

Guidelines for *Listeria* Testing of Environmental, Raw Product and Finished Product Samples in Smoked Seafood Processing Facilities

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SUMMARY

The Smoked Seafood Working Group (SSWG), a collaboration of two US industry trade organizations, smoked seafood processors and academia, developed guidelines for controlling *Listeria monocytogenes* in smoked seafood operations. To minimize the potential for *L. monocytogenes* contamination of finished products, it is necessary to have sanitation procedures that prevent contamination of product contact surfaces and eliminate niches where *L. monocytogenes* can become established, grow, and persist. Environmental testing can be used to help identify problem areas or locate contamination sources in the plant, and to confirm that problem-solving procedures have been effective. Raw seafood and finished product testing can be used to evaluate raw product suppliers and verify the effectiveness of control procedures. Regular testing can also help to track performance over time and identify new sources or reservoirs of contamination in the processing plant environment. This paper describes considerations for developing effective environmental and product testing programs for *L. monocytogenes* and provides four examples to illustrate how testing programs could be structured for various types of smoked seafood processors.

INTRODUCTION

L. monocytogenes contamination has been found in both hot- and cold-smoked seafood (2, 6, 8). Efforts to control *Listeria monocytogenes* in the food processing plant environment can reduce both the frequency and level of contamination in smoked fish products, but it is not possible, given current technology, to completely eliminate the organism from the processing plant environment or totally eliminate the potential for contamination of finished products (4, 20). Although the process of producing cold smoked products does not include a heating step that will eliminate *L. monocytogenes* that may be present on the raw material, *L. monocytogenes* contamination during processing appears to be a major source of finished product contamination (5, 14). The potential for contamination of cold-smoked product after the smoking process must thus be evaluated and minimized. Although hot smoked seafood products do reach a high enough temperature for sufficient time (145°F (62.8°C) for 30 minutes) to kill *L. monocytogenes*

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(10), these products can also be contaminated at processing steps that occur after the product is smoked (16). To minimize the potential for *L. monocytogenes* contamination of finished products, it is necessary to have sanitation controls that prevent contamination of product contact surfaces and eliminate niches where *L. monocytogenes* can establish itself, grow, and persist (5, 22). Environmental testing can be used to help identify problem areas or locate contamination sources in the plant, and to confirm that problem solving procedures have been effective (20, 21). An ongoing testing or monitoring program can be used initially to help determine what control measures are most effective and where changes or modifications in plant procedures are needed. When these measures have been implemented, regular testing can then help to track performance over time and identify new sources or reservoirs of contamination in the processing plant environment (3, 19, 20).

Since 2001, representatives of the National Fisheries Institute and the National Food Processors Association, individuals from at least 10 smoked seafood firms, and food safety or seafood specialists from Cornell University, Virginia Tech and the Sea Grant programs in New York and Delaware have been working together as the Smoked Seafood Working Group (SSWG) to develop guidelines for the control of *Listeria monocytogenes* in smoked seafood manufacturing plants. This initiative was also conducted as part of a Cornell University project to develop control strategies for *Listeria monocytogenes* in food processing environments, funded under the National Food Safety Initiative in 2000 by the Cooperative State Research, Education and Extension Service of USDA, Project Number 00-51110-9768.

The following information is designed to offer smoked fish processing plants a variety of options for developing an effective environmental and product monitoring program. Examples are provided to illustrate different types of environmental testing programs for smoked fish processing operations. An effective monitoring program should include decisions as to:

- what areas of the plant are to be tested,
- the frequency of testing,
- what testing procedures will be used,
- how test results will be evaluated, and

- what actions will be taken when test results are positive.

A brief description of these elements, illustrated by four different examples of how these components could be integrated into a complete plan, follows.

ESTABLISHING A SAMPLING/MONITORING PROGRAM

Deciding on the test organism

One of the first steps in establishing a sampling/monitoring program is to determine whether to test for *Listeria* species (*Listeria* spp., or generic *Listeria*) or for *L. monocytogenes*. Within the genus *Listeria*, only *L. monocytogenes* is considered a foodborne pathogen (13). The term “*Listeria* spp.” includes *L. monocytogenes* along with non-pathogenic species. Generally, non-pathogenic species of *Listeria*, in particular *L. innocua*, are found more frequently than *L. monocytogenes* in many processing plants, (7, 15, 16, 19) but this can be plant specific. In some circumstances and in some types of samples only a small fraction (< 5 to 10%) of samples positive for *Listeria* spp. are actually also positive for *L. monocytogenes* (14, 15, 19). On the other hand, in some situations the majority (> 70–80%) of the samples positive for *Listeria* species may also be positive for *L. monocytogenes* (14, 15, 19).

Although newer, rapid systems are becoming available, differentiation of *L. monocytogenes* from other *Listeria* species and specific detection of *L. monocytogenes* through the use of traditional cultural methods is generally time consuming, often requiring at least 7 days. In contrast, testing for *Listeria* spp., which would include *L. monocytogenes*, is less expensive and generally requires only 2 to 3 days. Most environmental testing programs in the US food industry use tests for *Listeria* spp. as an indicator for the potential presence of *L. monocytogenes*. However, some companies elect to test for *L. monocytogenes*, as this is the organism of concern. A *Listeria* spp.-positive sample should be interpreted as an indicator of *potential*, not presumptive, *L. monocytogenes* contamination. Depending on the location of the positive, it may be appropriate to determine whether a sample that is positive for *Listeria* spp. contains *L. monocytogenes* or to treat it as if it were *L. monocytogenes*. For finished product testing, it is generally more appropriate to test specifically for

L. monocytogenes rather than only for *Listeria* spp., unless product in which *Listeria* spp. is found is treated as if *L. monocytogenes* had been found.

Deciding what to monitor or test

A monitoring/testing program for smoked seafood may involve selecting and testing several different kinds of samples, including:

- Raw products,
- Non-food contact surfaces in the processing plant environment,
- Food contact surfaces, and
- Finished ready-to-eat products.

Some monitoring programs may not test all types of samples. A processor must always remember that the goal of testing is to find the organism if it is present, not to obtain “negative” test results.

Raw seafood testing

Research has shown that *L. monocytogenes* can be isolated from many of the types of raw fish commonly used for smoking (1, 4, 9, 14, 17, 18, 19). These raw products can be one source of *L. monocytogenes* contamination that is constantly being introduced into a plant. Many smoked seafood facilities receive raw products that have undergone some processing (heading and gutting, filleting). Contamination levels can be higher if the raw product is not handled properly during harvesting and primary processing. Testing for *Listeria* spp. or *L. monocytogenes* in raw seafood can help processors understand contamination sources associated with different species or types of raw products and monitor the performance of suppliers. Raw seafood testing is predominantly used in the production of cold smoked fish, as there is no processing step lethal to *L. monocytogenes*. Raw seafood found to be positive for *L. monocytogenes* should not be used to produce cold-smoked products unless the raw products can be treated to reduce the risk (e.g., chemical washing steps, freezing product, addition of growth inhibitors or combination treatments (12)).

Non-food contact surface testing

Research has shown that *L. monocytogenes* can frequently be isolated from various areas in the smoked fish processing plant environment and can persist in niches in certain areas of the plant (1, 9, 16, 18, 19, 22). These areas can include

floors, floor mats, walls, drains, tubs or totes, conveyances used to move product from one area of the plant to another, racks, cooler coils and condensate collectors, seams and crevices in processing machinery, and sponges, mops and other cleaning utensils. Each plant should determine appropriate environmental sites to sample, as well as appropriate sampling frequencies based on the potential for finished product contamination and based on knowledge of the specific operation and controls that are in place, along with any microbiological data available. Sampling locations can include the areas noted above, equipment support structures, structures over areas where product will be exposed, and, in particular, the wheels and vertical supports on racks. Sufficient samples should be taken to be representative of the plant environment. Testing non-food contact environmental surfaces can help processors understand contamination patterns, identify *L. monocytogenes* niches, and evaluate the effectiveness of sanitation procedures.

When conducting environmental testing of non-food contact surfaces, weekly sampling is recommended initially for most wet areas, where *L. monocytogenes* can grow; in dry-cleaned areas, sampling may be less frequent. The number of sampling locations and the frequency of sampling may be adjusted based on sampling results obtained over time. For example, repeated negative findings may suggest reducing the frequency of sampling in a particular area or elimination of a sampling site. When potential *L. monocytogenes* contamination problems are identified, the number of samples and sampling frequency may need to be increased to pinpoint contamination sources and then to demonstrate that the control measures used to eliminate *L. monocytogenes* were effective.

Non-food contact surface samples may be taken at different times during production: pre-operational (pre-op), during operation and at the end of the production shift, prior to cleanup. Companies need to consider what information can be obtained from each type of test when setting up the sampling program. Pre-op sampling reflects the efficacy of cleaning and sanitation, but it usually provides little information on sites that potentially harbor *L. monocytogenes*. Generally, sampling several hours into production allows time for *L. monocytogenes* to work its way out of any harborage sites in which it may be present and contaminate the environment, the processing line, and, potentially, product (20). Thus, sam-

pling during production or at the end of the shift prior to cleaning and sanitation can provide the best indication of the presence of *L. monocytogenes* in the processing environment and help processors identify new or persistent niches in equipment or the plant environment. Sampling drains is another means of determining the presence of *L. monocytogenes* in the processing environment, since drains can serve both as harborage sites and as a collection point for microorganisms in the plant when they get flushed to the drain during cleanup.

Data from non-food contact surface monitoring should be tracked over time to identify the need to take action and to identify trends that may not be obvious from a single day's monitoring (3, 20). Detection of *Listeria* spp. in an environmental monitoring sample does not necessarily indicate a microbiological control problem; however, it does indicate that additional investigation should be undertaken (21). Plants should determine the action to be taken in the event that *Listeria* spp. is detected at frequencies exceeding the upper control limit, target, or "trigger" that the plant has set (although attention should be given to cleaning and sanitizing an area when any positive is found). Because the reasons for a positive finding are likely to be plant-specific, actions taken in response to positives will vary. Consider the following points in determining remedial actions for non-food contact surface positives (21):

- When results indicate a trend toward an increased incidence of *Listeria* spp. in the environment, or repetitive positives in a particular area, plants should investigate to determine the reason(s) for the increase and should take action to reduce the level again.
- Additional samples should be taken from the environmental area where the positive was detected. These samples may indicate that additional actions are needed in this area.
- If, after a remedial action has been applied, additional samples are positive, the environment should be intensively cleaned and re-tested.
- Plants should consider the need to sample food contact surfaces in the areas where environmental positives are detected.

Floor drains, floors, and floor mats represent almost constant problem areas (19, 20, 21). A separate sampling program

with specific goals for each of these areas may be appropriate. Actions taken in response to a positive in these areas, especially drains, may also be less stringent than for positives in other areas in the environment; for example, while positives in these areas may result in additional sanitation, these areas may not need to be re-sampled daily until a specified number of negative samples is achieved.

Food contact surface testing

It is recommended that food contact surfaces be sampled routinely for *Listeria* spp. to verify that environmental and sanitation controls are preventing *L. monocytogenes* contamination of surfaces. Although some facilities choose to sample food contact surfaces only when monitoring of non-food contact surfaces suggests that there may be a problem, this approach is not recommended, since food contact surface contamination is not necessarily preceded by non-food contact surface contamination. In addition to routine testing, many processors conduct food contact surface sampling to verify the effectiveness of sanitation procedures used to solve specific problems or to eliminate persistent contamination sources identified by routine sampling of non-food contact surfaces.

Plants should determine the locations to sample, the time of day for sampling, and the frequency of sampling based upon knowledge of the specific operation and the controls in place, as well as any available microbiological data. When testing equipment, it is best to run the units for a period of time prior to swabbing/sponging, as the movement of parts and equipment vibrations may dislodge microorganisms from harborage sites. A pre-determined plan of action should be developed to address the finding of food contact surface positives. It is particularly important that plants investigate the reason(s) for all positives on food contact surfaces. Investigational sampling must be capable of identifying equipment that contains niches where *L. monocytogenes* has become established. Examples of steps that may need to be taken as a result of positives on food contact surfaces include modifying cleaning and sanitizing procedures, re-designing equipment, and re-training employees to improve adherence to Good Manufacturing Practices (GMPs) and other policies, practices and programs. Finding *Listeria* spp. on food contact surfaces may indicate the need for product testing for *L. monocytogenes*. Factors to be consid-

ered when making this decision would include whether there are other positive tests that suggest this is not a sporadic positive, the likelihood that the *Listeria* spp. would be *L. monocytogenes* (based on knowledge about the prevalence of *L. monocytogenes* and *Listeria* spp. in the specific facility), the likelihood of transfer from the food contact surface to product, and whether product storage, handling and use could increase the risk of illness if low levels of *L. monocytogenes* were present.

Finished product testing

Manufacturers periodically test finished products to verify that sanitation and other *L. monocytogenes* control measures (both prerequisite programs and HACCP controls, if implemented) are effective. Some manufacturers may use finished product testing as part of their product release program (“hold and test,” wherein product is held until test results are available). Many manufacturers conduct product testing at the request of their customers. Firms that have a solid environmental monitoring program (food contact surfaces and non-food contact surfaces) with appropriate remediation strategies may be able to convince customers that reducing the frequency of finished product tests would not compromise the safety of the product.

Because current US regulatory policies require that any lot of product in commerce that tests positive for *L. monocytogenes* be recalled from the market, it is imperative that each firm adequately define what constitutes a production lot when finished product testing is conducted. Further, the product lot sampled should be held until laboratory test results are available.

When product is sampled, representative samples should be collected from the lot. Sampling plans may be based on information from the International Commission on Microbiological Specifications for Foods (ICMSF) (11). ICMSF categorizes microbial hazards according to risk – moderate, serious and severe – and it ranks *L. monocytogenes* as either a serious hazard in foods for the general population or a severe hazard in foods for restricted populations (high risk groups, e.g., those in hospitals and nursing homes). ICMSF describes 15 different “cases” of sampling plans, with sampling plan stringency based on degree of risk and the effect on risk of the conditions of use of the product. With respect to *L. monocytogenes* in refrigerated smoked seafood

for the general population, case 12 could be applied (serious hazard that could increase since refrigerated storage would allow growth of *L. monocytogenes*); the sample size (n) would be 20, and c (number of units that could be positive) would be 0 (11). If product is to be held frozen, case 11 (n = 10, c = 0) could be applied (the risk does not increase since the product is held frozen.). If more stringent sampling is desired (e.g., product for nursing homes), sample size could be increased to 30, or even 60, samples. To reduce testing costs it may be possible, based on data for meat (R. Huffman, American Meat Institute Foundation, personal communication), to composite up to five samples (up to 125 g) for testing as a single unit without sacrificing sensitivity. It is highly recommended that, to minimize the chance of contaminating the samples, intact samples be sent to the laboratory and that, if any compositing is done, the laboratory do it. Samples from different lots should not be composited, since this could delay identification of which lots are contaminated when a positive occurs.

TESTING PROCEDURES

Sampling guidelines for *Listeria* testing

For environmental sampling, sponge samples are generally preferred to swab samples, as sponges can cover larger areas. However, swabs are useful to sample cracks and crevices that can serve as harborage sites for *L. monocytogenes*. When taking swab or sponge samples, a scientifically acceptable method should be used. Consistent sampling techniques should be used to ensure that results can be compared over time. It may be necessary for smoked seafood processors to get additional guidance or training on proper sampling techniques from a testing laboratory or from other food safety professionals. Product contact surface and non-product contact surface samples should be taken from an area as large as practical. Unless a processor is attempting to enumerate *L. monocytogenes* in a specific location (an expensive procedure not generally needed), a consistent-sized area need not be sampled.

Determining who will conduct the tests

Companies need to carefully assess whether the samples they collect will be tested at their own in-house facility or sent out to a contract laboratory. In most in-

stances the latter will be preferable, as this will eliminate the risk of the laboratory serving as a source of *L. monocytogenes* contamination for the plant. Special precautions must be taken if a laboratory that is located in a plant conducts pathogen testing. The laboratory may need to be completely separated from the plant, and control protocols will need to be implemented to ensure that people, sampling equipment, etc. do not carry pathogens from the laboratory to the plant. Actual costs for *Listeria* spp. and *L. monocytogenes* tests will depend on variables such as the amount and frequency of testing, test methods used, the sample collection supplies provided, and shipping costs. Before implementing a testing program, it is prudent for any company to discuss its testing needs with several laboratories to evaluate and determine which laboratory has the best price, services, and logistical arrangements to meet the company’s needs. However, primary consideration should be the laboratory’s capability to conduct accurate testing for *Listeria* using good laboratory practices and to handle the company’s volume of tests in a timely manner. Consideration may be given to the use of accredited laboratories (e.g., to ISO 17025), although this is not essential.

Actions taken based on sampling results

Firms should clearly recognize that the purpose of sampling and testing for *Listeria* spp. is to gather information that can be used to identify and eliminate potential sources of *L. monocytogenes* contamination. The goal of this testing is to find the organism if it is present so that the potential for contamination of the finished product can be minimized. Each firm should expect to find positives and determine, prior to starting such a testing program, the type of response or action that will be taken when test results are positive. The type of response will be different depending on whether tests are positive for *Listeria* spp. or *L. monocytogenes* and depending on the potential implications for finished product contamination.

For example, a firm that routinely monitors for the presence of *Listeria* spp. on non-food contact surfaces should decide on an appropriate “trigger” for further actions based on the number of positive test results and their location. Positives from non-food contact surfaces may trigger additional environmental testing, testing of food contact surfaces, and, in

some cases, testing of product. Positive tests for *Listeria* spp. do not necessarily indicate that finished products may be contaminated, but it may indicate that specific sanitation control measures to eliminate *Listeria* are not effective or are not being conducted properly or that personnel are not observing appropriate practices to minimize *L. monocytogenes* contamination. As noted previously, further investigation and sampling should be conducted to identify the contamination source and eliminate it. If environmental testing is conducted for *L. monocytogenes* instead of *Listeria* spp., processors will need to evaluate the source of any positive sample and determine the likelihood that product contact surfaces or finished products may have been contaminated. In addition to actions to eliminate *Listeria* at the site, more intensive sampling of the area may need to be conducted, as well as testing of product contact surfaces and possibly finished product(s). The finding of *Listeria* on food contact surfaces, particularly when there are multiple positives on a line, or after actions have been taken as the result of a positive sample, should be more likely to trigger product testing than the finding of a positive on a non-food-contact surface.

Problem solving

When an effective control program for *L. monocytogenes* is in place, finding multiple positives in the environment or product may indicate that the primary source of *L. monocytogenes* is a harborage site where the organism has become established and is multiplying. This can lead to line-specific contamination (21), in which the contamination will often flow downstream along a processing or packaging line. Mapping of the contamination sites on a layout of the area can assist in locating the source of contamination or, at least, suggest additional sites to sample (20). It is critical that the harborage site be found and eliminated. This usually means taking many samples of food contact surfaces along the line and in the adjacent environment. Line samples should be taken throughout the day (e.g., every 2 hours). To pinpoint the location of the harborage site, samples should be analyzed individually, not as composites. Suspected pieces of equipment should be torn down, and samples from suspicious sites or materials should be collected. Equipment should be cleaned and sanitized as it is being reassembled; the equipment should then be re-sampled. This is the preferred approach to finding *Listeria* on equipment surfaces and is usually ad-

equated to eliminate the contamination (20). However, if this process is unsuccessful, it may be necessary to remove sensitive electronics, oil and grease and to heat equipment surfaces to 160°F (71.1°C) for 20–30 minutes (5, 20). Lower temperatures for longer times may also be effective. Small parts can be placed in an oven or a hot water bath; larger equipment can be shrouded with a heat-resistant tarp and steam introduced under the tarp. It is also possible that employee practices may be a factor involved in contamination incidences. Refresher training in the controls necessary to prevent *L. monocytogenes* contamination may thus be indicated if repeat positive samples are found.

EXAMPLES OF LISTERIA MONITORING PROGRAMS IN SMOKED FISH PLANTS

The following examples describe four different hypothetical monitoring and testing programs for smoked fish operations to illustrate the guidelines provided above.

Example 1 (Company A) illustrates a program in a high volume plant that produces only cold-smoked salmon.

Example 2 (Company B) is a program in a medium-sized plant that produces 12 different types of hot- and cold-smoked fish.

Example 3 (Company C) describes a program in a small plant that produces 5 different types of hot smoked fish.

Example 4 (Company D) is a program in a medium-sized plant that uses a “zone” concept for its testing program.

It is important to keep in mind that these examples are provided for information purposes only. As noted previously, there is no one sampling or testing program that is appropriate for all smoked fish operations or even specific types or sizes of operations. The examples do not cover all possible scenarios that may arise during such testing programs. It is unlikely that any one of the examples will exactly match the unique conditions or procedures used in any particular plant. Rather, they are intended to help firms evaluate testing options and develop their own monitoring and testing programs as one component of a complete *Listeria* control plan. Cost estimates (which may be highly variable, depending on the number and types of tests conducted, logistics and testing method used) are included to help processors understand the costs that may be associated with various testing strategies.

EXAMPLE I

Company A produces a variety of cold-smoked salmon products for sale to retail stores, restaurants, catering companies, and institutional food service customers. Over 1 million pounds (approximately 450,000 kg) of headed and gutted (H&G) frozen salmon are purchased from 8 different suppliers in North America, South America and Europe each year. Frozen, brined salmon fillets are purchased from 2 large international suppliers. The plant operates all year and has 50 employees, all of whom work on a single shift (7 a.m. to 4 p.m.), except for 3 individuals who monitor the smokers in the evening and at night. The plant has a loading dock where raw products are delivered and stored in designated freezers. Raw product is thawed and prepared for brining in a raw material handling room. Products are brined in tubs in a cooler designated for brining. Brined products are placed on racks in the raw material room and moved into the smoking chambers. After the smoking cycle is completed, the smoked fish is moved to a designated finished product cooler. Smoked product is sliced and packaged in a finished product handling room. Company A has 3 slicing machines. Finished product is portioned and weighed by hand and then vacuum packed. Product is then stored at 36°F (2.2°C) for 2–3 days and shipped or is frozen until shipment. Company A has a sampling and testing program that includes routine testing of environmental (non-food contact) sites and product contact surfaces, periodic testing of finished product, and routine testing of raw seafood (Fig. 1).

Routine environmental testing

Each week Company A collects 12 samples from 6 different types of non-food contact sites in the exposed finished product handling area and tests them for *Listeria* spp. All swab or sponge samples are collected before processing. Two environmental samples are collected from each of the following five sampling sites: floors near the slicing machines; the wheels of carts used to transport in-process products and packaged products; coolers where smoked product is stored before it is packaged; the edges of and underneath tables where finished products are portioned and weighed; and underneath product conveyor belts. In addition, 2 samples are taken from floor drains, but results are treated differently (focused cleaning and testing only when there are positives). Test results are evaluated by tracking the total number of positives at each site over time. Whenever a

positive is detected (including in samples from drains), special attention is focused on cleaning and sanitizing that site. If 2 or more samples, including those from floor drains, are positive, or if the same site comes up positive two or more times in a month, extra attention is given to cleaning and sanitizing those sites. Except when the positives are from a floor drain, swab samples are then taken daily until the samples are negative for three consecutive days, and the routine weekly monitoring schedule is resumed. If there are any positive test results in 3 consecutive days, trouble-shooting procedures are implemented, which include shutting down lines in the affected area; using different sanitizers; more aggressive cleaning and application of sanitizer; use of heat sanitizing if necessary and feasible; or using other methods until there are 3 consecutive negative test results.

Product contact surface testing

Samples for *Listeria* testing are collected each week from 6 different product contact surfaces in the area of the plant where exposed finished products are handled and processed. A total of 12 different test sites on slicers, conveyor belts, scales, skinning machines and trim knives have been identified. Swab or sponge samples are taken at six of these sites each week so that all sites are sampled twice per month. Two of the monthly samplings are done before processing begins and two at mid-shift break, i.e., twice a month all the sample sites are tested before processing begins, and twice a month the sites are sampled at the mid-shift break. For machinery or equipment with moving parts, when pre-op samples are taken, the equipment is run for 15–30 minutes without product prior to sampling in order to dislodge any contamination from hidden, inaccessible areas. A pre-op positive from a piece of equipment suggests poor cleaning and sanitation or possibly persistent contamination (a harborage site). Re-sampling at selected sites using historical data to identify potential hot spots may help identify the contaminated area. When a product contact surface sample is positive, extra attention is given to the area, breaking the equipment down as necessary and cleaning and sanitizing this site. Pre-op samples from this site are then tested daily for 3 consecutive days. If results of at least 2 days of tests are negative, then routine sampling of the area is continued. If positives are found on 2 or more days during this 3-day testing period, the line is shut down; equipment is disassembled and thoroughly cleaned and sanitized with a different sanitizer than

the one routinely used. Swab samples are taken again before the line is put back into production and for 3 consecutive days after production has resumed. If tests are negative on two or more days, the routine sampling schedule is resumed. If positives still occur on two or more of these testing days, samples of finished product produced since the line was restarted are taken (20 packages, tested as 4 composites of 5 samples) and the corresponding lot of product is held until test results are obtained. Product that is negative is released. Product that tests positive must be destroyed or cooked, since reprocessing as cold smoked fish is not an option.

Finished product testing

Company A tests a random sample from a single lot of finished product once each quarter for *L. monocytogenes*. This company has determined that a lot is identified as a single type of product from one processing line produced during a specified period of time (usually a single day of production). Four composite samples, each consisting of 5 finished packaged products from a single lot, are collected. The lot from which the samples are taken is isolated until test results are obtained. The composite samples are tested for *L. monocytogenes*. If test results are negative, routine monitoring continues. If one or more of the samples test positive, the lot of product that the sample was taken from must be reprocessed into a hot-smoked product or fully cooked, or destroyed. Monitoring of product contact surfaces for *Listeria* spp. is then conducted daily for one week using the weekly testing protocol. If a positive test result is found, intensive sanitation procedures are conducted at the site. When test results are negative for three consecutive days, routine sampling is resumed.

Routine raw product testing and screening for new suppliers

The raw product testing and new supplier sampling program of Company A is as follows:

Screening new suppliers

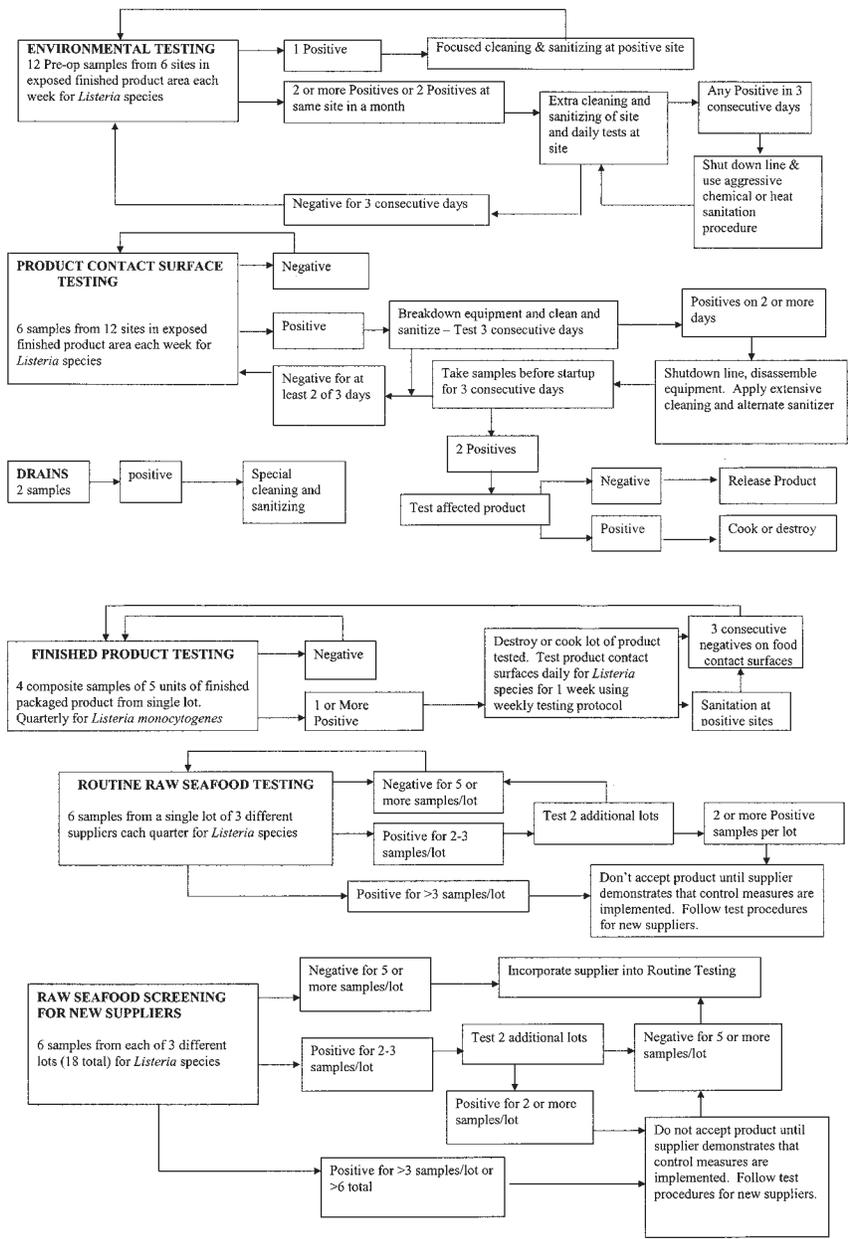
Company A has a policy requiring that samples of product be tested for *Listeria* contamination before the company establishes a business relationship with and accepts large shipments of frozen H&G salmon or brined fillets from any new supplier. This initial screening process requires that 6 samples from at least three different lots of product be tested

for *Listeria* species. If 5 or more of the samples from each lot of product are negative, the new supplier will be incorporated into Company A's routine raw material testing program. If more than 3 samples from any one of the three lots or more than 6 samples overall are positive, then Company A will not accept product from this supplier until they are able to demonstrate that effective *Listeria* control measures have been implemented and the screening process is repeated to confirm supplier controls are effective. If 2 to 3 tests from any of the initial lots of product are positive, additional samples are taken from two new lots of product from that supplier. If at least 5 or more of the samples from each of these additional lots are negative, the supplier can be incorporated into the routine raw material testing program. If 2 or more samples per lot from these additional tests are positive, Company A will not accept product from the supplier until it can demonstrate that effective *Listeria* control measures have been implemented and the screening process has been repeated and passed. Lots that test positive for *Listeria* spp. are returned to the supplier.

Routine raw product testing

Samples are taken randomly from 3 different suppliers on a quarterly basis. Six samples are taken from a single lot from each supplier, for a total of 18 routine raw material samples per quarter. Lots that test positive for *Listeria* spp. are returned to the supplier or sold to local restaurants to be used for cooked product. If 5 or more of the samples from a single supplier are negative, the supplier is returned to the routine testing schedule. If 2 to 3 samples from a supplier are positive, two new lots of product from this supplier will be tested. If 5 or more of the samples in each of the additional lots are negative, the supplier is returned to the routine testing schedule. If 2 or more of these additional tests are positive, Company A will notify the supplier of the problem and work with them to ensure that effective *Listeria* control measures are being used. When assurance is received that problem-solving measures have been implemented, Company A will then re-test the supplier. If more than 3 of the initial samples from a single supplier are positive, then Company A will notify the supplier of the problem and work with it to ensure that effective *Listeria* control measures are being used. When assurances have been received that problem-solving measures have been implemented, Company A will then re-test the supplier.

Figure 1. Flow Diagram for *Listeria* Testing – Company A (Example 1)



Testing program costs

Based on the sampling program outlined, Company A estimates that 1,248 environmental samples, 72 routine raw material samples, and 72 screening samples from four new suppliers will be tested per year for *Listeria* species. This represents approximately 1,400 routine *Listeria* species tests annually. At a cost of \$25 per test, the annual cost would be approximately \$35,000. In addition, Company A estimates that up to 75 additional tests will be needed to solve problems when occasional test results are positive. The cost of these additional tests at \$25 per test would be \$1,875. Finally, 16 finished product composite samples will be tested for *L. monocytogenes*. At a cost of

\$30 per test, the annual cost would be \$480. Additionally the company budgets for 8 product composites to be tested in the event of product contact surfaces testing positive (\$240). Based on these estimates, Company A determined that it must budget an additional \$40,000 annually in operating expenses specifically for the *Listeria* testing program outlined in this example.

EXAMPLE 2

Company B annually produces approximately 400,000 pounds (~181,400 kg) of smoked fish products that are sold to retail stores, delicatessens and restaurants. Approximately 40% of the finished prod-

uct is vacuum packed cold-smoked salmon, 15% is vacuum-packed sablefish and the remainder is air packed hot-smoked products such as trout, whitefish, mackerel, salmon, bluefish and eels. Salmon is received in the H&G frozen form from suppliers in North and South America. Sablefish is obtained frozen from suppliers in the Pacific Northwest. Frozen trout fillets are received from domestic suppliers, and all other products are received as fresh whole fish from suppliers in the US and Canada. The plant operates all year and has 26 employees. There is one shift from 8:00 a.m. to 5:00 p.m., with 2 employees managing the smoking operation and 3 employees assigned to sanitation duties conducted in the evening and at night.

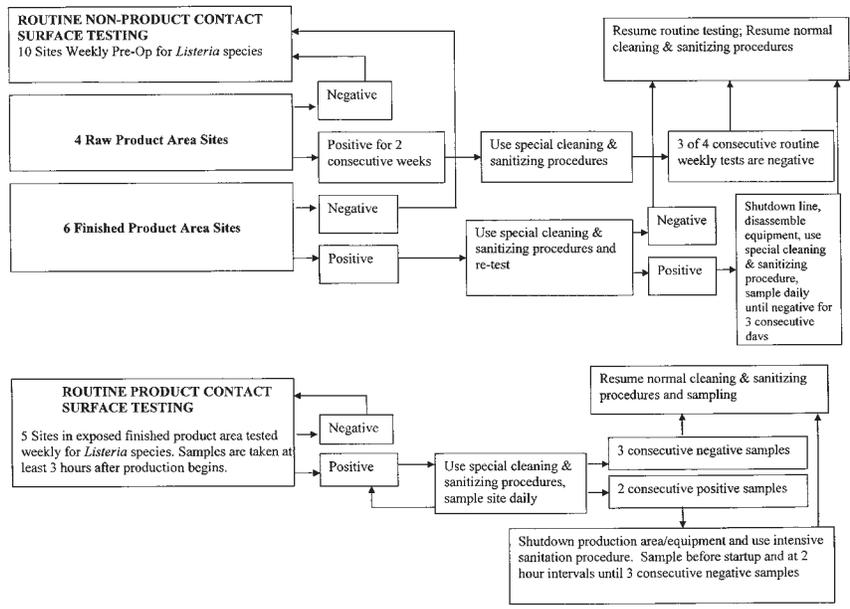
Raw fresh fish are received and stored in a designated cooler before processing. Frozen salmon and trout are delivered daily from a nearby storage warehouse to meet daily production needs. All raw products are handled and processed in a raw production room. One side of the room is designated for thawing frozen products and filleting and trimming products to be cold smoked, and the other side of the room is used for cleaning and preparing whole fish for brining and hot smoking. All products are wet brined and stored in a designated brining cooler. After smoking, all finished products are stored in a designated finished product cooler. Finished products are trimmed, sliced (if necessary), and packed in a separate packing room. Two slicers are used to prepare cold-smoked salmon. All individual packages are portioned and weighed by hand by plant workers and either vacuum or air packed. Finished products are stored in a refrigerated cooler set at 32°F for orders that will be shipped within one week. Some product will be frozen for longer-term storage.

Company B tests non-product contact surfaces and product contact surfaces for *Listeria* spp. (Fig. 2). The company assumes that, given the mixture of raw products used in the plant, all species of seafood may contain *Listeria*, and it has implemented an aggressive routine sanitation program in both the raw and finished product handling areas to control the organism. Products to be cold smoked are treated with an alkaline treatment (lime solution, pH 12) to reduce *Listeria* contamination levels (12).

Non-Product contact surface testing

Company B monitors 10 different environmental non-food contact sites in its plant on a weekly basis. Six sites are tested

Figure 2. Flow Diagram for *Listeria* Testing – Company B (Example 2)



in the finished product area and 4 sites are tested in the raw material handling area. Pre-op sponge samples are taken in the finished product area as follows: 1 sample on a slicing machine from an area that does not directly touch product; 2 samples from the edges and underneath work stations where product is packed and weighed; 2 employee contact surfaces such as the door handle to finished product coolers, sites on the slicer or skinner or a knife handle; and 1 sample from wheels or surfaces of carts used to move finished product. Sites in the raw product room include 1 sample from each of the following areas: the edges and underneath tables used to prepare raw products; the floor of the brining cooler; the raw product cooler; and the frame of a smoker rack. All samples are taken before production begins.

Test results are monitored over time. While raw-material areas are expected to have a higher frequency of positive samples, they represent a lower risk for contributing to finished product contamination. Finished product areas are expected to have a lower frequency of positive results, but these areas pose a greater risk for finished product contamination. Tests in the raw material area are used to monitor patterns of contamination. Sites that have a positive result for 2 consecutive weeks will receive a more stringent cleaning and sanitizing procedure, along with sanitizer rotation until at least 3 of 4 consecutive tests are negative, at which time the normal cleaning and sanitizing

procedures will be resumed. When a positive is detected in the finished product area, the sanitation crew is immediately notified and that site will receive special attention in the cleaning and sanitation protocol until the results are available from the re-test of this site, which is done within 24 hours of finding a positive. If the re-test result is negative, normal procedures are resumed. If the re-test result is positive, a supervisor will shut down the line, if necessary, and ensure that additional procedures for sanitation and equipment disassembly are implemented. Daily sampling of this site will occur until 3 consecutive negative samples demonstrate that the contamination source has been eliminated, and then the normal cleaning and sampling routine will be resumed.

Product contact surface testing

Five product contact surfaces are tested weekly in the finished product handling area only. A swab or sponge sample is taken from the following areas: the blade of a slicing machine, a scale used to weigh product before packaging, a conveyor belt (skinning machine belt or packaging belt), a trimming knife, and one of the totes or racks used to move cold smoked products. All samples are taken at least 3 hours after processing has started. If a sample is positive, intensive cleaning and sanitizing procedures are focused on that area and samples are taken daily from the positive site. This process continues if there are any positives. When 3 consecutive

days of negative samples are obtained, normal cleaning and sanitizing and sampling procedures are resumed. If samples are positive on 2 consecutive days, the area is shut down and extensive sanitation procedures are implemented. Swabs are taken before start-up and at two-hour intervals until negative samples for 3 consecutive days demonstrate that the contamination source has been eliminated, after which routine testing is resumed.

Finished product testing and raw material testing

Company B does not perform any routine testing of finished products or raw products.

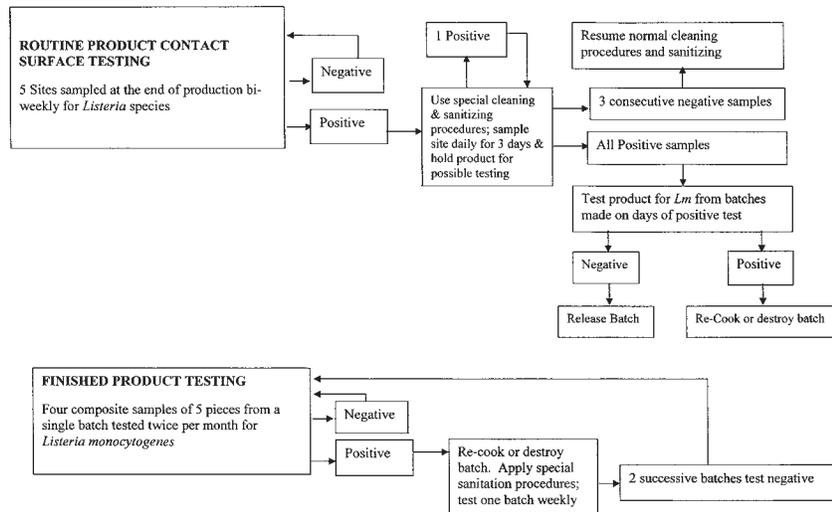
Testing program costs

Based on the sampling program outlined above, Company B estimates that 780 environmental samples will be tested per year for *Listeria* species. At a cost of \$25 per test, the annual cost would be approximately \$19,500. In addition, Company B estimates that up to 50 additional tests will be needed to solve problems when occasional test results are positive. The cost of these additional tests at \$25 per test would be \$1,250. Based on these estimates, Company B has determined that it must budget an additional \$21,000 annually in operating expenses specifically for the *Listeria* testing program outlined in this example.

EXAMPLE 3

Company C is a small “boutique” processor that produces 10,000 to 20,000 pounds (approximately 4,500 to 9,000 kg) annually of hot smoked fish products for sale to area retail stores, restaurants and catering operations. The primary products are hot smoked salmon, trout, eel, bluefish and mackerel; fish are purchased fresh from local fishermen or wholesalers. The plant is a single large room with 4 employees. Production occurs daily from May to October, and a single batch of product is smoked each day. One to three batches are smoked per week during the remaining months of the year. Raw products are prepared in a designated area for brining. Because space and equipment constraints do not allow complete separation, all products (raw fish, products being brined, and finished products) are stored in the same cooler (which presents a higher risk of recontamination of finished product). All finished products are placed in open plastic containers after smoking.

Figure 3. Flow Diagram for *Listeria* Testing – Company C (Example 3)



After an initial 8-hour cooldown period, lids are placed on finished product containers during storage to minimize the potential for cross contamination. Customer orders are assembled just prior to delivery, and all finished products are air packed.

Company C has a monitoring program that involves testing product contact surfaces, with periodic finished product testing (Fig. 3). All of the products produced by this firm undergo a step that requires the internal product temperature to reach a minimum of 145°F (62.8°C) for 30 minutes, which is lethal to *L. monocytogenes*. For this reason the primary concern for this firm is post-processing contamination of finished products from the plant environment, and the firm does not test raw products. Testing product contact surfaces is used to demonstrate that the firm's *L. monocytogenes* control measures are effective. Periodic finished product testing is used for further confirmation of the effectiveness of these control measures.

Product contact surface testing

Company C swabs 5 different product contact surfaces on a bi-weekly basis and has them tested for *Listeria* spp. Swab or sponge samples are taken at the end of production, prior to cleaning and sanitizing, at the following sampling sites (one sample per site): the table used to pack orders, two different cutting boards used to trim or cut product into portion sizes, one of the containers used to store

smoked products, and the scale used to weigh customer orders. If a positive test result is obtained, the affected site or equipment is thoroughly cleaned and sanitized using intensive procedures. Daily swabs are taken for 3 days. This process continues if there are any positives. When 3 consecutive days of tests are negative, routine sampling and cleaning and sanitizing procedures are resumed. Finished product from the batch produced when the sample was taken is held and tested for *L. monocytogenes* if a product contact surface is positive; consequently, whenever product contact surfaces are tested product is placed on hold. If tests are negative the product is released. If tests are positive the lot is destroyed or re-processed with a full cook reaching a minimum internal temperature of 145°F (62.8°C) for 30 minutes.

Finished product testing

Four composite samples consisting of five different 25-g pieces from a single batch of finished product is tested twice each month for *L. monocytogenes*. A lot is comprised of a single batch of product smoked in the processor's single smokehouse. The lot is held until test results are obtained. Additional lots produced at the same time may also be tested. If any product test is positive, the product is destroyed or re-cooked if possible through the full cycle to ensure that it reaches a minimum internal temperature of 145°F (62.8°C) for 30 minutes, and special sanitation procedures are used until 2 successive batches test negative.

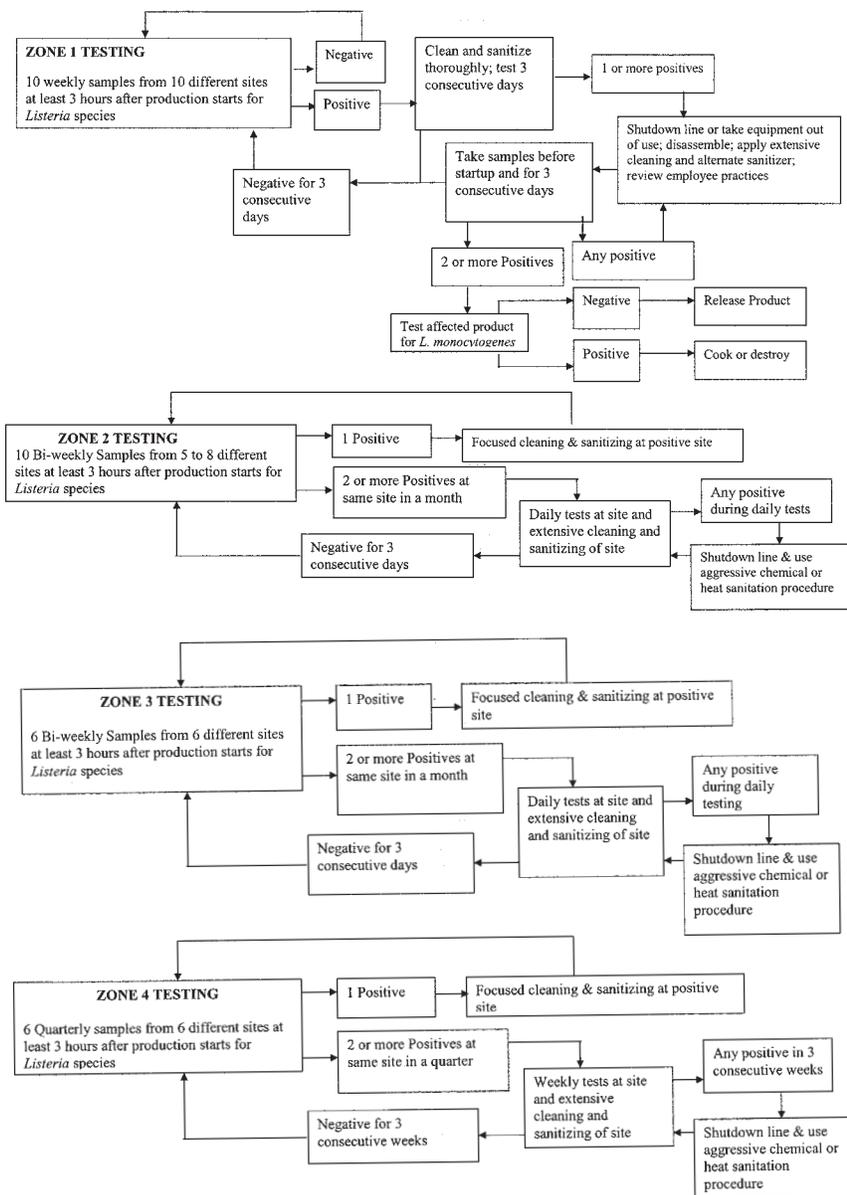
Testing program costs

Based on the sampling program outlined above, Company C estimates that 130 environmental samples will be tested per year for *Listeria* species. At a cost of \$25 per test, the annual cost would be approximately \$3,250. In addition, Company C estimates that up to 20 additional tests will be needed to solve problems when occasional test results are positive. The cost of these additional tests at \$25 per test would be \$500. Costs for finished product testing when food contact surfaces are positive are estimated to be \$600. Company C also will test 96 (8 composites per month × 12 months) routine finished product samples per year for *L. monocytogenes*. The cost of these finished product tests at \$30 per test would be \$2,880. Company C has also budgeted for additional testing if product were to test positive. They estimate that this might happen twice a year, requiring 4 additional tests of product composites for a cost of \$120. Based on these estimates, Company C has determined that it must budget an additional \$7,350 annually in operating expenses specifically for the *Listeria* testing program outlined in this example.

EXAMPLE 4

Company D produces cold-smoked salmon and a variety of different hot smoked ready-to-eat products for sale to retail stores, restaurants and commissary operations. The primary raw material used in the plant is frozen H&G salmon and brined salmon fillets from suppliers in North and South America. Trout is purchased from aquaculture suppliers in the US and Canada; raw products for other specialty items are both wild caught and farm raised. The plant operates year round and has 50 employees, all of whom work on a single shift, except for the cleaning crew and the smokehouse operators. Whole salmon and fillets are stored in a frozen storage warehouse and delivered to the plant to meet production needs. Other raw products are stored either in the in-plant freezer or a raw material cooler. Frozen products are thawed and prepared for brining in a raw material handling area. From there, product moves into an in-process area where brine is prepared and fish are rinsed after brining and loaded onto racks for smoking. After smoking, the finished product is moved to a designated cooler for holding. Smoked product is then moved into a finished product handling and packing room

Figure 4. Flow Diagram for *Listeria* Testing – Company D (Example 4)



where the product is trimmed, sliced, portioned and packed. Finished vacuum- and air-packed product is either stored at 36°F (2.2°C) or frozen until orders are packed and product is shipped to customers.

Company D has implemented an environmental *Listeria* testing program that divides plant operations into four different zones that were identified by evaluating the relative potential risk that they represent in terms of possible direct finished product contamination. Zone 1 includes all direct product contact surfaces in the finished product handling area that could harbor *Listeria* and directly contaminate finished product, including equipment such as slicers, skinners, trimming

knives, scales, work tables, conveyor belts, carts, racks, totes used to transport finished product, and employee hands or gloves. Zone 2 includes non-food-contact surfaces in the finished product handling area in close proximity to product contact surfaces that could indirectly contaminate food contact surfaces or finished products, such as the exterior of equipment, floors, stress mats, cart wheels, metal framework, coolers where finished product is stored, drains, employee aprons, and shoes. Zone 3 includes product contact surfaces in the in-process areas of the plant that could harbor *Listeria*, including fillet tables and knives, smokehouses, brine tubs, brining coolers, smoker racks, and employee aprons, as

well as drains in the in-process area. Zone 4 includes those areas that are remote from the finished product handling areas, such as raw material storage coolers, thawing tubs, storage areas for ingredients and packaging materials, and staging areas. Company D's environmental *Listeria* testing program identifies how and when testing will occur and appropriate responses to test results for each plant zone (Fig. 4).

Zone 1

Company D collects a single swab or sponge sample from each of 10 different sites in Zone 1 weekly and tests them for *Listeria* spp. Equipment samples from slicer blades, skinning machines, etc. are taken after at least three hours of production and up until the end of the day's production to "shake-out" any potential contamination that may not have been eliminated from the previous day's cleaning and sanitizing activities (due to a harbor-age site) as well as to pick up contamination that occurs during production. Sites included in each weekly sample collection include at least 2 samples from slicer blades, 1 sample from the skinning machine, 2 samples from work tables and/or conveyor belts, 1 sample from a scale, 1 sample from a randomly selected employee's hands, 1 sample from a trimming knife, and 2 samples from carts, totes, or racks used to transport exposed finished products. If a sample is positive (other than an employee's hands or a trimming knife), special attention is devoted to cleaning and sanitizing procedures and the site is re-tested for 3 consecutive days. If the site is negative for 3 consecutive days, routine testing at that site is resumed. If results of any tests are positive tests, the equipment or line will be shut down and intensive cleaning and sanitizing procedures will be applied, including disassembly of the slicer or skinning machine, if positive, and heat or chemical sterilization if possible. An additional sample is then taken before startup and again for three consecutive days, holding product produced on the line those days, until 3 consecutive negative samples are obtained. If any positive is found, sanitation and test procedures continue, with more aggressive cleaning and sanitation and more extensive sampling in the area to determine the root cause of the positive. If 2 or more additional positive samples are found during the 3 days of testing, the lot of product produced on that line or piece of equipment is tested for *L. monocytogenes*. If test results are negative, product can be released and intensive cleaning and sanitizing procedures

and daily testing are reapplied until consecutive negative results for three days are found. If the product test for *L. monocytogenes* is positive, the isolated lot is destroyed or cooked or hot smoked to a minimum internal temperature of 145°F (62.8°C) for at least 30 minutes. If a trimming knife is positive, employee practices are reviewed and revised as needed and employee refresher training is conducted; the type of sanitizer used for trim knives may be changed. If an employee's hand or gloves tests positive, a supervisor will review company hand washing and personal hygiene policies at the work site and re-test the same employee the following week.

Zone 2

Company D collects 10 samples every two weeks from 5 to 8 different non-food contact surfaces in the finished product handling area. Swab or sponge samples are collected during production and tested for *Listeria* species. Sample sites include 2 samples from non-food contact sites on equipment used for finished product such as slicers, packaging equipment etc.; 1 sample from metal framework of work tables or packaging equipment; 1 sample from stress mats or the floor near slicers; 1 sample from an employee apron or shoes; 1 sample from the wheels of carts used to transport exposed finished product; 1 sample from the cooler used to store exposed finished product; and 1 drain sample. If a site tests positive, focused cleaning and sanitizing procedures are used at this site until the results of the next scheduled test are obtained. If this subsequent test result is negative, routine procedures are resumed. If 2 positive samples at the same site are obtained in the same month, intensive cleaning and sanitizing procedures are implemented at this site and, except when the positive samples are from a drain, daily tests are conducted. If test results are negative for 3 consecutive days, routine sanitation and testing procedures are resumed. If any test is positive during this daily testing, the line is shut down and heat or intensive chemical sanitation procedures are applied until daily tests are negative for 3 consecutive days.

Zone 3

Company D collects 6 samples every two weeks from 6 different sites in this zone. Swab or sponge samples are collected after at least three hours of production and tested for *Listeria* species.

Sample sites include 1 sample from a fillet table; 1 sample from a brine tub; 1 sample from a drain; 1 sample from the brining cooler; 1–2 samples from smoker racks or the smokehouse; and 1 sample from an employee apron or gloves. The same protocol for responding to positive samples described for Zone 2 is used for Zone 3.

Zone 4

Company D collects 6 samples quarterly from 6 different sites in this zone. Swab or sponge samples are collected at the same time samples are being taken from other zones and tested for *Listeria* species. Sample sites include 1 sample from raw material storage cooler; 1 sample from empty raw material thawing tubs; 1 sample from drains in the thawing area; 1 sample from empty tubs or totes used to move thawed product into the in-process area; 1 sample from wheels of carts used to move product into the in-process area; and 1 sample from a bathroom door. A protocol similar to that described for Zones 2 and 3 is used to respond to positive samples from Zone 4. However, in Zone 4, samples are taken quarterly rather than every two weeks, and the re-sampling frequency for responding to a positive test result is weekly rather than daily.

Raw and finished product testing

Raw products are treated with an alkaline treatment (lime solution, pH 12) to reduce *Listeria* contamination levels (12), and no raw product or supplier testing is conducted. Company D does not conduct any routine finished product testing; such testing may be conducted in conjunction with Zone 1 positives, as noted above.

Testing program costs

Based on the sampling program outlined above, Company D estimates that 520 samples will be tested per year for *Listeria* species in Zone 1; 260 samples in Zone 2; 156 samples in Zone 3 and 24 samples in Zone 4. The total number of samples tested for *Listeria* species per year is 960. At a cost of \$25 per test, the annual cost would be \$24,000. In addition, Company D estimates that up to 60 additional tests will be needed to solve problems when occasional test results are positive. The cost of these additional tests at \$25 per test would be \$1,500. The company does not anticipate the need to test product; however, it includes \$1000 in the budget as a contingency. Based on these estimates, Company D has determined that it must budget an additional \$26,500

annually in operating expenses specifically for the *Listeria* testing program outlined in this example.

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