

► THE PRACTITIONER'S TOOL KIT

Practical Strategies for Monitoring Bioaerosols

Editor's Note: The National Environmental Health Association (NEHA) strives to provide relevant and useful information for environmental health practitioners. In a recent membership survey, we heard your request for information in the *Journal* that is more applicable to your daily work. We listened and are pleased to feature this column from a cadre of environmental health luminaries with over 300 years of combined experience in the environmental health field. This group will share their tricks of the trade to help you create a tool kit of resources for your daily work.

The conclusions of this column are those of the authors and do not necessarily represent the official position of NEHA, nor does it imply endorsement of any products, services, or resources mentioned.

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Monitoring bioaerosols has two strategic components: 1) air sampling and 2) air monitoring.

Air sampling analyzes the airborne environment at a particular point in time (i.e., incidence). It provides essential information to identify the root causes of airborne contamination. By sampling specific operations—such as food preparation, material movement, and sanitation, along with active and quiet periods of these activities—we can develop a framework for effective remediation and prevention strategies.

Air monitoring documents changes in air quality and contamination over time (i.e., prevalence). Sampling aerosols for the presence (or absence) of microbes provides valuable information on the movement and ultimate fate of small, suspended particles generated by normal human activity and other environmental influences.

Both monitoring components are necessary to understand the dynamics of bioaerosols.

Sampling

The sampling strategy consists of both passive and active monitoring and sampling.

Passive monitoring uses settling plates (e.g., Petri dishes). The sampling media is exposed to the air for specified time periods. Ideally, these time periods are during activity or production, clean-up, and sanitation, and after shutdown. Passive monitoring helps determine which microbiological particles might be present and suspended in the environment, as they settle out over time onto the media surface of the Petri dishes. The exposed plates are incubated and analyzed. The data

as any environment where contamination and disease risk are critical factors.

To control the aerosolization of microbes, we need information on their numbers and types to relate these data to activity and air-flow characteristics. Moreover, controlling aerosols complements requirements from the Food and Drug Administration's *Food Code*.

The Science

The basis of a bioaerosol monitoring program is systematic sampling and testing of the airborne environment for potential sources of contamination, such as pathogens, allergens, spoilage organisms, and other microbial contaminants that might not be inherent to the facility. It is a useful tool for validating the effectiveness of ventilation systems that are missed by surface sampling alone.

In short, sampling solid surfaces and liquids provides information about the presence of contaminants and indicator organisms. It answers the questions of what is present and where it is present, but it does not tell us why it is there or how it got there in the first place. Air sampling and characterization of ambient air, along with both mechanical and passive ventilation, will help provide those answers.

Introduction

This topic might be far afield from what we regularly do as regulators. Air sampling for bioaerosols, however, is part of the environmental monitoring program (EMP) conducted by food processors. Guided by regulations such as the Food Safety Modernization Act, an EMP is designed to proactively identify potential safety issues, enhance sanitary conditions, and ensure regulatory compliance, thereby safeguarding consumers and protecting brands. While the core of the EMP is surface sampling, air monitoring is used to diagnose and identify problems encountered when finding pathogenic bacteria in strange places and under questionable conditions.

Retail kitchens are simply smaller, but often more complex versions of larger food plants. Knowing the science and application of sampling bioaerosols will definitely help us understand testing data and its interpretation. It also helps in assessing the risk of cross-contamination from moisture, splashes, and spray. Knowing the fate and transport of droplets and droplet nuclei provides background and support information on refining operations and, specifically, cleaning procedures. It can target food preparation and production areas, as well



Photo 1. Centrifugal impactor sampler with agar sampling strip.



Photo 2. Sieve impactor sampler with a RODAC (replicate organism detection and counting) sampling head.

yields comparative findings by activity and location. Recovery of the most prevalent organisms can be enhanced by varying incubation temperatures. If the exposed plates are initially incubated at 22 °C for 24, 48, and 72 hr and subsequently incubated at 36 °C, both psychrophilic (molds and heterotrophs) and mesophilic (potential disease causing) organisms are recovered.

By using selective and nonselective media, a qualitative analysis of bioaerosols is possible. It could even provide insight into the ventilation system's effectiveness and identify biological characteristics and patterns of air movement.



Photo 3. Slit-to-agar sampler with a clock mechanism.

Active air sampling provides both qualitative and quantitative analyses of the microbiological bioburden. Mechanical air samplers are used to force air into or onto a nutrient agar-based test medium. Active monitoring devices are ideally suited for sampling low microbial concentrations because a greater volume of air passes through them. Using selective and nonselective media, both disease-causing and spoilage organisms can be found, identified, and quantified. Specific operations or environmental conditions within the facility can easily be targeted. In a food environment, active air monitoring can yield data on aerosol generation during production, internal movement, plating, packaging, serving, and, particularly, during sanitation activities.

Active air monitoring devices are used for shorter sampling periods (e.g., 1–60 min) and require preplanning and continu-

ous monitoring during operation. Organism recovery might be enhanced by varying incubation temperatures. The data from bacterial and/or fungal counts (CFUs) collected can be identified and comparisons made before, during, and after periods of specific but normal activities. Outcomes might vary within the same area due to changes over time and activity. Patterns are readily recognizable, however, and bias can easily be eliminated.

Both passive and active monitoring have their strengths and weaknesses. Passive monitoring is not aggressive and can miss critical microbes. It does, however, allow for a lengthy (approximately 4 hr) sampling period at a low cost. Active monitoring requires equipment purchases or rental, additional training, and, in most instances, device qualification. Device qualification refers to ensuring that the monitoring equipment is properly positioned, operates as intended,

and performs consistently under normal conditions as detailed in its user manuals.

Choosing between passive and active air sampling really depends on the testing requirements and perceived achievable goals. In most cases, both methods complement each other in terms of qualitative and quantitative analyses, cost, and time needed for testing. Because there is no standardized protocol for collecting air samples, it is difficult to determine which method is more effective. Knowing the differences, however, can help determine which solution will work best within a defined environment.

Instrumentation

There are five active air monitoring systems to choose from: 1) centrifugal, 2) slit-to-agar, 3) sieve samplers, 4) filtration, and 5) liquid impingement. The first three systems, when used in concert with one another and passive settling plates, offer the most useful, efficient, and overall representative picture of the aerobioburden of an area being sampled.

The centrifugal impaction sampler, also known as RCS (Reuter centrifugal impaction sampler), uses an agar strip that is fed into the sampler head (Photo 1). Air is drawn into the top of the unit by means of an impeller, and the collected organisms of all sizes and types are deposited onto the agar strip by centrifugal force. The sampling time can be adjusted according to the expected aerobioburden, ranging from 1 to 10 min. The power source is four, D-cell batteries. Results are reported as organisms per L or ft² of air. These units are lightweight, fast, and easy to use. Because of their portability and ease of use, centrifugal samplers are ideal for identifying biological hotspots, characterizing airborne organisms by activity and location, and mapping bioburden throughout any area of concern.

Sieve impaction sampler, also known as an Andersen sampler or Andersen impactor, draws air through a series of small holes in the sampling head, and collected organisms are impacted on an agar surface (Photo 2). The sampler was originally designed to measure airborne particles by respirable versus nonrespirable size and is therefore ideal for characterizing droplets generated near the sampling site and droplet nuclei that remain in suspension and travel to remote locations. These air samplers have the distinct advantage of using either 55 mm RODAC (replicate

organism detection and counting) plates, which can also be used for surface sampling, or 100-mm agar-containing Petri dishes, which can double as settling plates. The sieve impaction sampler is also used to evaluate the presence of target organisms in the bioaerosol when used with selective media. As with other sampling strategies, organism selectivity can also be achieved by varying the incubation temperature. The sampling time is typically 10 min per run. The portability of the sieve sampler, similar to that of the centrifugal impaction sampler, allows sampling at multiple sites within a defined area. This sampler is particularly useful in providing information on where microbial aerosols are generated and how far they can travel from their point of generation.

The slit-to-agar sampler measures bioaerosols in real time (Photo 3). The sampler consists of a stationary sampling head and a revolving base. The most common slit-to-agar sampler is the Mattson-Garvin sampler. It uses a commercially available 150 mm × 15 mm agar plate for sample collection. As the sample plate rotates, air is drawn through the slit, and microorganisms are deposited on the agar surface. A full rotation of the plate is typically 60 min. Organism distribution on the agar is time- and concentration-dependent. The CFU placement on the agar plate correlates with the sampling time and can be read like a clock dial, which is particularly useful for assessing bioaerosol generation across different operations, conditions, and movement in real time. These data allow for further refinement to prevent aerosol generation during a specific activity. Because these samplers are cumbersome, a single sampling site is chosen and maintained throughout the sampling run.

Sampling Protocol

Determination of sampling sites, sampling frequency, and collection times for any sampling device depends on need and practicality. When selecting sampling sites, begin by mapping the area, describing the activities within it, and identifying equipment, furnishings, and construction materials relevant to the movement and cleaning of the area. In addition, characterize the airflow patterns during periods of activity and inactivity.

Monitoring three additional air parameters will be invaluable for understanding the results of the bioaerosol qualitative and quan-

titative testing. These parameters include temperature, humidity, and characterization of airborne particulate matter in both the ±2.5-micron and ±10-micron range. The sampling devices for these air quality measurements do not need to be exact; they are used for reference and to help interpret the test results in a dynamic environment. As we mentioned earlier, devices used for measuring these parameters are portable, inexpensive, and easy to use. As a plus, they are valuable tools for other air-related applications.

Bottom Line

Including bioaerosol evaluations in any environmental monitoring program provides answers to why a contaminant is there and how it got there in the first place. This information is essential for developing effective corrective and preventive measures for the control of contamination from airborne sources. ✨

Photo credit: Photos courtesy of Dr. Robert Powitz.

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