Simplify Coliform Screening as Part of Your Environmental Monitoring Program

Potential microbial contamination can come from seemingly anywhere: through raw materials, the production process, the equipment, the environment and of course, through employees. That's why it's important to check all areas of your facility with a structured sampling and environmental monitoring program (EMP). This article will help you identify the best approach for screening, depending on your facility.

The food industry has adopted a preventative, proactive approach to correcting potential safety issues. As part of the preventative approach, food manufacturers should use additional monitoring methods to determine the general hygiene of food processing areas. This includes microbiological testing which must be done to verify the effectiveness of any EMP. However, it can often be difficult to test large areas or batches of product. In cases where a full-scale test isn't possible, or for more frequent testing, it can be beneficial to test for organisms whose presence tends to coincide with pathogens, contamination, or spoilage. These organisms are called indicator organisms, and they can act as early warning sign. **An indicator can signal a microbiological problem that could impact the quality or the safety of a food ingredient, finished product, or production/processing/handling equipment.** They can also be used as an indication of a potential hygiene problem. Essentially, they let you know there's a problem before it becomes a real problem.

Selecting an Indicator Organism

The challenge arises when determining which indicator organism(s) to use. One must consider multiple criteria that are in place for the food product or environment in question. Variables such as the possible sources of pathogenic microorganisms; their incidence on or in the product; production, harvesting and processing practices; survival and growth of pathogens or spoilage organisms in the product; and specific analytical methods available for detecting the indicator, are all important considerations in indicator selection. However, in general, several key features are found in an "ideal" indicator:

1. The indicator microorganism is usually non-pathogenic.
2. Methods for detection of the indicator should be rapid, simple, inexpensive, widely available, and easy to interpret.
3. The indicator should have readily identifiable characteristics so it can be easily discriminated from natural, competing microflora.
4. If possible, indicator microorganisms should occur at a sufficient frequency and/or level when a food safety system is under control, such that:
   a. Baseline levels of the organism are clearly identified.
   b. Out-of-control conditions are reflected in a significant increase in the level of the indicator.
   c. Corrective actions can be taken before a pre-determined limit is exceeded.
5. Decision criteria for the indicator microorganism should be relevant at multiple points along the food chain.
6. If possible, indicator assays should provide enumerative results so that degrees of contamination can be assessed.
7. When used as a surrogate, the characteristics of the indicator should correlate with the characteristics of the target (i.e., same habitat, clear correlation between presence/absence of target and indicator, correlation between level of target and level of indicator) and survival/resistance of the indicator should be comparable to the target.

Choosing Coliform Bacteria

As noted above, different indicator organisms in food can point to different problems, so it's important to know what you're looking for and cover all your bases. Historically, coliform bacteria have been used as indicators of unsanitary conditions in water and foods for over a century. This concept originated in the late 1800s after *E. coli* was found to be ubiquitous to feces, and its detection in water was used to "indicate" an increased likelihood that pathogens such as *Salmonella* Typhimurium (typhoid fever) were in the water as well (i.e., an indicator of unsanitary conditions).

Coliform bacteria belong to the *Enterobacteriaceae* family and include species of the following genera: *Citrobacter, Enterobacter, Escherichia, Hafnia, Klebsiella, Serratia*, and *Yersinia*. The presence of coliform microorganisms in drinking water represents a sign of fecal contamination and indicates the potential contamination also with pathogenic bacterial species such as *Shigella* spp., *Salmonella* spp., or *Vibrio cholerae*. They are used traditionally as indicators of water quality because the testing methodology is simple and inexpensive. In addition, they have been used as a potential measure of the presence of enteric pathogens in food.

One of the most common applications of coliform bacteria as indicator organisms is their association with hygienic conditions and overall quality, especially concerning heat processed food – they are typically killed during most heat processing conditions. If found in the final product, their presence generally indicates an inadequate heat processing or a post-processing contamination. The presence of any *Enterobacteriaceae* typically means that contamination most likely occurred after heat processing or cleaning was inadequate.

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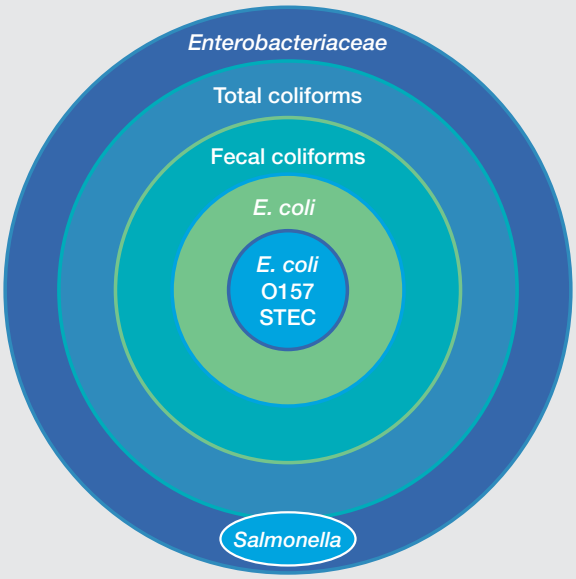

The relationship between *Salmonella,* pathogenic *E. coli* (*E. coli* O157/ STEC) and the commonly used *Enterobacteriaceae* hygiene indicator, and related hygiene indicators.

Testing Options for Coliforms

As noted in **BAM** (Bacteriological Analytical Manual), three groups of coliforms are used as indicators but in different applications. Detection of total coliforms is used as an indicator of sanitary quality of water or as a general indicator of sanitary condition in the food-processing environment. Fecal coliforms remain the standard indicator of choice for shellfish and shellfish harvest waters, and *E. coli* is used to indicate recent fecal contamination or unsanitary processing.

Hazards from the coliform, *E. coli*, can specifically be prevented by heating sufficiently to kill the bacteria, holding chilled products below 40° F, preventing post cooking cross-contamination and preventing ill employees from working in food operations. Other coliforms can be eliminated in similar ways. If processes run out of control, pathogenic strains of *E. coli* and other coliforms could survive, posing risks to consumers, especially the young and immunocompromised.

While strict, defined operations can prevent contamination, they don't ensure that the final product is free of all possible pathogens of concern. Therefore, it is critical to include indicator organism testing as part of your facility's EMP. (Note: indicator organism testing is not a substitute for testing for pathogens; additional testing must also be performed).

For each specific indicator, simple pass/fail criteria can be established by reviewing historical data for the past six to twelve months. The EMP and the target/baseline are unique for each plant, each type of product and the baseline is different for different zones. Many facilities rely on traditional culture methods for detection of contamination.

One of the most common indicator tests is Total Plate Count (TPC). Also known as Aerobic Plate Count, it provides information on the total population of bacteria present. It is used as a measure of general sanitation, effectiveness of intervention steps, microbiological quality, and spoilage in matrices including ready-to-eat (cooked) foods, pasteurized milk and spices. Additional plating methods are used to detect coliforms (in raw ground meat, water, and spices), fecal coliforms (in raw ground meats, water and seafood), *Enterobacteriaceae* (in infant formula and ready-to-eat foods), and *E. coli* (in seafood, water, and ready-to-eat foods).

All these testing methods are time-consuming and require additional plating and biochemical testing for specific species or strains. After days of testing, if positive results for a pathogen are obtained, then the product made on that line must be held until further confirmative test results are available, and it will most likely initiate a recall situation or discarding of precious product. This increases the overall costs for the facility – discarded product, line stoppage, additional cleaning and retesting.

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Rapid Testing Solutions

The best way to reduce product contamination is to ensure proper hygiene and sanitation in between TPC testing. As an alternative to plating, facilities should consider more rapid testing to indicate the presence of any indicator organisms. Tests must be simple, sensitive, accurate, easy to use and provide results as quickly as possible, preferably within a shift (6 - 8 hours). One solution meeting these criteria is MicroSnap™ rapid test devices. Tests are available for total viable counts (TVC), coliforms, *E. coli*, and *Enterobacteriaceae*. Whether testing a raw material, plant environment, or finished product, MicroSnap can be used to screen for microbiological contamination.

MicroSnap™

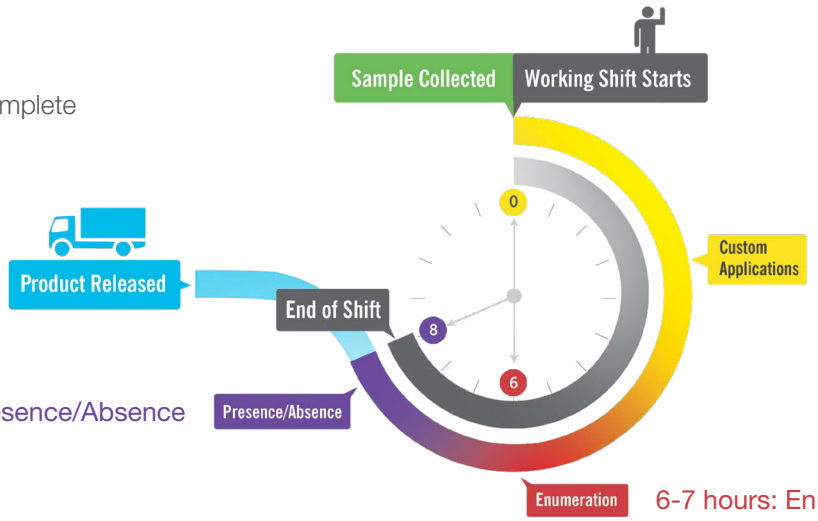
Product	Organisms Detected
EB	All *Enterobacteriaceae*
E. coli	All *E. coli*, including O157
COLIFORM	All coliforms, including *E. coli*
TOTAL	Gram positive and Gram negative bacteria

MicroSnap is a novel, rapid test system capable of detecting bacteria using a modification of the adenosine triphosphate (ATP) bioluminescence reaction, which most processors are familiar with from routine ATP sanitation monitoring. This assay can detect bacteria at low levels in a variety of sample types to give results in 6–8 hours using the easy-to-use, sensitive, accurate EnSURE™ Touch luminometer from Hygiena™. Utilizing a novel, bioluminescent test methodology, the light generating signal can be quantified in the multi-functional EnSURE Touch system and stored in a cloud-based software application (SureTrend™) for further analysis, reporting and trending. The system has been AOAC-RI*PTM* validated for the enumeration of total bacteria (TVC), coliforms and *E. coli*.

Implementing this simple, rapid, low-cost test system can provide you with results in the same working day or shift – ideal for monitoring the quality and safety of food as well as post-cleaning surface hygiene. This platform uses the same foundation of traditional pour plates or sample-ready media film with an enrichment step that allows the microorganisms to grow.

Same-shift results

The entire MicroSnap test process is complete in less than 8 hours vs standard testing methods taking 2-3 days.



Instead of counting colonies, MicroSnap uses a unique formulation and rearrangement of the ATP bioluminescence biochemistry coupled to specific substrates to detect specific bacteria at low levels in just a few hours. These tests detect between 1–10,000 CFU, whereas sample-ready media film typically detects between 10–300 CFU. The test is flexible for environmental sampling with a built-in sample collection swab for assessing surface contamination and environmental hygiene. The unique self-contained packaging format simplifies the technology, so the test is simple and easy to use in any facility by any user. In addition, the use of this platform reduces costs, especially for facilities using outside laboratories for these quality tests. Learn more about these solutions at **www.hygiena.com**.

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