No required refrigeration for samples – transportation/storage at room temperature is sufficient
- Comes with a sturdy, tamperproof seal with a unique serial number
- Multiple analyses can be performed from a single sample
- Shelf life of 1 year from the date of manufacture (which is four times longer than most other sampling methods using culture plates)

Options for Analysis

Direct Microscopy – Samples can be prescreened with direct microscopy, allowing for user to decide on suitability of further analysis techniques.

Culturable Sampling – Collect viable organisms for culturing on agar media plates.

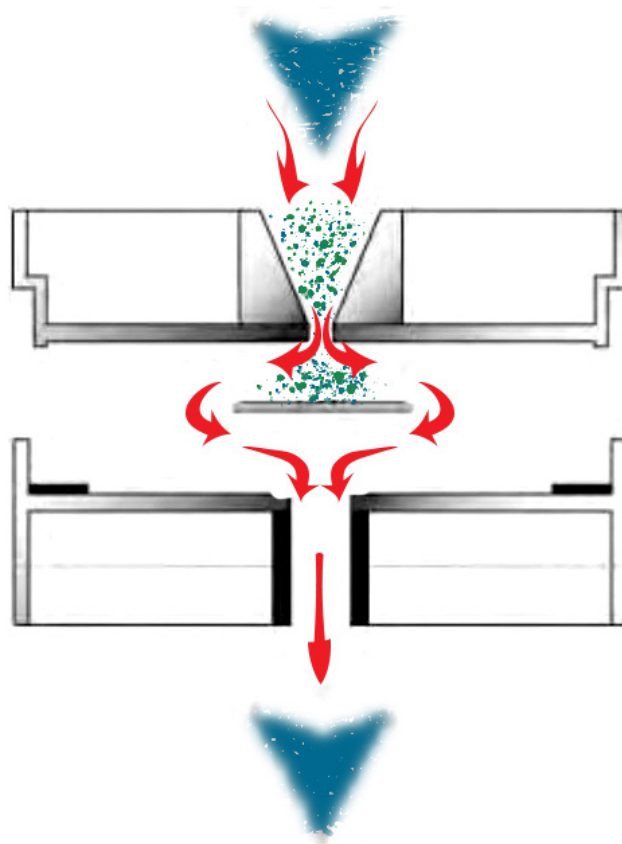
PCR/Chemical Analysis – Since Via-Cell has a water soluble collection media, it is perfect for PCR and other types of chemical analysis techniques.

Operation Principles – Inertial Impaction

Illustration of the Air Flow Path through the Via-Cell® Sampling Cassette

The Via-Cell® Sampling Cassette works as follows:

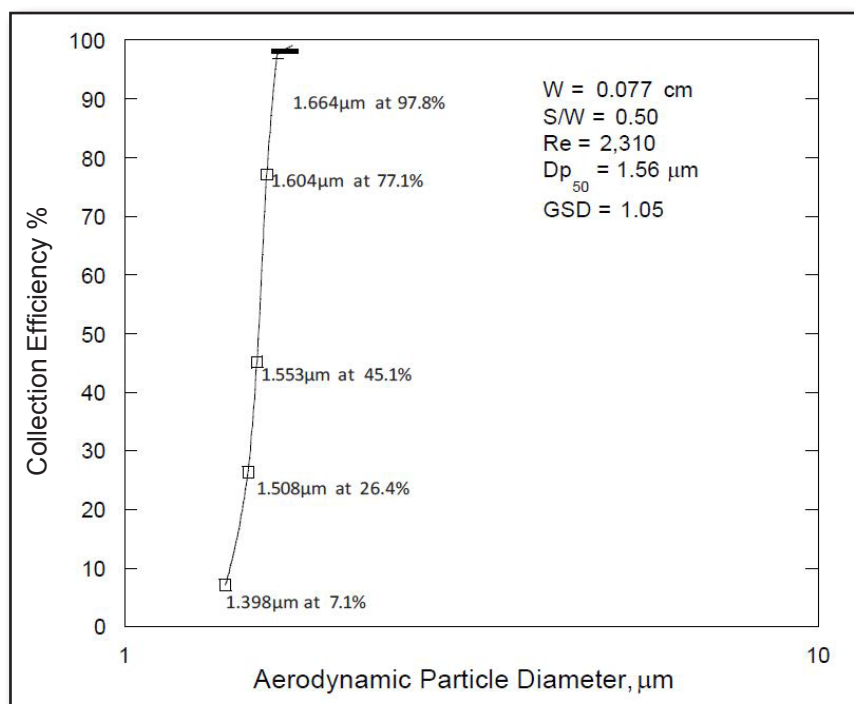
1. Using inertial impaction, the particles in the air stream are accelerated as they approach the tapered inlet opening and are drawn through a small slit aimed directly at a glass slide containing a water soluble sampling media. The sampling media holds the particles intact and maintains viability for later analysis.
2. As the particles come through the slit, the air velocity forces the particles to impact into the sampling media.
3. Then, the air stream makes a sharp 90° turn and proceeds around the slide and out of the cassette.



Operational Performance

The Via-Cell sampler has been evaluated in commercial and university laboratories to determine the collection efficiency and operational aspects of the collector. With impactor-type samplers, the most common measure of efficiency is defined by its “cut-point” curve. The cut-point (also known as the “D-50” point) is defined by the particle size at which 50% of the particles are collected and 50% pass through the sampler at a pre-determined flow rate. The ‘cut point’ for the Via-Cell Sampler has been determined to be 1.56 microns at a flow rate of 15 LPM. This is extremely useful when studying for fungal analysis, where particles smaller than this are not of interest, and if included in the sampling, can even create enough background debris to obscure appropriate viewing of the sample, thus compromising reliable analysis.

The graph (to the right) displays the collection efficiency of the Via-Cell sampler when used at a flow rate of 15 LPM. Properly designed samplers will exhibit upper and lower curve limits that are very sharp. A sampler will reach 100% efficiency almost immediately after the cut-point, in the ideal scenario.



Suggested Field Sampling Procedures

The Via-Cell sampler is designed to efficiently collect bio-aerosol particles while also providing safe transport to the laboratory. The results achieved when using the sampler requires proper use, including pump calibration and handling techniques. Therefore familiarization with the Via-Cell Sampler collection procedure is recommended prior to use .

Required Sampling Equipment

- Via-Cell Sampling Cassettes
- Sampling pump capable of pulling 15LPM
- Tubing to attach cassette to pump, if required (1/4" ID or 1/2" ID)
- Pump calibration device

Handling the Via-Cell Sampler

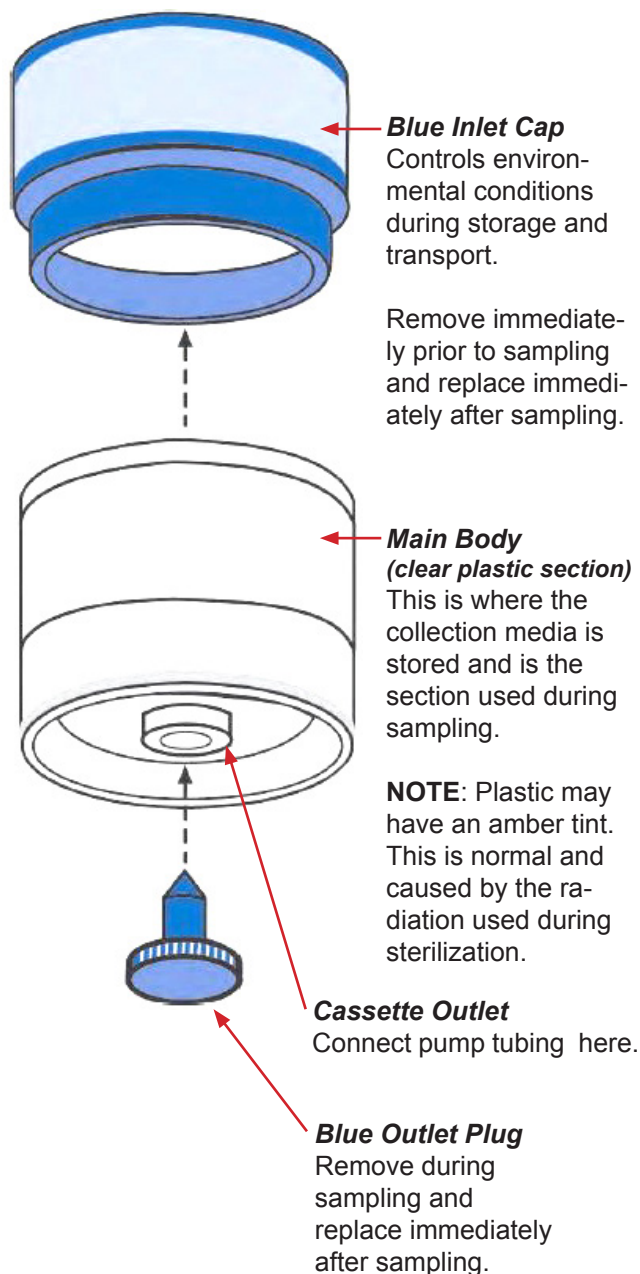
- The Via-Cell Sampler is sterile and ready-to-use when received.
- DO NOT contaminate the inner-surface of the cassette through careless handling
- DO NOT insert anything into the cassette through the inlet or outlet port.
- Ensure that only the sample is collected. **NOTE:** The act of collection may render the sampler unsterile due to the collected particles. Inadvertently extraneous material added to the cassette could alter the outcome.

Handling Tips and Considerations

- ALWAYS store Via-Cell cassettes between 50 - 80°F.
- The Via-Cell sampler does not require refrigeration during storage or during shipment to the analysis laboratory.
- Prolonged exposure to high temperatures (such as in a hot warehouse or in a car) may alter the performance of the product .
- Extremes in environmental conditions (i.e. very hot or cold weather) may affect the collection performance.
- Ensure that the Via-Cell samples laboratory receives the samples within 24 hours after collection. The laboratory should process the samples within 24 hours of receipt.



Do not open the blue inlet cap assembly! The materials used inside may be irritating to your skin.



Sample Preparation – Calibration

The sampling pump needs to be calibrated at a flow rate of 15 liters per minute to ensure proper collection. To ensure accuracy, calibrate at least once a day. Ideally, calibration should be done prior to and after each collection. It is strongly recommended that a primary standard device such as a soap bubble meter/tube or other NIST certifiable device be used to calibrate the sampling pump. Secondary calibration devices such as a rotameter may be used if it is calibrated against a primary standard.

NOTE: Some pumps will only work with specific calibration devices, so please read the manual for the pump that will be used to ensure it is compatible with the Via Cell sampler.

A few examples of typical devices that can be used for calibration:

- Zefon Bio-Pump®Plus (Catalog # 65211-12)
- mini BUCK Primary Flow Calibrator (Catalog # 65240-63 or # 65240-64)

Sample Collection

1. Calibrate the sampling pump to a flow rate of 15 lpm.
2. Clean hands or wear clean, disposable, powder free gloves.
3. Tear open the foil package using the tear strip on top. **NOTE:** Use care when opening as this package is resealable and is required to be used after sampling for transport to the laboratory.
4. Remove the Via-Cell from the package.
5. Remove the blue inlet cap and record the Via-Cell serial number, and any other pertinent information on the front of the foil package. **NOTE:** The serial number can be viewed through the top of the cassette once the blue inlet cap is removed. It is printed directly on the collection slide. *See illustration at right.*
6. After recording the serial number, replace the blue inlet cap until ready to sample.
7. Remove the blue outlet plug from the cassette and place the plug into the foil pouch for safe keeping.



About the Foil Packaging

Keep the resealable foil package for transportation of the cassette to the laboratory. This package, along with the blue inlet and outlet caps, controls the environmental conditions inside the cassette after sample collection. This is critical in maintaining the viability of the particles if culture analysis is required. The included safety seal label is used to ensure the collected sample remains uncontaminated until it reaches the laboratory.



The interior of the Via-Cell sampler is sterile. When the plugs are removed, handle the product very carefully so that the sampling port and outlet port do not become contaminated with materials that may alter the collection or analysis of the sample.

8. Connect the Via-Cell sampler to the pump; position the cassette in the desired location. **NOTE:** The Via-Cell sampler is capable of operating in any vertical or horizontal orientation and in confined spaces such as ducts, plenums or wall cavities.
9. Remove the large blue inlet cap from the cassette and place into the foil package for safekeeping.
10. Turn on the pump and run for the desired amount of time. Reference the chart on the next page for assistance in determining the correct sampling time.

11. Replace the blue plug in the outlet and the blue cap over the inlet when the sampling period is completed. **NOTE:** Handle these items by the outer finger grips. The plugs will prevent contamination of the interior of the cassette during transport, and ensure proper environmental conditions of the collected sample.
12. Place the Via-Cell sampler into the special foil bag and zip it closed.
13. To ensure the integrity of your sample until it reaches the laboratory, apply the red safety seal label over the top of the foil bag opening. Note the sample time and other important sampling information.
14. The sample cassette must arrive at the analysis facility within 24 hours after being collected, so schedule pickup accordingly. **NOTE:** Refrigeration of the collected sample is not required during transport.

Sampling Time Recommendations

The Via-Cell sampler collection period is dependent upon the density of the particulate in the environment. Dusty environments can produce particles overlaying each other on the slide, causing difficulty in direct microscopy analysis. **NOTE:** particles will not adhere to the adhesive media if the adhesive is already covered by particles.

The following table suggests typical sampling times to attain a particle deposition on the slide in which the edges of the trace are sharply defined and the particles dispersed properly to enable good microscopic evaluation.

NOTE:

- Longer/shorter collection times may be required.
- Each situation must be evaluated according to specific conditions and sampling time must be adjusted to meet those conditions.

| RECOMMENDED SAMPLING TIMES | |
|---|---------------|
| Environmental Dust Conditions | Sampling Time |
| Clean office environment or outdoors (no visible dust) | 10 minutes |
| Indoor environment, high activity | 5 minutes |
| Indoor environment, evidence of drywall renovation or industrial dust | 1 minute |

Control Sample Recommendations

- Collection of an outdoor sample may help to determine the amplification of indoor particles.
- Collection of a sample in a 'non-suspected' area may assist with the comparison of results. This can also sometimes be used in place of an outdoor sample when inclement weather conditions are present.
- Collection of field blanks is also recommended.

Laboratory Analysis Strategies

NOTICE

It is imperative that aseptic (sterile) handling procedures be employed by laboratory personnel to prevent sample contamination that could affect analysis.

The Via-Cell sampler is unique in that the particulate collected may be analyzed using one or more analytical methods such as microscopy, culturing, PCR (polymerase chain reaction), or other types of analysis. Since the lower half of the sampling cassette has a built-in trough and the slide media is water soluble, the sample collection may be suspended in water directly in the cassette without the use of additional glassware. This suspension can then be transferred and used in any number of analytical methods that can use this suspension.

Process the collected sample in a timely manner. Experiments have shown that collected mold particles (for example, *Cladosporium cladosporioides*) can maintain viability inside the Via-Cell® up to 5 days after collection. However, the best viability and resulting growth was achieved within 3 days, after that there is diminished viability. Therefore, it is recommended that the collected sample reach the laboratory within 24 hours after collection and that the sample be processed by the laboratory within 24 hours after receipt. Processing may include screening, creation of the suspension and culture. If analysis such as PCR is done, the collection can remain on the collection slide much longer as viability of the particles is not a factor.

NOTE: If culturing of the collection is anticipated, staining of the collection will not be possible for prescreening the particles with direct microscopy since stain kills the viable particles. If PCR analysis is going to be done, staining may be possible; it will be up to the user to determine the suitability of the stain and possible interference with the analysis.

NOTICE

Each Via-Cell sampler should have a plug installed into the outlet port of the cassette as well as the cap over the inlet port when received by the laboratory. These are necessary to seal the cassette to control the environmental conditions as well as prevent contamination during shipment. The outlet plug will seal the bottom of the cassette so water can be added to dissolve the media and suspend the particles. If the plugs are not re-installed after collection, the mold particles may not grow as required for study.

Sample Preparation – Culturing or PCR

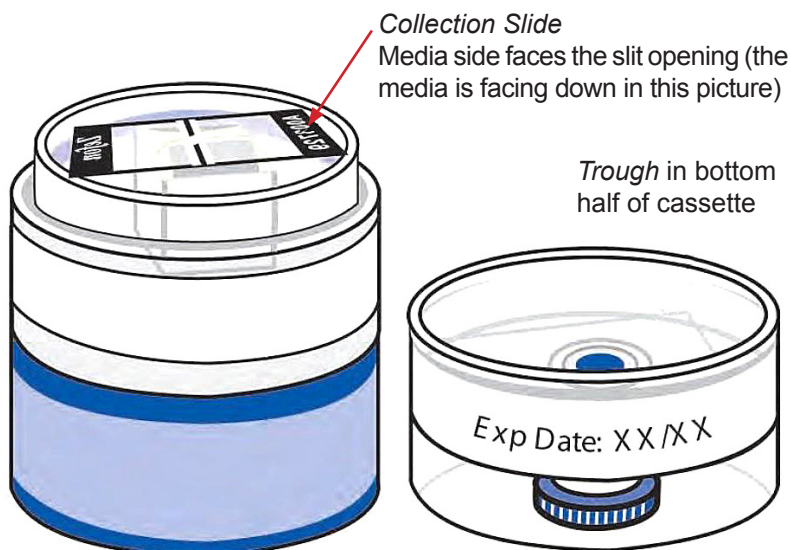
The Via-Cell sampler can be prepared for culturing, PCR and other analysis by suspending the particles in sterile water, then dispensing the suspended particles onto agar plates or other media.

Required Supplies

- Syringe - 3cc sterile, or sterile pipettes
- Sterile water
- Culture plates with appropriate agar
- Sterile particle spreaders
- Self-closing tweezers
- Controlled environmental chamber

Directions

1. Verify that the plug and cap are installed in the cassette.
2. Record the sampler serial number onto the lab worksheet. **NOTE:** The serial number is printed directly on the slide, not on the outside of the cassette.
3. Clean the outer surface of the sampler to remove any excess dirt.
4. Cut the protective tape. Open the cassette in the controlled environmental chamber, keeping the slide in position as shown above. **NOTE:** Be careful not to get pieces of tape inside the sterile cassette.
5. The collection slide will be facing media-side down toward the V shaped inlet sampling port. When you can read the word “ZEFON” on the collection slide, the media will be facing you.
6. Put 2 cc sterile water into the trough in the bottom half of the cassette. After cleaning the tweezers tip with alcohol, place the slide upside down (media side down) on top of the water. If any large bubbles are trapped under the slide, gently lift one side of the slide and slowly lower to remove bubbles. **NOTE:** Be sure the alcohol has completely dried before handling the slide.
7. Leave the slide exposed to water for a minimum of 5 minutes.
8. After 5 minutes, raise the slide with the forceps and flush the face of the slide with 1/4 cc of sterile water. Allow water to fall into cassette. If the adhesive does not completely wash off the slide, let the slide sit an additional 5 minutes to dissolve the remaining adhesive.
9. Draw the suspension from the cassette housing into a sterile syringe or pipette, then dispense onto a culture plate.



Culture Plating Tip – Sometimes it is desirable to split the suspension onto several culture plates. Particles such as molds and fungi germinate in less than 100% RH (liquid water). If more water is used to dissolve the media, it may result in a longer time for the water to evaporate from the surface of a culture plate and for growth to occur.

Direct Microscopy Procedures

Direct microscopy allows the analyst to examine the particles on the slide and then determine the next course of action to take (e.g. culture the sample). If culturing the sample after microscopic analysis is a possibility, it is important to maintain sterility of both the slide and the inside of the cassette. Ensure that the slide is handled with sterile instruments.

Required Supplies

- Standard 1 x 3 microscope slide
- Microscope
- Stain or linking solution
- Self-closing forceps
- Controlled environmental chamber
- Zefon Via-Cell Sterile Microscopy Linking Solution
- Cover Glass



Stains such as Lacto-Phenol Cotton Blue or Saffron Red contain solvents that will destroy the ability for spores to grow. **DO NOT** use any type of stain if you plan to culture the sample after examination.

Sample Preparation

1. Prepare a standard microscope slide by thoroughly cleaning with alcohol.
2. Record the Via-Cell sampler serial number onto the lab worksheet.
3. Cut the adhesive label around the Via-Cell cassette.
4. Aseptically open the cassette in the environmental chamber taking care not to touch the inner surfaces of the cassette.
5. Remove the slide using forceps (tips 'sterilized' with alcohol) and place onto the sterilized microscope slide with the media side up (when the media is facing up, you can read the printing on the slide). **NOTE:** DO NOT place the Via-Cell slide face down onto the microscope slide as transfer of the adhesive and/or particles may occur.
6. Place the slide under the microscope for examination. **NOTE:** Be careful not to contaminate the slide.

Direct Microscopy Slide Preparation

Use when standard direct microscopy is required and the sample may be cultured later.



The Use and Application of Linking Solution

Linking solution is an important step when examining Via-Cell slides under direct microscopy and/or when viability of particles needs to be preserved.

The adhesive on the Via-Cell slide is frosty in appearance in its natural form and needs to be cleared before use in direct microscopy. The linking solution works by partially dissolving the adhesive. It is therefore necessary to place enough solution onto the slide that will allow the cover slip to carry the solution completely under the cover slip. Otherwise blotchy, non-clarified areas may be seen under the cover slip.

Directions

- 1 Place the Via-Cell® slide onto the microscope slide media side up. **IMPORTANT:** Do not permanently mount sample to microscope slide. This preserves the ability to culture the sample.

Via-Cell® slide with media side up → 

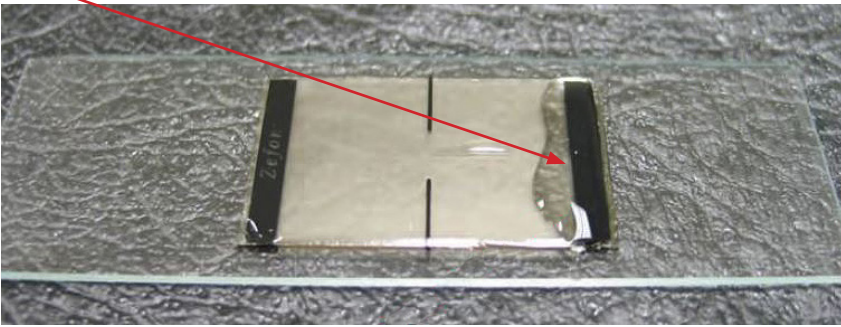
Sterile microscope slide → 



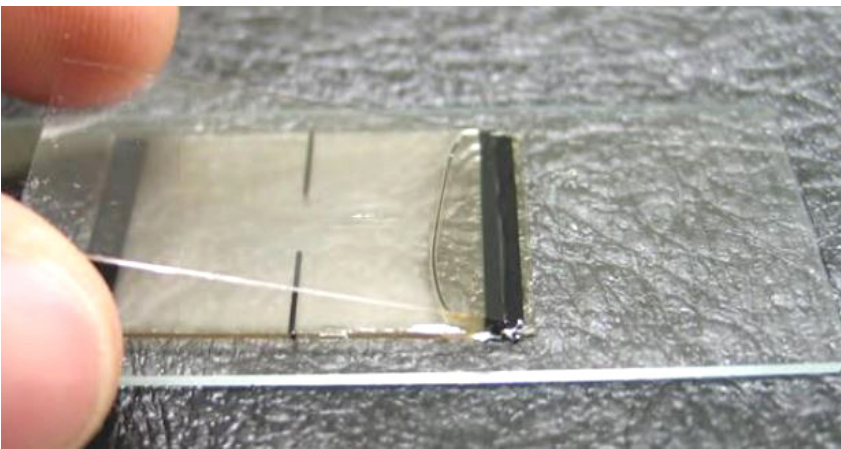
IMPORTANT!

Apply linking solution very carefully and in a specific manner to avoid disturbing the collected sample trace. Please follow the directions regarding proper application of linking solution precisely.

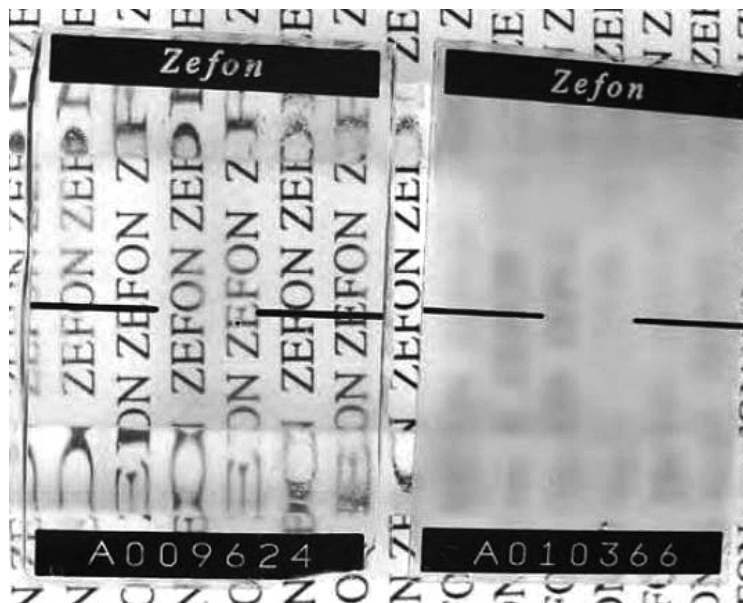
- 2** Place a generous amount of linking solution along the edge of the slide. *DO NOT* place drops of linking solution directly on the trace area, as this will result in excessive disturbance and shifting of particles.



- 3** Lower the cover slip on top of the slide gently at an angle as shown. **NOTE:** Placing the cover slip on the slide this way allows the linking solution to spread across the slide while disturbing the trace as little as possible.



- 4** Preparation is now complete!
Slides take several minutes to clear. Here is an example of a before and after look at the slide. The completed, processed slide should exhibit clarity similar to the example on the left.



Direct Microscopy Preparation Application Summary

- ALWAYS sterilize a cover glass to place over the collection, if you intend to culture the sample later.
- Sterilization is not necessary if you DO NOT intend to culture the sample.
- Use of any type of stain or solvent should only be used if you do not intend to culture the sample. Some stains may not clear the Via-Cell slide adequately. If this happens, contact EMS and we can provide guidance or sell a stain that will clear adequately.
- If using immersion oil, apply it carefully. If immersion oil makes contact with the Via-Cell® adhesive, it will probably result in killing any viable particles.
- Always rinse the cover glass with about 1/4 cc water into the cassette trough to collect any particles that have adhered to it.

Estimation of Collected Particle Load

With direct microscopy, counting and quantification of the collected particles enables the analyst to estimate the particle concentration in the sampled area. This estimation is conducted by counting calibrated cross-sections of the deposited sample trace. The number and type of particles estimated per cubic meter of air is calculated based on the:

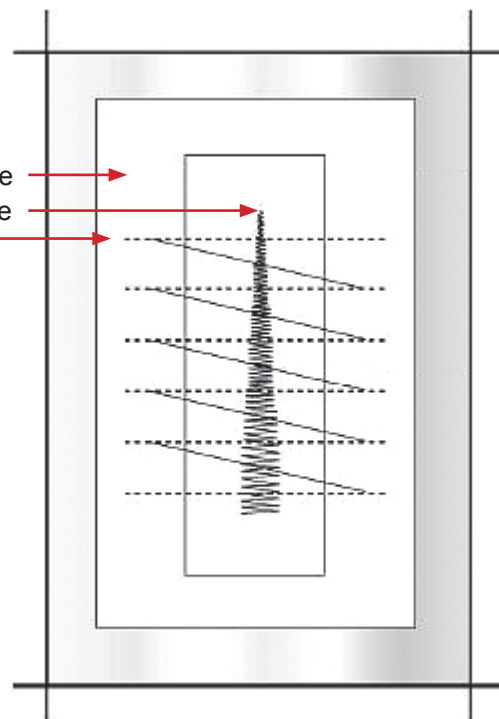
- length of the deposition trace
- length of trace actually examined
- volume of air collected
- number of particles counted

The Via-Cell® Sampler particle deposition area is approximately 0.9 mm wide by 14.4 mm long, for an approximate area of 12.96 mm². The width of the deposition trace may vary slightly in particle density from the middle to outer edges of deposition. For this reason, using the deposition trace area is not recommended for direct calculation of particle concentrations. The recommended procedure for calculating particle concentrations is based on using the Via-Cell® Sampler trace length and microscope field diameter. One field of view counted is defined as the calibrated diameter of the microscope field of view (in mm) covering one cross-sectional pass or "traverse" across the sample deposition trace. A typical sample preparation and microscopic counting procedure is illustrated below.

Counting Procedure

Start analysis at the first traverse. Complete and then move to 2nd traverse. Repeat until the end is reached.

Via-Cell® slide
Particulate sample



The calculation of particle concentration per cubic meter of air can be performed by using the following equations:

- 1** Determine the actual air volume collected in cubic meters (m³) using the following calculation:

$$\text{Air volume (m}^3\text{)} = (\text{Sampling rate (liters per minute)} / 1000) \times \text{Number of minutes}$$

- 2** Determine length of sample trace counted based on the microscope field of view and number of fields of view counted. **NOTE:** To accurately calibrate and measure the diameter of the microscope field of view, use a stage micrometer slide. **NOTE:** Each microscope is different, and each different combination of ocular and objective lens must be calibrated separately. **NOTE:** Stated lens magnifications are rarely precise. The microscopist should then record the number of complete traverses examined across the width of the deposition trace and use the formula given below to calculate the actual length of the deposition trace examined.

$$\text{Trace Length Counted (mm}^2\text{)} = \text{Microscope field diameter (mm)} \times \text{number of traverses}$$

- 3** Determine the concentrations of particles (cts/m³) by using this equation:

$$\text{Cts / m}^3 = \frac{\text{Trace length (14.4 mm)}}{\text{Total length of trace counted (From Step 2)}} \times \frac{1}{\text{Air Volume (m}^3\text{)} \text{ (From Step 1)}} \times \text{\# of particle counts}$$

EXAMPLE

Mold Spore Sample

Microscope field diameter at (900X) = 0.240 mm

Number of traverses = 10

Sample volume (15 lpm @10 minutes) = (15/1000) x 10 = 0.150m³

Mold spore counts = 50

$$\frac{14.4 \text{ mm}}{0.240 \times 10} \times \frac{1}{0.15} \times 50 = \frac{14.4}{0.36} \times 50 = 2000 \text{ ct/m}^3$$

Microscopic Counting Recommendations

| Particle Type | Counting Recommendation |
|----------------|--|
| Pollen | Entire trace or 100 grains (whichever comes first) should be examined at a minimum magnification of 200X. Identification and speciation should be performed at minimum magnification of 400X. |
| Mold Spores | A minimum of 15% of the entire trace should be examined or a minimum of 100 mold spores counted (whichever comes first). Identification and speciation should be performed at a minimum magnification of 400X. |
| Fibers | The entire trace or 100 fibers, (whichever comes first) should be examined at a minimum magnification of 200X. |
| Other Aerosols | Skin cell fragments, combustion emissions, insect parts – A minimum of 10% of the entire trace should be examined or a minimum of 100 particles counted (whichever comes first). |

For any questions or for ordering information,
please contact Customer Service at

1-800-523-5874

Thank you for choosing
Electron
Microscopy
Sciences!

www.emsdiasum.com
sgkcck@aol.com

Tel: 215-412-8400 ♦ *Fax:* 215-412-8450

Electron Microscopy Sciences
P.O. Box 550
1560 Industry Road, Hatfield, PA 19440