USER MANUAL FOR

BIOAIRE™ CASSETTE
Impaction Particle Sampler

Quantitative Analysis of:
Bioaerosol ♦ Molds ♦ Pollen
**BioAire™ Cassette** Impaction Particle Sampler

**APPLICATION**

The BioAire™ cassette impaction particle sampling cassette manufactured for A. P. Buck, Inc. is a unique sampling instrument specifically designed for the rapid collection and analysis of a wide range of airborne aerosols including but not limited to bioaerosols (mold spores, pollen, insect parts, skin cell fragments, etc.), fibers (asbestos, fiberglass, cellulose, etc) and inorganic particles.

**APPLICATIONS**

**Indoor Air Quality Testing**

Mold spores, pollen, insects, skin cell fragments, plant fragments, dust fibers, combustion emissions, etc.

**Allergy Testing**

Mold spores, pollen, insects, etc.

**Clean Room Monitoring**

Evaluation of low airborne dust and contaminants from personnel (skin cells, clothing fibers, cosmetics, etc.)

**Restoration of Buildings**

Evaluation of mold spore contamination before, during and after remediation

This device combines the convenience of using pre-loaded disposable filter cassettes for rapid microscopic analysis while preventing cross contamination.

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**RECOMMENDED MICROSCOPIC COUNTING GUIDELINES**

**Counting & Identification Guidelines**

<table>
<thead>
<tr>
<th>Aerosol Type</th>
<th>Minimum Examination Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pollen</td>
<td>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.</td>
</tr>
<tr>
<td>Mold Spores</td>
<td>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 minimum magnification of 600x.</td>
</tr>
<tr>
<td>Fibers</td>
<td>The entire trace or 100 fibers, (whichever comes first) should be examined at a minimum magnification of 200x.</td>
</tr>
<tr>
<td>Other Aerosols</td>
<td>Skin cell fragments, combustion emissions, insect parts a minimum of 10% of the entire trace should be a minimum of 100 particles counted (whichever comes first).</td>
</tr>
</tbody>
</table>

The BioAire™ Cassettes are available in boxes of 10 or in cases of 50.

**ORDER NUMBER**

<table>
<thead>
<tr>
<th>Number</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>SAM-500810</td>
<td>Box of 10</td>
</tr>
<tr>
<td>SAM-500850</td>
<td>Case of 50</td>
</tr>
</tbody>
</table>

**NOTE:** The BioAire™ Cassette was designed to be used as a stand alone air sampling device. Since the product has been on the market, several “accessory” items for use in specific situations have come on the market which are NOT manufactured by A. P. Buck. A. P. Buck remains independent of all these devices as the collection and interpretation of data may vary significantly from standard air sampling procedures. It is important to understand this and to obtain information from the suppliers of these devices regarding the data it will provide.
Two example calculations for mold spores and pollen grains are given below:

<table>
<thead>
<tr>
<th>Example 1</th>
<th>Mold Spore Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microscope field diameter at (900x)</td>
<td>0.240 mm</td>
</tr>
<tr>
<td>Number of traverses</td>
<td>10</td>
</tr>
<tr>
<td>Sample volume (15LPM @ 10 minutes)</td>
<td>(15/1000) x 10 = 0.150 m³</td>
</tr>
<tr>
<td>Mold spore counts</td>
<td>50</td>
</tr>
</tbody>
</table>
| \[
\begin{array}{ccc}
14.4 \text{ mm} & \times & 1 \\
0.240 \times 10 & \times & 50 \\
\end{array}
\]
| \[
\begin{array}{ccc}
0.15 & \times & 0.36 \\
\end{array}
\]
| \[
14.4 \times 50 = 2000 \text{ ctm}^3 \\
\]

<table>
<thead>
<tr>
<th>Example 2</th>
<th>Pollen Example</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microscope field diameter at (200x)</td>
<td>1.11 mm</td>
</tr>
<tr>
<td>Number of traverses</td>
<td>10</td>
</tr>
<tr>
<td>Sample volume (15LPM @ 10 minutes)</td>
<td>(15/1000) x 30 = 0.450 m³</td>
</tr>
<tr>
<td>Pollen counts</td>
<td>25</td>
</tr>
</tbody>
</table>
| \[
\begin{array}{ccc}
14.4 \text{ mm} & \times & 1 \\
1.11 \times 10 & \times & 25 \\
\end{array}
\]
| \[
\begin{array}{ccc}
0.45 & \times & 5.0 \\
\end{array}
\]
| \[
14.4 \times 25 = 72 \text{ grains/m}^3 \\
\]

**BioAire™ CASSETTE ADVANTAGES**

- Detection limit improvement of approximately 100 to 1000 times over conventional filter sampling.
- Eliminate sample loss to cassette walls known to occur with filter samples from vibration or static charge during sampling and shipment.
- Eliminates the need for direct handling of collection media or microscope slides in the field.
- Eliminates potential cross-contamination between samples and during shipping that may occur with other slit impaction devices.
- Unique optically transparent and smooth collection media allows direct staining and examination by bright filed, dark filed, and phase contrast microscopy!\
- The sampling media is compatible with a wide range of biological stains and refractive index oils allowing for direct quantitative analysis of biological and inorganic particles.
- The BioAire™ cassette does NOT require the purchase of specialized (sampling) umps and will operate with conventional air vane or diaphragm high volume pumps.
PRINCIPLE OF OPERATION

The BioAire™ cassette operates on the well established principle of inertial impaction. Particles in the air stream are accelerated to a minimum velocity of approximately 40 mph through a tapered slit (known as the Hirst slit) and aimed directly at a sticky and optically clear sampling media. The design of the BioAire™ cassette forces the air stream to make a sharp 90° turn at the surface of the sampling media prior to exiting the back of the sampling cassette. The inertial velocity and mass of the particle combine to cause the particle to impact and adhere to the sampling media rather than continue following the air stream. The sticky media utilized in the BioAire™ permanently fixes the impacted particles in place.

Although the efficiency of impaction collection decreases with particle size and density, mold spores as small as 3.0 µm in diameter are collected with a theoretical efficiency of approximately 85%. Large particles such as pollen grains (15-80 µm in diameter) are captured with an efficiency of greater than 95%. The air flow path through the inlet orifice of the BioAire™ and around the sampling media to the exit of the cassette is illustrated below in Figure 1.

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

First determine the actual air volume collected in cubic meters (m³) by following the calculation given in Equation 1.

**EQUATION 1:**

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

Second, determine the length of sample trace counted based on the microscope field of view and number of fields of view counted. Accurately calibrate and measure the diameter of the microscope field of view using a stage micrometer slide. Remember, each microscope is different and each different combination of ocular and objective lens must be calibrated separately. 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 in Equation 2 to calculate the actual length of the deposition trace examined.

**EQUATION 2:**

\[
\text{Trace Length Counted (mm}^2\text{)} = \text{Microscope field diameter (mm)} \times \text{Number of Traverses}
\]

The concentrations of particles (cts/m³) can then be determined by using Equation 3.

**EQUATION 3:**

\[
\frac{\text{trace length (14.4 mm)}}{\text{total length of trace counted (From Equation 1)}} \times \frac{1}{\text{Air volume (m}^3\text{) (From Equation 2)}} \times \text{number of particle counts}
\]
MICROSCOPIC EXAMINATION

Analysis of the collected sample should be performed by an experienced Microbiologist, Aerobiologist, or Environmental Microscopist. Counting and quantification of sample components is conducted by counting calibrated cross-sections of the deposited sample trace. The number and type of particles counted per cubic meter is calculated based on the length of the deposition trace, length of trace actually examined, volume of air collected and number of particles counted.

The BioAire™ particle deposition area at a flow rate of 15 LPM is approximately 1.1 mm wide by 14.5 mm long yielding an approximate area of 15.95 mm². The width of the deposition trace will vary slightly with flow rate and media thickness, and will 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 BioAire™ trace length and microscope field diameter and will be discussed below. 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 in Figure 2.

RECOMMENDED SAMPLING PROCEDURES

General:

The BioAire™ sampler is designed to operate at an optimal flow rate of 15 LPM. The user can employ any sampling pump capable of a minimum flow rate of 15 LPM. Sampling flow rates of up to 30 LPM can be employed for specific applications. The impaction sampler is capable of operating in any vertical or horizontal orientation or in restricted access spaces smaller than 2 inches in diameter. As a result the BioAire™ is ideally suited for sampling in HVAC ducts, plenums, wall cavities or other confined spaces.

Sampling of Ambient Static Environments:

A Rotameter calibrated to a primary standard should be used to calibrate the sampling pump to a flow rate of 15 LPM. The rotameter should be connected directly to the BioAire™ cassette to calibrate the pump flow rate.

The BioAire™ cassette is connected to the sampling pump using flexible tubing and the tape seal covering the inlet is removed and placed on the side of the device. The sampling pump is turned on for an appropriate sample time ranging from 1 to 10 minutes and the seal replaced after sampling is complete. Unlike other spore trap impaction or filter devices, the BioAire™ cassette can be oriented in any vertical or horizontal direction, without concern for sample loss of large particles or vibration. “Outside background” samples should always be collected for comparison purposes.
Sampling in HVAC Systems:
The BioAire™ cassette is the only disposable single use impaction sampling device on the market that allows for isokinetic sampling of aerosols in Heating, Ventilation and Air Conditioning (HVAC) Systems. Sampling can be conducted at the supply diffuser or inside most conventional ducts. The inlet of the cassette should always be facing into the flow stream. The inlet orifice as a cross-sectional area of approximately 11 mm x 15 mm (165.0 mm²) tapering to a slit with dimensions of 1.255 mm x 14.4 mm (15.19 mm²). The flow velocity can be increased up to 30 LPM with conventional sampling pumps, however, air flows exceeding 20 LPM may potentially damage some bioaerosols or cause "bounce off". Isokinetic sampling can be conducted in most air duct systems with flow rates of up to approximately 600 FPM. Approximate face velocities for the BioAire™ cassette are given below for both the entrance orifice and slit exit in Table

BioAire™ Theoretical Face Velocities

<table>
<thead>
<tr>
<th>Flow Rate (LPM)</th>
<th>Orifice Face Velocity (FPM)</th>
<th>Orifice Face Velocity (MPH)</th>
<th>Slit Face Velocity (FPM)</th>
<th>Slit Face Velocity (MPH)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15.0</td>
<td>299</td>
<td>3.4</td>
<td>3110</td>
<td>35.3</td>
</tr>
<tr>
<td>20.0</td>
<td>399</td>
<td>4.5</td>
<td>4146</td>
<td>47.2</td>
</tr>
<tr>
<td>25.0</td>
<td>499</td>
<td>5.7</td>
<td>5183</td>
<td>59.0</td>
</tr>
<tr>
<td>28.3</td>
<td>564</td>
<td>6.4</td>
<td>5867</td>
<td>66.8</td>
</tr>
<tr>
<td>30.0</td>
<td>598</td>
<td>6.8</td>
<td>6219</td>
<td>70.6</td>
</tr>
</tbody>
</table>

Recommended Sampling Time Intervals:

As mentioned above, flow rates exceeding 20LPM have been known to cause “bounce off” of large particles such as pollen grains. Flow rates lower than 10 LPM will not collect the small mold spores (such as Aspergillus and Penicillium) as efficiently. Recommended sampling times (at 15 LPM) for different environmental sampling conditions are given in Table 2.

<table>
<thead>
<tr>
<th>Environmental Dust Conditions</th>
<th>Sampling Time In minutes at 15 LPM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Outdoor sampling on a clean windless day</td>
<td>10.0 - 60 min</td>
</tr>
<tr>
<td>“Clean” office environment or outdoors (no visible dust)</td>
<td>10.0 min</td>
</tr>
<tr>
<td>“Indoor” environment, high personnel activity</td>
<td>5.0 min</td>
</tr>
<tr>
<td>“Indoor” environment, evidence of drywall renovation, or industrial dust</td>
<td>1.0 min</td>
</tr>
<tr>
<td>“Indoor” environment, visible dust emissions from point sources present</td>
<td>0.5 min</td>
</tr>
</tbody>
</table>

RECOMMENDED ANALYSIS PROCEDURES

Slide Preparation

One to two (1-2) drops of staining or mounting media (Lacto Phenol Cotton Blue is recommended for mold spore analysis) should be placed in the center of a clean pre-labeled slide. BioAire™ cassettes should only be opened in the laboratory. The sealing band should be removed, and the glass cover slip (containing the sample trace) removed and slowly placed on an angle with the media collection side down onto the staining solution. **Do not press down on the slide during or after staining!** Excess staining solution should be removed from around the edges of the cover slip with a tissue wipe or cotton swab after 10 minutes has elapsed. This will ensure even staining of the sample.