Establishing enhanced *Listeria* control: Coupling environmental monitoring with risk-based product testing

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**Abstract.** Prevention and control of *Listeria monocytogenes* remains a challenge in food manufacturing facilities and methods adopted vary across different production systems and food categories. Regulatory policies also vary from region to region, although there is a convergence across the world towards risk-based approaches. Given these inconsistencies, the objective of this commentary is to reiterate two fundamentally critical components of *Listeria* control and prevention, and the potential benefits of actively coupling food contact surface testing and risk-based product testing programs.

1. Introduction

Foodborne illness associated with *Listeria monocytogenes* remain a significant global public health burden. Outbreaks of Listeriosis resulting from consumption of food contaminated with *L. monocytogenes* are related to a diverse range of products. Significantly, susceptible populations such as the elderly (>65 years), neonates and pregnant women, and immunocompromised individuals and those suffering from chronic disease conditions (cancer, diabetes, cardiovascular problems, etc.) account for majority of cases [1, 2, 3, 4]. Consequently, disease incidence is typified by high mortality rates of up to 30% [1,5]. Efforts to impact disease reduction have been targeted at various stages in the farm to fork supply chain, yet incidence of invasive listeriosis remains high across the world.

Unlike other pathogens, Listeria thrives in cool and moist environments and is problematic in foods stored at refrigeration temperatures. Unsurprisingly listeriosis outbreaks are predominantly associated with high-risk foods that are stored at these temperatures. Farber et al. noted major Listeriosis events over the past decade resulted predominantly due to contaminated ready-to-eat (RTE) foods considered to be high risk (which support growth of the pathogen) such as sliced delicatessen meats and soft cheeses [6]. More recently listeriosis cases and outbreaks have also been identified with low-risk foods (RTE foods which do not support growth of the pathogen), such as frozen foods (ice cream, frozen vegetables, frozen chicken products, etc.) [7, 8, 9, 10]. The term low-risk is ascribed to frozen foods for example due to their storage temperature state, conditions which do not support the growth of the pathogen. It should be emphasized invasive listeriosis is primarily problematic with the high-risk category of foods, quantitative modeling shows greater than 90% of these cases are caused by ingestion of RTE food containing >2000 colony forming units (CFU)/g [11]. Foods stored in vacuum or in modified atmospheric packaging with extended shelf life in refrigeration, allow growth of the pathogen to high levels during shelf life. The distinction of whether a food supports or does not support growth of the pathogen in RTE foods is thus at the center of developing a risk profile and importantly at the way both
the industry can implement management practices and how regulatory policies are framed and enforced across the world.

As consumer eating habits and diets evolve to comprise more RTE foods, there is also a growing concern of listeriosis risk with these foods and most global regulatory policies continue to target the presence of *L. monocytogenes* in RTE foods with zero tolerance for foods that support growth. Codex, European Union, Canada, Japan all employ a risk-based tolerance of 100 CFU/g with respect to RTE foods that do not support growth [11, 12, 13, 14, 15]. Current scientific information and data relative to the nature of the pathogen, potential growth in food, and public health impact (illnesses and outbreaks) indicate *L. monocytogenes* is a remote public health risk in low-risk foods such as frozen foods. Even still, U.S. regulatory policies (both U.S. Food and Drug Administration (FDA) and Food Safety Inspection Service (FSIS)) broadly enforce a ‘zero’ regulatory action limit for the presence of the hazard, *L. monocytogenes* across all foods without relying on a risk-based distinction [16, 17].

### 2. Current U.S. regulatory policies for meat and poultry products

FSIS, the agency under U.S. Department of Agriculture which oversees all food safety regulations relevant to meat and poultry products, enforces its *Listeria* rule across all RTE foods (reference) and provides specific recommendations to prevent cross contamination of post-lethality exposed RTE meats and poultry products. However, the rule also explicitly precludes Not-RTE food products with on-package validated cooking instructions.

Importantly, under FSIS regulations, RTE foods exposed to the post-lethality environment should implement robust sanitation programs, appropriate hygienic equipment design principles, pathogen testing regimen, and other pre-requisite programs including Good Manufacturing Practices (GMPs) as part of the food safety system. Adulteration is defined either by the presence of *L. monocytogenes* in the RTE food (contaminated food) or as resulting from food coming into direct contact with a surface that is contaminated with the pathogen. The FSIS *Listeria* regulation delineates three alternative methods to control *L. monocytogenes* contamination of post-lethality exposed RTE foods.

- **Alternative 1:** Use of a post-lethality treatment to reduce or eliminate *L. monocytogenes* and antimicrobial agent or antimicrobial process to suppress or limit the growth of *L. monocytogenes*.
- **Alternative 2:** Use of a post-lethality treatment or an antimicrobial agent or process to reduce, eliminate or prevent the growth of *L. monocytogenes*.
- **Alternative 3:** Do not use a post-lethality treatment or antimicrobial agent or process, instead relies on sanitation alone to control *L. monocytogenes* in their post-processing environment (sanitation controls)

A variety of validated post-lethality treatments are noted by FSIS and include steam and hot water pasteurization, radiant heating, high pressure processing, ultraviolet and infrared treatments, and drying to achieve low water activity. The agency expects a post-lethality treatment is designed to achieve at least a 1-log lethality of *L. monocytogenes* before the product exits the production facility.

Antimicrobial processes and agents used to suppress or limit growth of *L. monocytogenes* should be designed to achieve no more than 2-logs of growth over the shelf-life of the food. Antimicrobial agents include modified atmosphere packaging, addition of salt, organic acids and other additives, curing with nitrates, addition of growth inhibitors such as lactates and diacetates, which may be added to the formulation, finished food product or packaging material as appropriate. Common antimicrobial processes include fermentation, drying and freezing that are supported by studies documenting the effectiveness of these processes in limiting or suppressing growth during shelf-life.

No doubt a combination of the above strategies is encouraged to be used through the hurdle concept, including appropriate use of pH, water activity, moisture-protein ratio, and storage temperature to reduce the level of the pathogen during processing and to limit growth during shelf-life.

As stated above a third alternative is to utilize sanitation controls alone where a post-lethality treatment, antimicrobial process or agent are not applicable to control *L. monocytogenes* in the post-processing environment. A sanitation standard operating procedure may be incorporated as a pre-requisite program including the ability to escalate sanitation procedures to counter repeat positive
findings of the pathogen or their indicator microorganisms. Its important to note, regardless of which alternative is applied, maintenance of a robust sanitation program and microbial testing of food contact surfaces for *L. monocytogenes* or an indicator microorganism to verify effectiveness of these activities is expected.

3. Current industry *Listeria* management practices

As prevalence of *L. monocytogenes* in the food supply continues to present complexities, its critical for food manufacturers to rethink their *Listeria* prevention and control management strategies to drive down *Listeriosis* incidence across all food categories.

Consider the fact that unlike most other foodborne pathogens, *L. monocytogenes* is essentially an environmental pathogen that finds entry and the potential for harborage in manufacturing milieu through for instance, incoming raw materials, movement of personnel and vehicles, and the uncontrolled use of water. These conditions result not only in niches that support growth of the pathogen but also serve as transfer points to spread contamination across the facility and ultimately to food. Conceptually, measures to mitigate the presence of *L. monocytogenes* in foods can broadly be placed in to four important segments: 1) Formulation: use of low pH, low water activity, antimicrobial agents, etc.; 2) Process: use of heat and other lethality treatments; 3) Facility: use of robust sanitation and environmental monitoring programs; 4) Food: Modified atmospheric packaging and storage temperature. Each of these approaches can be effective yet none alone achieves the level of control needed to address the risks typified by the dynamic nature of contamination and occurrence of *L. monocytogenes* in food production environments. In addition, as *Listeria* is everywhere, we need everyone involved in food manufacturing to be at the forefront of pathogen management, and this in turn needs increased awareness of *Listeria* risks through appropriate education and training regimen for personnel.

4. Addressing potential post-lethality contamination in the facility

Among the most frequent causal regions in the facility that engender cross-contamination is the post-lethality environment where food may be exposed prior to packaging. In this discussion, concepts related to environmental monitoring and product sampling will be used to increase the likelihood of identifying potential food contamination during processing and decrease the likelihood of contaminated food from entering commerce.

5. Environmental monitoring and food contact surface testing

Food safety programs to assess growth and harborage of *L. monocytogenes* in a food manufacturing facility have largely revolved around swabbing of surfaces for different microorganisms, particularly indicator microorganisms to determine sanitation efficiency and potential pathogen contamination. As it may be challenging to monitor the entire production environment constantly, care should be taken to select the appropriate monitoring sites that are likely to harbor the pathogen and serve as transfer points for cross contamination. For instance, environmental monitoring of the post-lethality environment is critical in identifying and controlling *L. monocytogenes* in facilities involved with the production of RTE food.

An effective environmental monitoring plan should adopt the ‘seek and destroy’ approach wherein positive indicator findings are deemed as rationale for continuous learning and development of cleaning and sanitation programs. [18] Microbial testing methods that assist with the environmental monitoring program should be directed toward an indicator microorganism such as *Listeria* spp. Seeking out the presence of *Listeria* spp. instead of *L. monocytogenes* is advantageous because they are non-pathogenic and generally found more frequently in the environment than the pathogen. This approach provides a path to identifying growth niches before *L. monocytogenes* can find harborage in the facility.

The challenges of evolving and administering a mature environmental program that can identity harborage of *L. monocytogenes* in a timely manner and take corrective actions rests on good knowledge of the pathogen, the facility and its people and operational footprint. Testing non-food contact surfaces (non-FCS) can provide a good start to a nascent environmental program, and these steps should develop in to both higher frequency activities and cover more relevant regions in the facility where risk is elevated, such as food contact surfaces (FCS). Positive FCS findings reveal both a breakdown in the sanitation system as well as a potential transfer from other areas in the facility. They should be followed
with recleaning, re-sanitizing steps, and retesting procedures to ensure safe production. In some cases, a complete overhaul of these procedures may be warranted including reassessing chemical concentrations, personnel behaviors and tools used for these activities. If positive FCS results are sustained even after these interventions, then root cause analysis steps must be undertaken to provide a more comprehensive assessment of existing risks in the facility.

The criticality of sampling and testing post-lethality regions of the facility and their FCS cannot be understated. Niches or locations in the facility where *Listeria* *spp.* are found even after cleaning and sanitation have been applied may correspond to transfer points of the pathogen from these areas which lead to persistent product contamination. Indeed *L. monocytogenes* harborage can form anywhere in the facility, but generally, they occur in areas that are not easily identified or accessible for cleaning and thus not controlled. If not identified and addressed in a timely manner, the pathogen slowly migrates from their niches to outer surfaces of the equipment, reaching FCS and contaminating food products.

The presence of any transient *Lm* can be eliminated by following good manufacturing practices (GMPs), effective cleaning and sanitation and a robust environmental program. This approach must include FCS sampling and testing to ensure cross contamination is avoided within the facility and to product. Furthermore, facilities should administer goals that will reduce the number of *Listeria* *spp.* positives and steps toward continuous improvement of sanitation programs.

**6. Risk-based finished product sampling and testing**

First and foremost, it should be recognized that depending on the sampling plan and test method, product testing may demonstrate the presence or absence of *L. monocytogenes* in food, but it is not a reliable indicator of the prevalence of the pathogen in a facility. No doubt, food contaminated with *L. monocytogenes* is difficult to assess as it is unlikely to be uniformly distributed throughout the product.

Farber et al. in their recent review of *L. monocytogenes* science recommended that low-risk foods with a regulatory tolerance for the pathogen may incorporate specific end-product verification testing using novel three-class sampling plans as an alternative to existing two-class presence/absence sampling plans [6]. The authors demonstrate some three-class sampling possess higher test performance in detection of the pathogen such that it may serve as a “warning management indicator.” The benefit of enumeration (for instance, a quantitative microbiological limit of less than 100 CFU/g) combined with detection (for instance, a qualitative detection limit of 1 out of 4 analytical units of 25g each) include establishing that product contamination is not widespread (only one positive) and not at a very high level (less than 100 CFU/g) of the pathogen in the food.

Of course, any product testing program must follow test and hold procedures to avoid potential food withdrawals or recalls resulting from positive findings among tested samples. Whether testing of products is instituted in a limited way or as a routine measure, utilizing scientifically validated high performance plans can only further generation of more data and inform more accurate risk assessments related to the presence of *L. monocytogenes* in foods.

**7. Coupling environmental monitoring and product testing**

Historically, product testing was viewed as a futile exercise with many risks for food processor companies, yet many industry supply chain programs incorporate product testing requirements while circumventing the need for carrying out robust environmental monitoring programs. As discussed above, implementing both programs present challenges for food manufacturers and consequently, neither reliable product testing nor adequate environmental monitoring are used to effectively prevent and control the presence of *L. monocytogenes* in the global food supply.

Decades of experience shows the presence of transient *L. monocytogenes* can be eliminated by following good manufacturing practices (GMPs), effective cleaning and sanitation, and robust environmental monitoring programs. The ability to track and eliminate more persistent pathogenic strains and undercut the potential for food contamination requires greater rigor in a facility’s food safety verification system. An environmental monitoring program with FCS testing coupled with a scientifically valid product sampling and testing regimen are reliable and needed anchors for the overall food safety system targeting the presence of this ubiquitous pathogen.
8. Conclusion

Even systematic approaches such as good facility hygiene and the ‘seek and destroy’ process continue to challenge persistence of L. monocytogenes in processing facilities, especially in the post-processing environment. Addressing these issues while complying with regulatory zero-tolerance for the presence of the pathogen in food and food contact surfaces is a demanding effort. However, risk-based regulatory approaches can engender effective risk management practices aimed at controlling L. monocytogenes. Specifically, a strategy to link food contact surface testing and risk-based testing programs help identify potential entry and harborage of the pathogen, prevent cross contamination, and ultimately minimize the occurrence of the pathogen in food. Both practices can trigger corrective actions to avoid food contamination and restrict the likelihood of allowing potentially contaminated food from entering commerce.

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