ABSTRACT

Title of Dissertation: USE OF RISK ASSESSMENT MODELING TECHNIQUES TO DEVELOP QUANTITATIVE RISK-BASED HAZARD ANALYSIS AND CRITICAL CONTROL POINT (RB-HACCP) PLANS

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Hazard Analysis and Critical Control Point (HACCP) is the internationally recognized system to assure the safety of food products and the foundation of food safety programs worldwide. However, its success is limited by its inability to relate stringency to measurable public health impacts and its inherent qualitative nature. The aim of this research was to incorporate quantitative microbiological risk assessment (QMRA) techniques into HACCP to develop risk-based HACCP (RB-HACCP) plans. The research hypothesized that the Critical Control Points (CCPs) are steps in the process that significantly reduce the mean and/or variance of a hazard and that these

can be identified and quantified using risk assessment modeling techniques such as sensitivity analysis (SA) and what-if scenario analysis, as well as providing a more objective means in considering Critical Limits (CLs). QMRA models were developed for two distinctly different commercial food products, frankfurters and cold-smoked salmon (CSS). The former has a definitive inactivation step while the latter achieves control through a series of partial control steps. Modular Product Pathogen Pathway risk assessment models were developed to identify potential risk-based CCPs (RB-CCPs) for the control of *Listeria monocytogenes*. Steps of the processes within modules were evaluated and prioritized using SA to determine the relative contribution of the process steps to control *L. monocytogenes*. What-if scenario analyses were subsequently used to quantitatively determine the consequences of system deviations, thereby allowing risk-based CLs (RB-CLs) to be set and the most-effective risk mitigation strategies to be identified. This conceptual framework, combined with relevant plant-specific data, was used to identify RB-CCPs and RB-CLs, thereby producing RB-HACCP plans that are linked with public health goals to lower the risk of listeriosis. This allowed a direct comparison between current industry HACCP plans for frankfurters and CSS with RB-HACCP plans derived from the risk assessments. The comparison suggests that the use of RB-HACCP plans may offer advantages in developing the “preventive controls” risk management food safety plans required under the FDA Food Safety Modernization Act of 2011.

USE OF RISK ASSESSMENT MODELING TECHNIQUES TO DEVELOP QUANTITATIVE RISK-BASED HAZARD ANALYSIS AND CRITICAL CONTROL POINT (RB-HACCP) PLANS

by

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Dedication

The author dedicates this dissertation to her daughter Victoria.

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List of Abbreviations

AFDO	Association of Food and Drug Officials
AIDS	Acquired Immune Deficiency Syndrome
ARS	USDA Agricultural Research Service
CAC	Codex Alimentarius Commission
CAST	Council for Agricultural Science and Technology
CCFH	Codex Committee on Food Hygiene
CCP	Critical Control Point
CDC	Centers for Disease Control and Prevention
CFR	Code of Federal Regulations
cGMP	Current Good Manufacturing Practices
CI	Confidence Interval
CL	Critical Limit
CFIA	Canadian Food Inspection Agency
CFU	Colony Forming Unit
CSFII	Continuing Survey of Food Intakes by Individuals
CSS	Cold-Smoked Salmon
CUSUM	Cumulative Sum
6D	Six-Logarithm Reduction
DNA	Deoxyribonucleic Acid
EFSA	European Food Safety Authority
EGR	Exponential growth rate
ELISA	Enzyme Linked Immunoabsorbent Assay
ERS	USDA Economic Research Service
FAO	Food and Agriculture Organization of the United Nations
FDA	U.S. Food and Drug Administration
FIFO	First-In-First-Out
FNR	Frankfurter Not Reheated
FR	Frankfurter Reheated

FSAI	Food Safety Authority of Ireland
FSANZ	Food Standards Australia New Zealand
FSIS	USDA Food Safety and Inspection Service
FSMA	Food Safety Modernization Act
FSO	Food Safety Objective
FSP	Food Safety Plan
FSPCA	Food Safety Preventive Controls Alliance
GAP	Good Agricultural Practices
GHP	Good Hygiene Practices
GMP	Good Manufacturing Practices
GPFH	Codex General Principles of Food Hygiene
GRAS	Generally Recognized as Safe
HACCP	Hazard Analysis Critical Control Point
HARPC	Hazard Analysis Risk-based Preventive Controls
HHS	U.S. Department of Health and Human Services
IAFP	International Association for Food Protection
ICMSF	International Commission on Microbiological Specifications for Food
IFT	Institute of Food Technologists
ILSI	International Life Sciences Institute
IOM	U.S. National Academies of Science Institute of Medicine
LPD	Lag phase duration
MC	Microbiological criterion/criteria
MLE	Maximum Likelihood Estimations
MPD	Maximum population density
MPN	Most Probable Number
MPPP	Modular Product Pathogen Pathway
NACMCF	U.S. National Advisory Committee on Microbiological Criteria for Foods
NAHMS	National Animal Health Monitoring System
NAL	USDA National Agricultural Library
NASS	USDA National Agriculture Statistics Service

NHANES	National Health and Nutrition Examination Survey
NHDSC	National Hot Dog and Sausage Council
NRC	National Research Council
NSHA	National Seafood HACCP Alliance for Training and Education
oPRP	Operational Prerequisite Program
OIE	World Organization for Animal Health
OSU	The Ohio State University
PAHO	Pan American Health Organization
PC	Performance Criteria
PcC	Process Criteria
PCHF	Preventive Controls for Human Food
PCQI	Preventive Controls Qualified Individual
PCR	Polymerase Chain Reaction
PdC	Product Criteria
PO	Performance Objective
PRP	Prerequisite program
QALY	Quality-Adjusted Life Year
QMRA	Quantitative Microbial Risk Assessment
RB-CCP	Risk-Based Critical Control Point
RB-CL	Risk-Based Critical Limit
RB-HACCP	Risk-Based Hazard Analysis Critical Control Point
RTE	Ready-to-eat
RSI	Risk Science Institute
SA	Sensitivity analysis
SD	Standard deviation
SEM	Scanning electron microscopy
SPC	Statistical process control
SPS	Sanitary and Phytosanitary Agreement of the World Trade Organization (WTO)
UN	United Nations

U.S.	United States
USDA	U.S. Department of Agriculture
WHO	World Health Organization
WTO	World Trade Organization

Chapter 1: Introduction

Hazard Analysis Critical Control Point (HACCP), the internationally recognized food safety management system, is widely used to assure the safety of food products. Despite HACCP's international adoption, this system's qualitative focus lacks the ability to directly relate food safety stringency to measurable public health impacts, i.e., its focus is qualitative. For example, current HACCP plans are supposed to identify Critical Control Points (CCPs) wherein hazards can be prevented, eliminated, or reduced to acceptable levels of risk. To that end, HACCP could benefit by combining this system with techniques available through quantitative food safety risk assessment to allow a more quantitative consideration of risk. The ability to link HACCP programs to food safety public health outcomes is crucial to ultimately developing risk-based food safety systems at the facility level.

The overall goal of this dissertation is to develop quantitative microbiological risk assessment models for commercial frankfurter and cold-smoked salmon (CSS) operations to use the risk assessments as tools to develop risk-based HACCP plans incorporating food safety risk management metrics. The quantitative models developed could serve as prototypes for HACCP plans that are real-time risk management tools, and HACCP plans that are more directly related to public health impacts, thereby allowing for risk-based selection of CCPs and CLs, and improved determination of equivalence for international trade. Thus, the dissertation presents two case studies to demonstrate the potential benefits of advancing HACCP plans by providing more objective quantitative measures that incorporate reliable food safety risk management metrics.

Buchanan and Williams (2013) provided an in-depth discussion of what a HACCP system is and how it functions. To move beyond the shortcomings of this food safety system to develop food safety risk management metrics, it is important to provide a brief overview of

the key points of the HACCP system. To that end, this chapter begins with a summary of the origins, evolution, and limitations of the current HACCP system. This is followed by a discussion on the emergence of risk analysis and its impact on HACCP, including recent examples of HACCP's continuing evolution. Specifically, the chapter is organized as follows: A general introduction (Section 1.1), origins of HACCP (Section 1.2), evolution of HACCP (Section 1.3), limitations of traditional HACCP (Section 1.4), emergence of risk analysis and its impact on HACCP (Section 1.5), recent examples of the continuing evolution of HACCP systems (Section 1.6), and a summary of these key points (Section 1.7) including an introduction to the working hypotheses.

1.1 *General Introduction to HACCP*

The combination of the Hazard Analysis and Critical Control Point (HACCP) system and current Good Manufacturing Practices (cGMPs) is the gold standard worldwide for the management of food safety risks (NACMCF 1998; CAC 2009b; Buchanan and Williams 2013). Understanding the 50-year evolution of HACCP (Sperber and Stier 2009) and its emergence as the primary food safety risk management system for foods is critical to shaping the future of food safety risk management, and requires one to be knowledgeable about its origins and expansion over time.

1.2 *Origins of HACCP*

To fully appreciate the origins of HACCP it is important to understand the reasons for its emergence, its early history, and its key concepts and principles. In addition, it is worthwhile to understand the early developments of this system not just in the U.S. but in other countries.

1.2.1 Reasons for the Emergence of HACCP

The traditional belief about the origins of HACCP is that it was conceived by the Pillsbury Company, in collaboration with the National Aeronautics and Space Administration (NASA) and the U.S. Army Laboratories at Natick, to ensure the safety of astronauts' food during the early days of the U.S. space program (Sperber and Stier 2009). However, an additional key factor that allowed HACCP to emerge was that Good Hygienic Practices (GHPs) did not fully control food safety concerns faced by the food industry, in particular, the elimination of foodborne disease (Buchanan 1990).

Several other systems or mandates collectively led to the conceptualization and establishment of HACCP, specifically: (1) the Critical Control Point (CCP) engineering management mandate by NASA, (2) the development of reliability engineering systems, and (3) the use of Failure Mode and Effect Analysis (FMEA) by the Army Laboratories at Natick (Mortimore and Wallace 2001). The engineering design criteria used in HACCP were based on several existing aerospace documents, including the *Handbook of Instructions for Aerospace Personnel Subsystem Designers* (USAF Manual 80-3) (Lachance 1971), which served as the primary source of personnel subsystem information applicable to the management, design, and development of control systems. Focused on identifying "critical failure areas," Pillsbury and NASA worked to eliminate such liabilities from the system. Using the analysis to identify and implement effective means of control, the team successfully used this approach to evaluate potential hazards at each step in the operation process. In particular, the application of FMEA concepts to improving food systems was an innovative reimagining of the concepts developed by NASA.

1.2.2 Early History of HACCP

The HACCP system, formalized in the U.S. in 1972, had been conceived more than a half-century ago. One of its initiating events was the launch of the world's first satellite,

Sputnik, by the Soviet Union on October 4, 1957. Five days later, on October 9, 1957, President Dwight D. Eisenhower committed the U.S. to the space program by signing the National Aeronautics and Space Act (H. R. 12575) into law on July 29, 1958 (Hagerty 1958). The goals of the space program were first to launch a scientific satellite into orbit and second, to put a "man-on-the-moon" (Piland 1958 p. 2). Developing safe, nutritious, well-designed food for astronauts was an integral component of the plan. Scientists from the Quartermaster Food and Container Institute of the United States Armed Forces, now known as United States Army Laboratories in Natick, Massachusetts (Bauman 1993), and NASA's Manned Spacecraft Center in Houston, Texas (Dick and Launius 2007) teamed up with the Pillsbury Company in 1959 to develop food for manned space missions (Bauman 1993). At that time, food safety and quality systems were generally based on end-product testing. While end-product testing was better than no testing, it had its limitations. Scientists had already concluded that end-product testing had two major shortcomings. First, a considerable amount of testing had to be done to provide a high degree of assurance. Second, end-product testing involved a reactive rather than a proactive approach to hazard control. In response to these concerns, the scientists began to design a preventive approach to food manufacturing that incorporated safety measures into the formulation, production, and packaging of foods to more effectively ensure the safety of food (Mossel 1969). This new food safety approach focused primarily on making strict microbiological assessments. Working on the strict microbiological requirements for space food, the team of scientists applied unique criteria, using pathogens rather than indicator organisms, to specify and standardize pathogen limits on all foods destined for consumption in outer space (Lachance 1993). While Pillsbury's initial focus was using HACCP to assure microbiological safety, it quickly adopted HACCP principles to manage physical and chemical hazards. In 1970-71, when confronted with a serious food safety matter (glass contamination in farina for infants) in one of its commercial food products, Pillsbury quickly adopted the HACCP system for the production of its food (Ross-Nazzal 2007).

1.2.3 Key Concepts and Principles

Prerequisite programs for HACCP include procedures such as Good Manufacturing Practices (GMPs), which address operational conditions and provide the foundation for the HACCP system (SHA 2011). Prerequisite programs outline the basic environmental and operating conditions necessary for the production of safe food (Scott and Stevenson 2006). Likewise, Good Hygiene Practices (GHPs) are expected to be implemented in virtually any food facility and distribution operation. These foundational practices may be sector-specific, such as GMPs for food processing, or commodity-specific, such as Good Agricultural Practices (GAPs) for primary production (Buchanan and Williams 2013).

Programs that may be considered prerequisites include sanitary design, personnel hygiene and training, production equipment, control of raw materials, sanitation, environmental monitoring, chemical control, pest control, allergen management program, glass control, receiving, storage/distribution, product tracing/recall, and maintenance (Scott and Stevenson 2006). GHPs are not product- or line-specific, and do not help to identify steps that pose the greatest risk in a processing line. Having effective prerequisite programs in place, however, simplifies the development and maintenance of a HACCP plan (Sperber and others 1998), and may result in a more manageable implementation. Many prerequisite program practices are specified in federal, state, and local regulations and guidelines. Like HACCP plans, these programs should be well-documented with written Standard Operating Procedures (SOPs) (Scott and Stevenson 2006). While this section provides a brief overview view of some prerequisite programs, a more in-depth discussion of the prerequisite programs, regulations, and guidance for GHP compliance has already been presented by Buchanan and Williams (2013).

1.2.3.1 Systems Thinking and HACCP

Buchanan and Williams (2013) explain that the basic concept underlying 'systems thinking' is that for complex systems (like food production) understanding the component parts of the system can be best understood in the context of their relationships among themselves and with other systems. These techniques have been successfully employed in highly complex industries such as the aerospace products. In fact, it was the aerospace program where system-thinking approaches (Jenkins 1969 p. 11) and methods for performing risk and reliability assessment originated (NASA 2011). Examples of methods that NASA relied on for systems safety assessment early in the Apollo program include Hazard Analysis (HA) and Failure Mode and Effects Analysis (FMEA) (NASA 2011). While many other industries have adopted the FMEA method, modifying it to meet their specific needs (Modarres and others 2010), this was not the case with the food industry. Granted that the HACCP system evolved out of FMEA, over time it actually moved away from the FMEA model following a reductionist approach. Since its inception in the mid-1960s, FMEA continued to evolve by incorporating various analytical and informatics tools. An example of this evolution is Failure Mode, Effects, and Criticality Analysis (FMECA). As stated in IEC (2006), "FMEA might be extended to incorporate an investigation of the degree of severity of the consequences, their respective probabilities of occurrence, and their detectability, thereby becoming a Failure Mode, Effects, and Criticality Analysis (FMECA; see IEC 60812)" (FDA 2006). By highlighting failure modes with relatively high probability and severity, FMECA allows preventive measures and/or mitigation strategies to be allocated more effectively where they will greatly minimize the risk. FMECA's methods of risk identification, which should be used in conjunction with other reliability tools, are applicable to extremely complex system and, provide the necessary visibility into such systems (NASA 1993). The use of FMECA has become a fairly standard practice in aerospace and other industries, including individual failure modes for corrective actions in the design phase of the processes. Without a formal FMECA process, the system

design integrity would be determined by the experience and rigor of individual design engineers. As a result, there would be no means of verifying that design risk had been minimized to an acceptable degree (NASA 1993).

In the case of food safety, "the efficacy of any HACCP system . . . rely on . . . employees having the appropriate HACCP knowledge and skills . . ." (CAC 2003). Also, "risk decisions" established after developing HACCP plans are often non-transparent and not fully supported by an adequate assessment. The lack of a FMECA-based or risk-based HACCP system in the food industry has probably increased the HACCP system reliance on personnel knowledge and skills (Buchanan 2010). Numerous safety-based reliability and systems engineering applications have been developed for aircraft safety and safe practices at nuclear facilities. Examples of these reliability techniques are Fault Tree and Success Tree Analysis (e.g., Binary Decision Diagrams), Event Tree Analysis, Master Logic Diagram, Probabilistic Scenario Analysis, Reliability Block Diagram Analysis, Influence Diagrams, and human reliability. In addition, an extension of Bayesian Belief Networks (BBNs) applications for reliability and risk assessment is the proposed method Qualitative-Quantitative Bayesian Belief Networks (QQBBN), which enables the use of both qualitative and quantitative likelihood scales in inference. The inclusion of qualitative scales is especially useful when quantitative data for estimation of probabilities are lacking and experts are reluctant to express their opinions quantitatively (Wang and Mosleh 2010).

These techniques could potentially improve the safety and reliability of the food supply by enhancing the identification and ranking of specific hazards and by improving the reliability of personnel and equipment within a system approach. These techniques are usually applied to more advanced technological industries with higher profitability and margins (i.e., aviation, nuclear, and aerospace). As a result, a cost-benefit analysis could be beneficial when incorporating reliability techniques at the food industry level. In the food industry, Bayesian

methods have been used to estimate uncertainty and variability in the prevalence of *Bacillus cereus* spores (Malakar and others 2004) and *Listeria monocytogenes* (Delignette-Muller and others 2006) in specific food processes. In addition, Smid and others (2010) compiled the strengths and weaknesses of BBNs and Monte Carlo simulation models in microbial risk assessments, and proposed a practical framework for the construction of a biotracing model (Smid and others 2011). The food industry has yet to adopt many of these tools that might potentially enhance the HACCP system. Moreover, a larger role for risk assessment modeling techniques and approaches was recommended by a number of food safety scientists (Buchanan 1995; Notermans and others 1995; Wilson 1997; Mayes 1998; Buchanan and Whiting 1998; Serra and others 1999; Gaze and others 2002; Buchanan 2010; Buchanan and Williams 2013). Despite the many recommendations, the HACCP system evolved little from the general approach originally articulated by the pioneers of the space program.

1.3 *Evolution of HACCP*

The following section on the evolution of HACCP covers its early adoption and acceptance by regulatory agencies, the Codex endorsement of HACCP at the international level, and the expansion of HACCP.

1.3.1 Early Adoption of HACCP

In 1970 and 1971, incidents of *Clostridium botulinum* contamination, as well as several cases of illness and deaths due to botulism, were attributed to under-processed, low-acid canned foods (LACF), specifically canned soup. These incidents coincided with the 1971 introduction of the concept of HACCP at the National Conference on Food Protection (United States Department of Health, Education, and Welfare 1972 cited by WHO/ICMSF 1980). In September, 1972, at the prompting of the U.S. Food and Drug Administration (FDA), Pillsbury organized and conducted a training program for FDA inspectors based on the newly developed

HACCP system for food safety, which included CCPs as a potential approach to regulating the processing of canned foods. The eleven-day training program consisted of classroom lectures, in-depth discussions and ten days of canning plant evaluations (Sperber 2006 as cited by Ross-Nazzal 2007). The first published use of the term "HACCP" is credited to this training program (Sperber and Stier 2009). In addition to training programs, in 1972 Pillsbury established internal specification systems and the following year published a comprehensive treatise on HACCP (FAO 2003). This document included three principles upon which the initial HACCP system was based: 1) conducting a hazard analysis, 2) determining critical control points, and 3) establishing monitoring procedures (Sperber 2005). This set of food safety tools was used to help identify the controls specified in the LACF regulations (Mortimore and Wallace 1994). Thus, canned products were the first foods for which HACCP guidelines were mandated (NAS 1985), and in 1973 the HACCP system was officially adopted for the LACF regulations (OSU 2002). Through experience with its new management system, Pillsbury subsequently adopted two additional principles: 4) establishing corrective actions to take when deviations occurred at a CCP, and 5) establishing critical limits for the required level of control at the CCPs (Sperber and Stier 2009).

In view of the emphasis given to the application of HACCP by the 1976 WHO Expert Committee on Microbiological Aspects of Food Hygiene, WHO, through its mandate on food safety, proposed a meeting with the main purpose of assessing HACCP's practical use in both developing and developed countries. The report of this WHO/ICMSF Meeting on "Hazard Analysis: Critical Control Point System in Food Hygiene" was published in 1980. By 1983, WHO Europe was largely recommending the use of the HACCP system. Similarly, in 1985 the National Academies of Science (NAS), recognized that despite the successful application of HACCP to microbiological control of LACF and its merits, the use of the HACCP system by the food industry was far from universal and urged that the HACCP system be more broadly

applied to various categories of non-canned foods. Specifically, the NAS proposed two plans of action. The first consisted of the universal application of the HACCP system in food protection programs within the food industry. The second consisted of the implementation of microbiological criteria (MC) for several categories of food products. Based on recommendations from the 1985 NAS report, the National Advisory Committee on Microbiological Criteria for Foods (NACMCF) was created and held its first meeting in 1988 (Scott and Stevenson 2006). In that same year, the International Commission on Microbiological Specifications for Foods (ICMSF) published the first book devoted to the development and implementation of HACCP ("Microorganisms in Foods 4: Application of the Hazard Analysis Critical Control Point [HACCP] system to Ensure Microbiological Safety and Quality").

In 1989, the NACMCF developed and approved a standardized and updated HACCP system, endorsed by several federal regulatory agencies. In 1992, NACMCF issued a revised document on HACCP and added two more principles to the HACCP system: 6) establishment of procedures for verification to confirm that the HACCP system is working effectively, and 7) preparing documentation concerning procedures and records applicable to these principles and their application (FAO 2003). In 1993, the Codex Alimentarius Commission (CAC) adopted and issued its first HACCP Guidelines, and in 1997 NACMCF issued a third revised document (NACMCF 1998). At the international level, Codex adopted the Guidelines for the Application of the Hazard Analysis Critical Control Point System (CAC/GL 18-1993) at its twentieth session (FAO/WHO 1998). These Codex guidelines were subsequently revised by the Codex Committee on Food Hygiene and adopted at the Twenty-Second Session of the CAC in Geneva (WHO 1997). CAC/RCP 1-1969, Rev. 4-2003-Annex represents the latest version of the HACCP guidance document adopted by Codex (CAC 2009b). The World Health Organization (WHO) and the Food and Agricultural Organization of the United Nations (FAO)

have recognized and promoted international acceptance of HACCP as being the most-effective means for prevention of foodborne illness.

At the national level, the U.S. FDA's seafood HACCP program was established in 1995 and became mandatory in 1997. Subsequently, the U.S. USDA/FSIS HACCP program for meat and poultry became mandatory for large facilities in 1998, for smaller ones in 1999, and for the smallest facilities in 2000. HACCP-based regulations were established by FDA for juices in 2001 and became mandatory for large processors in January 2002, small businesses in 2003, and very small businesses in 2004 (FDA 2001). As a result of a congressional request, a comprehensive study of gaps in public health protection provided by the food safety systems in the U.S. was initiated in 2008. The subsequent report "Enhancing Food Safety," which focused on the role of food safety programs at the FDA, was published by the National Academy of Sciences (NAS) in 2010. This report proposed an overall risk-based food safety management approach including requirements for preventive controls based on hazard analysis and risk (IOM/NRC 2010). While the 2010 NAS report noted that the FDA had already established the authority to impose preventive process control regimes on food facilities in their promulgation of the seafood and juice HACCP rules, the report recommended that the Federal Food, Drug and Cosmetic Act (FDCA) be amended to make the authority explicit and to mandate that all registered food facilities have such controls in place. These recommendations are reflected in the Food Safety Modernization Act (FSMA), a law that has profoundly reshaped food safety by incorporating risk-based approaches.

While the overall HACCP system was described in detail by Buchanan and Williams (2013), this chapter extends the discussion further by focusing on the state of science after 2013 and the key points of the HACCP system as they relate to both risk assessments and system approaches. Systems approaches were also discussed at a symposium entitled "The Next Risk

Analysis Challenge: Linking HACCP and Risk Assessments" at the 2013 Annual Meeting of the International Association for Food Protection (IAFP 2013).

1.3.2 Regulatory Agencies Acceptance of HACCP

In the U.S., HACCP is mandatory for the seafood, meat, poultry, egg products, and juice industries. HACCP compliance through the Code of Federal Regulations (CFR), is governed by either 21 CFR or 9 CFR, depending on the jurisdiction of the products. On January 4, 2011, the 111th Congress amended the Federal Food, Drug, and Cosmetic Act (FDCA) with respect to the safety of products regulated, mainly under 21 CFR (GPO 2011). The "FDA Food Safety Modernization Act" (FSMA) amended Chapter IV (21 USC 341 et seq.) by adding at the end of the FDCA and the 21 USC, sections 418 and 350, respectively. These and other changes are reflected in the PCHF and Produce Safety final rules previously mentioned.

1.3.3 Codex Endorsement of HACCP

The Codex Alimentarius Commission (CAC), a joint subsidiary body of the Food and Agriculture Organization of the United Nations (FAO) and World Health Organization (WHO), promoted the application of the HACCP system at the international level. That international support is one reason why, in the over fifty years since its genesis, the HACCP system has become the internationally recognized and accepted method for food safety assurance. As such, HACCP has been referred to as "the most revolutionary institutional innovation to ensure food safety of the twentieth century" (Ross-Nazzal 2007).

1.3.4 Expansion of HACCP

Some insist that HACCP was meant to be implemented mainly at the food processing establishment level for which it was originally developed (Kaufmann and Schaffner 1974 cited by WHO/ICMSF 1980; Jouve 1998). And yet, in 1980, the WHO/ICMSF recognized that this

was not necessarily true, since the use of HACCP had been extended to food service establishments (Bobeng and David 1977; Bryan and McKinley 1979) and the home (WHO/ICMSF, 1980). To that end, HACCP is recognized as a system that enhances the safety of foods from primary production through final consumption (FAO/WHO 2003).

1.4 *Limitations of Traditional HACCP*

HACCP was highly innovative when it was introduced in the 1970s, effectively pre-dating many of the tools that would have been useful to enhance its performance. As previously mentioned, the rush to standardize HACCP in an effort to make it more broadly applicable, and comparable among commodities and regions, discouraged further major modification of the initial approach. This reluctance for greater specificity resulted in the retention of a number of inherent limitations associated with HACCP, several of which are reviewed briefly below.

The most widely recognized limitation of HACCP as a risk management system is its general inability, other than in a qualitative manner, to link the stringency of a HACCP program directly to its public health impacts (Buchanan and Whiting 1998; Whiting 2003; Buchanan and Williams 2013). Metrics for assessing the public health impact of foodborne diseases involve the incidence of such disease cases and the number of outbreaks. However, due to a lack of quantitative foodborne disease attribution and the inability of food manufacturers and regulatory agencies to relate foodborne disease to something that is measurable and controllable in foods, the discussion quickly falls back to a hazard-based approach rather than one based on risk. This problem has been one of the driving forces behind the emergence of the food safety risk management metrics approach originally introduced by the ICMSF (2002).

In addition to implementation concerns such as: lack of expertise and resources for the development of HACCP plans, failure to maintain the HACCP system, and management

neglect of food safety, additional inherent limitations are presented considering each of the seven HACCP principles as follows**:**

1.4.1 Principle 1 Conduct a Hazard Analysis

To develop a HACCP plan, the team should first conduct a Hazard Analysis. Conducting a Hazard Analysis is the central pillar of any HACCP plan. However, limited guidance is available to HACCP teams on how to approach the application of this principle (Wallace and others 2014). NACMCF (1998) and CAC (2003) provide brief points to consider for principle 1. A hazard analysis is the process of collecting and evaluating information on hazards and conditions leading to their presence to decide which elements are significant for food safety and should therefore be addressed in the HACCP Plan (CAC 2003). Buchanan and Williams (2013) noted that the hazard analysis can be appropriately viewed as a qualitative risk assessment (or possibly a risk profile). In this regard, a limitation associated with principle 1 is its focus on hazards and not risks, which makes it difficult to objectively prioritize the hazards in terms of need for control. A related limitation is that hazard analyses are typically based on specific hazard/product pairs. This pairing tends to lead to a limited number of hazards being identified. In some cases, this limitation could be overcome by considering groups of hazards with common characteristics. For example, control of the risk associated with Gram-negative enteric pathogens might be a more practical way of capturing a variety of high and moderate risk pathogenic bacteria such as *Salmonella enterica*, *Shigella* spp., *Yersinia* spp., and Shiga toxin-producing *Escherichia coli*. Such an approach would have to be weighed against the benefits of hazard specificity and the likelihood that the different pathogens have different sources and methods of control. Weingold and others (1994) proposed a standardized classification system of foodborne disease outbreaks to group data more readily available to be used for the hazard analysis. This method, based on surveillance systems, was used by local health departments at the state level (Weingold and others 1995).

The current thinking is that the HACCP team should consider each raw material and process activity and list all potential hazards that might occur as the first step in identifying the significant hazards (Wallace and others 2011; Mortimore and Wallace 2013; Wallace and others 2014). Agreement on how to determine which of the potential hazards in a food are "significant hazards" remains a challenge. Since the term is only defined subjectively, disagreement occurs on exactly what hazards should be classified as significant and therefore included in the HACCP plan. A 1998 Joint FAO/WHO Consultation on the Role of Government Agencies in Assessing HACCP recommended that the appropriate Codex Committee undertake work to clarify the phrase "significant hazard." In 1999, ILSI defined "significant hazard" as "hazards that are of such a nature that their elimination or reduction to an acceptable level is essential to the production of safe foods;" however, this does not offer much more clarity than the original text by NACMCF (1998). Currently, most food processing companies determine the significance of hazards using judgment and experience. However, this is not typically an evidence-based process, and decisions can be difficult to defend if a client or a regulatory agency feels that additional hazards should have been included in the HACCP plan. An additional limitation on conducting of hazard analysis is that this first step in developing a HACCP plan has not been adapted to consider either food defense or economic adulteration hazards. Furthermore, this limitation could be extended to include its lack of ability to deal with emerging hazards in a timely manner.

1.4.2 Principle 2 Determine the Critical Control Points

In addition to not being able to assess what a "significant hazard" is, the current approach to determining CCPs is also qualitative to semi-quantitative, and is largely based on expert opinions or specified by regulatory authorities. To determine CCPs, decision trees work well for foods when there is an overwhelming inactivation step that effectively eliminates the hazard. However, for foods that rely on a series of incremental controls, none of which

eliminate the hazard, the decision trees are less definitive and the resultant HACCP becomes too complex (i.e., too many CCPs) for simple yes/no decisions, particularly if control requires synergistic effects among the control measures. The inability to quantitatively determine the degree of risk mitigation by incremental controls can lead to either the inclusion of CCPs that do little to improve public health or the absence of a CCP that is important only under specific combinations of conditions (Buchanan 2013).

Buchanan and Whiting (1998) concluded that while HACCP systems work well for microbiological hazards in which an intervention step provides a large reduction in the target microorganism (e.g., pasteurized milk, commercial sterilization of canned foods, cooked meats), it is less effective for foods that receive little to no reduction in microbiological populations. Instead, such foods (e.g., fresh produce, cold-smoked fish, fermented dairy products made from raw milk) rely on a series of controls that either partially reduce microbial populations or delay the growth of pathogenic microorganisms. Consequently, the ability to determine which steps in the manufacturing process are CCPs becomes much more difficult, particularly if there is substantial variability in the ingredients and processes. Typically, this variability results in a HACCP plan that is considerably more complex in terms of the number of identified CCPs. The ICMSF (1988) attempted to deal with the differences in the extent of control that can be applied to different types of food processes by distinguishing two classes of CCPs, i.e., CCP1 and CCP2. CCP1 "assure control of a hazard," whereas, CCP2 "minimize but cannot assure control of a hazard." In practice, CCP1 simply minimizes the risk with a higher level of assurance than CCP2. Nevertheless, this approach was not widely adopted and in 1990 NACMCF rejected the use of a two-class CCP system (Buchanan 1990).

1.4.3 Principle 3 Establish Critical Limits

Critical limits have often been established based on an "As Low as Reasonable Achievable" (ALARA) approach and not on the actual degree of control needed to manage

risks to a specified level of control. This approach can result in either insufficient control of hazards or greater stringency than actually needed. For example, Anderson and others (2011) evaluated the adequacy of traditional thermal processing versus a risk-based approach and concluded that the latter provides opportunities for alternatives to the traditional 12D treatments that achieve an equivalent degree of risk mitigation. A common practice in the industry is to increase CLs to provide an additional degree of protection and eliminate uncertainty. However, for processing that involves an overwhelming inactivation, such steps are not likely to achieve greater control since any residual risk is typically associated with recontamination after the inactivation step.

1.4.4 Principle 4 Establish a System to Monitor Control of the CCPs

Even after 40 years of use, there is still confusion in the minds of many over the differences between monitoring, verification, and validation. In all three cases, the frequency and sensitivity of testing are seldom designed to match the intended level of stringency contemplated in the HACCP plan, again reflecting the lack of quantitative evaluation of the facility's food safety risk management program. Perhaps the best example of this limitation is the use of microbiological testing for HACCP verification. It is well recognized that the ability for such testing to determine the effectiveness of control measures is dependent on the sensitivity of the methods employed, the frequency of testing, and the sampling plans selected (ICMSF 2002). However, since the stringency of the HACCP system itself is seldom determined, the appropriate selection of sampling plans is seldom based on the level of confidence that food operators or their purchasers are trying to achieve. This lack of specificity typically results in the selection of sampling plans that are not risk-based and are statistically inadequate to meet the expectations of the company (Buchanan 2013).

1.4.5 Principle 5 Establish Corrective Action based on Monitoring

In addition to monitoring the CCP, a HACCP needs to include a plan of action for when a deviation from critical limit occurs. Yet, when a process deviation occurs, the corrective actions to be taken are seldom evaluated in relation to the risk they pose. The establishment of corrective actions would benefit from a more formal risk evaluation so that the response is proportional to the increased risk caused by the process deviation. Performing such evaluations after a process deviation has occurred is generally unrealistic in the case of most foods due to the need to make decisions in a timely manner and avoid the appearance of employing ad hoc decision criteria.

A related limitation is the consideration of causality when addressing process deviations or verification testing that does not meet the established criteria. A key component to the response is determining whether such results are an indicator of a systemic deficiency in the food safety system. Such negative findings would generally be assumed to represent a loss of control. However, in any control measure there are instances where a negative result will be indicated but the system is still under control. In establishing a decision criterion, there will be a small portion of the samples that will exceed the designated value despite the system being in control. The portion of these "false-positives" is inherent in the decision criteria selected for monitoring or verification. Yet unless there is an adequate understanding of the variability of the control measures of the overall HACCP system, such findings will result in a frustrating search for a process failure that does not exist. The problem related to such residual risks can be reduced by clearly benchmarking the inherent variability of the control measures and understanding the risks of type 1 and type 2 errors in monitoring and verifications activities. An extremely useful group of tools are the statistical process control (SPC) trend analysis techniques. SPC tools are designed to enhance the collection and analysis of HACCP data. For example, Tokatli and others (2005) demonstrated the utility of multivariate process

monitoring and fault diagnosis techniques for HACCP programs involving food pasteurization processes. Likewise, Srikaeo and Hourigan (2002) demonstrated that SPC techniques could be used to enhance the validation of CCPs related to shell egg washing. Srikaeo and others (2005) used SPC techniques to examine biscuit baking and found that a number of the parameters that influenced the adequacy of the baking process were not in control.

1.4.6 Principle 6 Establish Procedures for Verification

Verification is the use of methods, procedures, or tests, in addition to those used in monitoring, to determine if the HACCP plan is being followed (Buchanan and Williams 2013) and the system is operating according to plan. Verification activities are typically carried out by quality control personnel, third party experts, and/or regulatory agencies. Differing results with monitoring and verification activities could signal a need to reevaluate and possibly revalidate the system of control measures. HACCP plans need to be reassessed periodically and revalidated whenever considerable changes occur in product formulations, equipment, processing procedures, or sourcing of raw ingredients (Buchanan and Williams 2013). Without established procedures for verification the HACCP plan lacks validation if consistent unbiased results cannot be replicated.

1.4.7 Principle 7 Establish Documentation and Record Keeping

To review, verify and validate a HACCP plan, efficient and accurate record keeping is vital to the entire system. While record keeping has long been part of the HACCP approach to food safety risk management, there has often been a propensity to collect the information as an archival activity, with the different records being largely kept independent of each other. However, returning to the system engineering roots from which HACCP emerged, the underlying premise is that when systems become complex enough, they can be best understood as a whole system and not as individual parts of a system. Further, limiting record keeping to

archival activities misses an opportunity to use such records to monitor and verify the entire system. Ideally, the HACCP would foster the use of advanced informatics that integrate the different factors influencing the performance of the system, thereby providing a real-time means of assessing performance. Certainly, it is recognized by industry, policy makers, and researchers that the use of statistical process control is critical for assessing performance over time and establishing the variability within the system. Likewise, food safety risk analysis researchers have emphasized the potential for combining good risk assessment models with informatics technologies to provide a means for integrating the various data streams into the next generation of food safety risk management tools. Of particular interest is the potential for doing advanced what-if scenario modeling to assess how changes in the HACCP program are likely to impact the public health protection the program is intended to deliver.

1.4.8 Other System-wide Limitations of HACCP

In addition to the limitations associated with the individual principles, there are several system-wide limitations associated with the lack of quantitative consideration of the factors that influence the system's performance. Of particular interest is the ability of industry and government agencies to determine the "equivalence" or "comparability" of HACCP systems (Buchanan and Williams 2013). To date, comparisons across facilities and regulatory agencies have been done either qualitatively or by the use of performance metrics such as process control testing against some specified criteria (e.g., *Salmonella* and generic *E. coli* testing required as part of the USDA/FSIS Pathogen Reduction/HACCP regulation for raw meat and poultry). A lack of comparability or equivalence is further compounded by the inability to assess and differentiate the relative contribution of HACCP and GMP to the overall control of identified hazards. This can lead to a "one size fits all" approach to multiple facility operations that can weaken the flexibility which should be inherent in HACCP systems.

That HACCP is not a stand-alone program, due of its reliance on prerequisite programs and its limited applicability to the primary production sector (Cerf and others 2011), could be considered a limitation of the system. Sperber (2005a; 2005b) emphasized the need for prerequisite programs to focus on effective interventions and CCPs to protect public health. Furthermore, Cerf and others (2011) concluded that the HACCP system is not fully applicable at the primary production level. The agricultural or primary production sector represents a challenge for HACCP based on the many factors that cannot be controlled (e.g., animals in the surrounding areas, environmental conditions such as rain causing floods, motivated agricultural workers adequately trained). However, prerequisite programs such as GAPs, evaluated through well-designed questionnaires/survey instruments, combined with the application of emerging technologies such as Geographic Information Systems (GIS), which have been applied by the FDA in collaboration with NASA in the U.S., could provide a more comprehensive solution to the limitations found in the implementation of the HACCP principles within the agribusiness sector.

Another limitation of HACCP is the lack of standardized training. It is evident that the diffusion of HACCP system as it was disseminated throughout the world reached different locales at different levels of sophistication and in some cases remained static. The Codex Alimentarius Commission (CAC) has published guidelines for HACCP application. These guidelines have been recognized as the international standard for food safety and are a benchmark for food safety national requirements (FAO/WHO 1998).

Although worldwide HACCP is based on the guidelines published by the CAC, there has been some controversy over the years regarding the classification of CCPs. As early as 1971, the National Conference for Food Protection (CFP) found that the location of CCPs was a priority. At this conference the CCPs were divided into the following categories: raw materials CCPs, Processing CCPs, Environment CCPs, Personnel CCPs, Finished Product

CCPs, and Distribution CCPs. In addition, food processes and food products were classified. As previously mentioned, ICMSF (1988) attempted to classify CCPs by the assurance level of control of a hazard. CCP1 assured the control of the hazard, whereas CCP2 minimized, but could not control, the hazard. This approach was rejected by NACMCF in 1990. Still, the classification, and most importantly, the ranking of critical control points could prove useful regarding prioritization of resources. Therefore, not being able to rank or classify CCPs in a simple and pragmatic way could be considered a limitation.

In 1985, the use and universal application of HACCP were recommended by the U.S. National Academies of Science (NAS) (IOM 2003). Although HACCP was publicly introduced in the early 1970's, it remained relatively dormant until late 1995 when the FDA published, as a final rule 21 CFR123, the "Procedures for the Safe and Sanitary Processing and Importing of Fish and Fishery Products." This rule requires processors of fish and fishery products to develop and implement HACCP systems for their operations (FDA 1999). This edict was followed rapidly in the United States by the USDA establishing HACCP-based regulatory frameworks for meat, poultry, and egg products (USDA 1998), and FDA's juice HACCP program (FDA 2001). Likewise, various HACCP requirements were established by many developed and developing countries. While the adoption by regulatory agencies has a positive impact by providing a standard to identifying and controlling foodborne hazards, it has had the unintended consequence of constraining its further evolution. The effort and difficulties associated with modifying regulations present real-world impediments to addressing the limitations of HACCP and incorporating new concepts and tools, such as those that have emerged based on 20 years of investments in risk analysis. Nevertheless, there have been recommendations for HACCP programs to take better advantage of advances in risk assessment and risk management (Buchanan 1995; Notermans and others 1995, 1996; Wilson 1997; Buchanan and Whiting 1998; Mayes 1998; Serra and others 1999; Gaze and others 2002;

Buchanan 2010; Buchanan and Williams 2013). For example, IOM/NRC (2010) recommended enabling the FDA to impose preventive controls based on hazard analysis and risk on all food facilities, including the seafood and juice HACCP rules.

1.5 *Emergence of Risk Analysis and Its Impact on HACCP*

The ability to link the performance of a HACCP plan to the achievement of a risk-based level of control is being actively explored by industry, governments, academia, and intergovernmental agencies. Some of the recent examples of how concepts of risk metrics are being used include how to evaluate the stringency needed for GHP/HACCP programs for *L. monocytogenes* in ready-to-eat foods (Perez-Rodriguez 2006; CAC 2007b; Tenenhaus-Aziza and others 2014), *Salmonella* in poultry meat (Membré and others 2007) and beef-derived foods (Tuominen and others 2007), and *C. botulinum* in commercially sterile foods (Anderson and others 2011).

In the late 1990s authors associated with the ICMSF gave their perspectives on the relationship between HACCP and risk analysis, including risk assessments. Jouve (1998) addressed the relationship between HACCP and risk analysis and concluded that this relationship still needs to be clarified. On the other hand, Buchanan and Whiting (1997, 1998) proposed risk assessment as a tool for linking HACCP and public health. Other authors, such as Mayes (1998), considered the potential benefits and burdens to the industry arising from the application of elements of quantitative risk assessment in HACCP. Most research on the use of risk assessments to develop enhanced HACCP plans has focused on the benefits of applying quantitative methods (Buchanan 1995; Notermans and others 1995; Wilson 1997; Mayes 1998; Buchanan and Whiting 1997; Serra and others 1999; Gaze and others 2002). Although this research provides us with a clear understanding of the necessity of applying quantitative risk assessments to HACCP, most of it does not fully address how to establish the link between HACCP and public health by providing a selection of appropriate methods.

In 2003, the phrase “risk-based HACCP” was introduced by Whiting, who predicted that a complete risk-based regulatory approach would specify the Food Safety Objective (FSO), and that industry would be responsible for demonstrating that the entire processing system meets that objective. In 2004, Havelaar and others used a risk assessment model to define quantitative criteria and suggested that the connection between public health and FSO was “directly applicable in the HACCP framework.” According to IOM/NRC (2010), a risk-based food safety system requires the analytical capacity to assess food safety risks and policy interventions, as well as the ability to access data from a broad array of sources. The IOM/NRC report recommended the establishment of a “centralized risk-based analysis and data management center” as an important step toward the implementation of a risk-based approach to food safety management in the United States. An example of such a tool is iRisk, a web-based system introduced by FDA in 2012, designed to analyze data for hazards in food and develop appropriate risk assessments.

1.5.1 Food Safety Risk Analysis

Risk analysis experts generally consider the hazard analysis phase of HACCP as a qualitative risk assessment (CAST 2006), although some proponents suggest a distinction between a hazard analysis and a risk assessment (Wallace and others 2011). This, in part, seems to stem from the qualitative nature of most hazard analyses and the mistaken assumption that an evaluation must be quantitative to be considered a risk assessment. Many of the tools available to risk assessors are qualitative in nature. This is reinforced by the NACMCF (1998) description of the hazard analysis process leading to the selection of “significant hazards”: “The purpose of the hazard analysis is to develop a list of hazards that are of such significance that they are reasonably likely to cause injury or illness if not effectively controlled. Hazards that are not reasonably likely to occur would not require further consideration within a HACCP plan.” Clearly, it was expected that the identification of significant hazards was to be based on

a consideration of probability and severity, i.e., risk. Regretfully, NACMCF did not provide guidance on how this should be done, which, on one hand, allows for the use of multiple hazard analysis methodologies, but, on the other hand, does not provide a standardized approach. A number of investigators have argued that HACCP could be strengthened by moving to a more quantitative approach (Buchanan 1995; Buchanan and Whiting 1998; Serra and others 1999; Whiting 2003; Buchanan and Williams 2013).

HACCP can be characterized as a semi-quantitative system largely based on a qualitative "hazard assessment" (Buchanan 2011). HACCP constitutes a management tool more focused on risk management than risk assessment (Gaze and others 2002). HACCP has been described as an integration of risk management and process control systems (Coleman and Marks 2003) and as a system of process and risk control (Buchanan and Whiting 1998). Since HACCP includes both an assessment phase (i.e., hazard analysis) and a management phase (i.e., HACCP plan development and implementation), it can be described as a risk analysis system.

Clearly, the inclusion of a hazard must be based on probability and severity, i.e., risk. This quantitative risk assessment, based on probabilities, will be further discussed, as numerous investigators have argued that HACCP could be strengthened by moving to a more quantitative approach (Buchanan and Whiting 1998; Serra and others 1999; Whiting 2003).

1.5.2 Food Safety Risk Management Metrics

As microbiological risk assessments (MRA) became increasingly feasible, the development of approaches for relating target levels of risk control to attributes that could be controlled by food manufacturers received greater attention over the past decade. Risk analysis systems have evolved since the 1995 establishment of the World Trade Organization (WTO)

Sanitary and Phytosanitary (SPS) Agreement which stipulates that "countries' SPS measures must be based on an appropriate assessment of the actual risks involved" (WTO 2010). This agreement was a major impetus underlying the rapid advancement of chemical and microbiological risk assessments and their increased use in the development of food safety public policy throughout the world. A great deal of the focus has been centered around the concepts of Appropriate Level of Protection (ALOP) and Food Safety Objectives/Performance Objectives/Performance Criteria (FSO/PO/PC). The former arose from the SPS agreement, while the latter was introduced by ICMSF (2002) and subsequently adopted and modified by Codex Alimentarius (CAC 2007a). The definitions for FSO, PO, and PC have also been harmonized by Codex (CAC 2015) and are depicted in the glossary. The FSO/PO/PC concepts are referred to as new risk management metrics whereas Process Criterion (PcC), Product Criterion (PdC) and Microbiological Criterion (MC) can be referred to as traditional food safety metrics. Definitions for these concepts are depicted in Table 1.2.

The first uses of the FSO/PO/PC concepts have largely been around the development of risk-based microbiological criteria (MC) by CAC, having been used in the development of new MC for *Cronobacter sakazakii* in powdered infant formula (CAC 2008), and *L. monocytogenes* in ready-to-eat (RTE) foods (CAC 2007a).

Since the introduction of these concepts, there has been substantial activity in developing and exploring potential applications and further enhancing risk-based metrics. The basic statistics underlying FSOs have been evaluated, including relating food safety stringency to ALOPs or other means for expressing public health goals (Havelaar and others 2004; Zwietering 2005). Other examples of potential applications include development of food safety policies (Walls and Buchanan 2005; FAO/WHO 2004b, 2006a, 2006c), establishment and validation of control measures (Stewart and others 2003; Szabo and others 2003; Perez-Rodriguez and others 2006; Membré and others 2007; Zwietering and others 2010; Anderson

and others 2011), establishment of risk-based MCs (FAO/WHO 2004b; Whiting and others 2006; van Schothorst and others 2009), and food chain management (Gorris 2005). This has resulted in the concept being increasingly used to consider the relationship between proposed food safety programs and performance criteria at the national and international level. However, this application has generally not specified how it could be used to link HACCP to public health outcomes, thereby addressing many of the limitations of HACCP and realizing the goal of moving from hazards-based to risk-based food safety risk management.

1.5.2.1 The International Commission for Microbiological Specifications for Foods (ICMSF) Equation

ICMSF (2002) recognized that the focal point for controlling microbial pathogens should be on the numbers of a pathogen in the food at the time of consumption, not just the performance of a single processing step. Therefore, the FSO concept was created and symbolically expressed as a "conceptual equation" that represents the underlying framework upon which their metrics were based:

$$Ho + \Sigma I - \Sigma R \leq FSO$$

Where Ho is the initial level of the hazard or the initial contamination, ΣI is the total (cumulative) increase of the hazard or the sum of all the increases in population, and ΣR is the total (cumulative) reduction of the hazard or the sum of all the reductions. This equation establishes that the initial contamination, reductions through inactivation steps, potential recontamination, and possible growth during storage should be such that at the time of consumption the pathogen will be below a specific level in every serving, termed FSO (CAC 1997, 1999; CCFI 2003; ICMSF 2002; Whiting and Buchanan 2007, 2008; Whiting 2011). This equation allows for meeting the food safety objective (FSO) by controlling the hazard of interest within a process, which in turn enables the production pathway to be viewed as a series

of inputs and outcomes that are consistent with a systems approach. This general approach could be used to describe individual subsystems or the entire farm-to-table chain. In the former, the equation could be modified to consider the performance objective (PO) values instead of FSOs:

$$Ho + \Sigma I - \Sigma R \leq PO$$

These conceptual equations, in conjunction with an adequate risk assessment model, provide a conceptual means of linking the risk management metrics to both public health metrics and traditional food safety metrics. General guidelines for the use of these food safety risk management metrics are included in the CAC "Principles and Guidelines for the Conduct of Microbiological Risk Management" (CAC 2007a).

1.5.2.2 Microbial Criterion as a Verification Tool

MC as well as other traditional metrics verify that a PO is being consistently achieved. Microbiological criterion must consider the degree of confidence expected by the risk manager that a PO is not being exceeded, taking into account the variability and uncertainty associated with the product and the sampling/testing methods. While at first glance a FSO/PO/PC bears a resemblance to a microbiological criterion, they are actually risk management values upon which a microbiological criterion or other risk management metrics would be based. In general, a microbiological criterion would be more stringent than its corresponding PO because it is necessary to take into account the variability and uncertainty associated with the sampling and testing methods.

1.6 *Recent Examples of HACCP Continuing Evolution*

Although HACCP has evolved in more than five decades (Sperber and Stier 2009), the recent evolution of HACCP-based programs at the national level with the incumbent Preventive Controls for Human Food (PCHF) rule, as well as at the international level with the

forthcoming revision (CAC 2017a, 2017b, 2017c, 2017d) to the Codex Alimentarius General Principles of Food Hygiene (GPFH) (CAC/RCP 1-1969) and its current Annex: HACCP System and Guidelines for its Application (CAC 2003), is notable in the context of HACCP's history. During the past decade, there have been considerable advances in microbiological food safety risk assessments. Further, there has been an increase in the use of risk analysis to help study and make informed decisions about food safety. Although many risk assessments exist, a clear connection between risk assessments and the HACCP system has not yet been established. For example, Domenech and others (2008, 2009, 2011, 2013) used predictive quantitative risk modeling to assess the effectiveness of CCPs and HACCP prerequisites; however, these authors did not draw a clear connection to public health goals or food safety objectives, and instead directed the efforts toward reduction of expenses, quality approaches, and product liability issues. Currently, HACCP, the primary food safety risk management system used worldwide to ensure the safety of food (WHO 1997, 2001), is limited in its ability to quantitatively consider the impact that control measures that determine the stringency of HACCP programs for "significant" foodborne hazards have on risks to public health. The current hazard-based approach to HACCP does not allow a direct linkage between HACCP performance and improvements in public health (Buchanan and Williams 2013). Microbiological quantitative risk assessment models and models developed to predict microbial survival and growth may serve as an integral tool to evaluate, control, document, and even defend the safety measures designed into food products (Baker 1995).

The movement of regulatory agencies toward risk-based preventive control programs is re-emphasizing the criticality of being able to link HACCP and quantitative risk analysis. As a means of showing this need to shift from hazard-based to risk-based food safety systems, this chapter reviews some of the most considerable advances in food safety risk analysis, highlighting food safety risk management systems and the emerging use of risk management

metrics. To explore linking HACCP with risk analysis, a review of HACCP as a food safety system, its current limitations, and its origins and evolution from the perspective of systems engineering will be covered. A framework for linking risk analysis and HACCP using a risk-based metrics system as a means of linking HACCP and risk analysis via the use of risk management metrics will be introduced. The overall goal is to introduce the potential pathway toward the development of risk-based HACCP plans that can lead to programs that are more science-based, risk-based, and transparent.

1.6.1 Food Safety Modernization Act

The requirement for Preventive Controls for Human Food (PCHF) within "FDA Food Safety Modernization Act (FSMA)" (GPO 2011) implies the use of such risk-based tools to prevent foodborne illness and protect public health. PCHF-based food safety management is one of two final rules that have been released by the U.S. Food and Drug Administration (FDA) as "Current Good Manufacturing Practice and Hazard Analysis and Risk-Based Preventive Controls for Human Food" and "Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption." PCHF acknowledges the necessity of a risk-based approach to protect public health. It is noteworthy that the PCHF final rule only affects products under the jurisdiction of the FDA that are not already covered under existing FDA HACCP regulations. These factors all point to the need for traditional HACCP plans to evolve into risk-based HACCP systems in the near future. Currently, food scientists are increasingly familiar with risk assessment techniques, and this should be the starting point for moving HACCP to a risk-based systems approach.

1.6.2 Joint FAO/WHO Codex Alimentarius Commission

The current evolution of HACCP-based programs at the international level with the forthcoming revision (CAC 2017a, 2017b, 2017c, 2017d) to the Codex Alimentarius General

Principles of Food Hygiene (GPFH) (CAC/RCP 1-1969) and its current Annex: HACCP System and Guidelines for its Application (CAC 2003) represents a remarkable event in the history of HACCP and its evolution. An explanation of how a RB-HACCP approach could contribute to the evolution of HACCP-based systems is provided in Chapter 7.

1.6.3 Other Countries

In the European Union (EU), the majority of food businesses have to meet the requirements of Article 5 of Regulation (EC) No 852/2004 of the European Parliament and the Council on the Hygiene of Foodstuffs, which require food business operators to put in place, implement, and maintain a permanent procedure based on Hazard Analysis and Critical Control Point (HACCP) principles.

Other government authorities in developed countries, including Canada, Australia and Japan, have adopted or are adopting HACCP-based food safety control systems (Scott and Stevenson 2006). Due to the flexibility and applicability of the HACCP system to all kinds of food industries, HACCP requirements have been adopted not only by developed countries but also by many developing countries (Vidal and Cueva 2001). It would be impractical to list HACCP regulations for the many developing countries. However, as an example of HACCP regulations established in a developing country, HACCP is mandated in Peru under the legal requirements relative to the food industry that were published in the 1998 Government Supreme Decree No. 007-98-SA: Regulation on Sanitary Control and Surveillance for Food and Beverages ("Reglamento sobre Vigilancia y Control Sanitario de Alimentos y Bebidas"). This document states that companies manufacturing food or beverages should prepare a HACCP plan and implement it in their manufacturing processes.

1.7 *Summary and Overview of the Dissertation*

In sum, the hypotheses of this research are that the Critical Control Points (CCPs) are

vital steps in the process that significantly reduce the mean and/or variance of a hazard and, that these steps can be identified and quantified using risk assessment modeling techniques such as sensitivity analysis (SA) and what-if scenario analysis, while also providing a more objective means in considering Critical Limits (CLs). These hypotheses will be explained in detail in the next chapter.

The dissertation is organized into to seven chapters. The first chapter provides the impetus for this research by providing a broad overview of HACCP along with its limitations. Chapter 2 provides greater detail of the problem, the hypotheses, and the background for both case studies. Next, Chapter 3 presents the methodology including the Monte Carlo Simulations, Sensitivity Analyses, What-if scenarios followed by the statistical analyses and Fit-for-Purpose Assessment. Afterward, the case-studies risk assessment and results are presented respectively in Chapters 4 and 5. Chapter 4 presents a first look at the baseline quantitative microbial risk assessment for Frankfurters. Chapter 5 presents a similar assessment for Cold-Smoked Salmon. The Risk-based Critical Control Points for both products are determined separately in Chapter 6. The chapter begins determining CCPs for Frankfurters followed by CCPs for Cold-Smoked Salmon. The final chapter of the dissertation ends with general discussion on the evolution of HACCP. Based on the two case study results, Chapter 7 provides comparisons of the risk-based HACCP plan to traditional HACCP plans. The chapter concludes with a discussion on the need for future research and recommendations.

Chapter 2: Problem, Hypothesis and Approaches

2.1 *Statement of the Problem*

The current use of the Hazard Analysis Critical Control Point (HACCP) system as a food safety risk management tool is limited in its ability to quantitatively consider the impact of risk factors on foodborne hazards and to associate the stringency of HACCP programs with risks to public health that need to be controlled.

2.2 *Statement of Goals*

The overall goals of the proposed project are (1) to identify, evaluate, and recommend a set of risk assessment modeling tools that could be used to consider the quantitative impact of risk factors associated with foodborne hazards, and (2) to use some of those tools to more effectively link HACCP plans to food safety risk management metrics.

2.3 *Statement of Working Hypotheses*

- Critical Control Points (CCPs) are steps in the process that significantly reduce the mean and/or variance of a hazard.
- There are risk assessment tools such as sensitivity analysis and scenario analysis that provide a more objective means in considering Critical Limits (CLs).

2.4 *Summary of General Approach*

The general approach to achieving the goals of this research was to develop quantitative risk assessments for *L. monocytogenes* in two commercially manufactured, ready-to-eat foods: frankfurters and cold-smoked salmon. These risk assessments were then used to identify the CCPs associated with each of the products while considering CLs and

verification strategies that could be related to public health outcomes. The risk-based HACCP plans developed in this manner were then compared against traditional approaches to HACCP plan development.

These products were selected, in part, because they represent the extremes in the continuum of means for controlling *L. monocytogenes* in RTE foods. Frankfurters are produced in a manner that includes an overwhelming inactivation step (i.e., hot smoking) that effectively limits *L. monocytogenes* to subsequent re-growth events. Conversely, cold-smoked seafood has minimal inactivation steps in its manufacture and must rely on a combination of marginally effective treatments to control the survival and growth of the pathogen. Additional reasons for selecting these two products include:

- Availability of a robust literature on the characteristics and control of *L. monocytogenes* in both products.
- Availability of risk assessments that have been performed on at least some aspects of *L. monocytogenes* in these products.
- Availability of predictive microbiology models and supporting microbiological data for the behavior of *L. monocytogenes* in these products.
- Ongoing national/international concerns on the effective control of *L. monocytogenes* in both products.

This research was divided into seven phases. In the first phase, the conceptual models for the development of quantitative risk assessments for each product were defined.

The second phase was the development of a facility-specific quantitative microbial risk assessment (QMRA) model for *L. monocytogenes* on frankfurters in order to identify risk-based critical control points at the processing level. This took advantage, to the greatest

degree possible, of data acquired from collaboration with a high production frankfurter manufacturing facility. This assessment was augmented by additional data drawn from the literature and other risk assessments.

The third phase was the development of a QMRA model for *L. monocytogenes* in cold-smoked salmon. In this case, data was acquired from the scientific literature. The decision to rely on literature data was based on the substantially larger number of studies and corresponding supplemental data available, the greater diversity of processes within the cold-smoked salmon industry, the time constraints associated with trying to acquire sufficient data from multiple cold-smoked salmon manufacturers, and the small likelihood that most cold-smoked salmon processing plants, which are typically small businesses, would be able to supply the necessary data.

Since the primary purpose of using two product risk assessments was to develop HACCP programs, the models emphasize in-plant operations. The post-manufacturing steps of the process prior to consumption were also modeled, accounting for the variability in certain factors, particularly the post-manufacturing cold chain.

The fourth phase of the research was to use the risk assessment models to identify likely CCPs for each of the foods. This was done using sensitivity analysis to identify steps in the process with the greatest impact on the concentration of *L. monocytogenes*, i.e., the sensitivity analyses were used to examine the relative impact of the different unit operations on *L. monocytogenes* contamination rates in the final product. Based on the results of the sensitivity analyses, “what-if” scenarios were selected to examine and considered the impact of different process deviations. Based on these results, the individual steps were prioritized by loss of process control, thereby identifying CCPs.

The fifth phase compared the CCPs identified by this research with those derived by more traditional qualitative approaches. The differences between traditional and risk-based HACCP plans were explored.

The sixth phase employed additional "what-if" scenarios for the two products to determine the impact of various CL values for the identified CCPs. These additional scenarios provided a means of assessing the factors that need to be considered by HACCP plan developers to make informed decisions about the impact of the CL selection on both products, and on the overall HACCP plans' operational characteristics.

The final phase of the research determined how risk assessment models could be used to inform management decisions related to the sensitivity and frequency of microbiological verification testing programs based on a microbiological criterion, thereby confirming that the HACCP was functioning as intended. This included consideration of the establishment of POs for the food safety system to enable manufacturers to move to risk-based HACCP systems.

2.5 *General Background on* Listeria monocytogenes *as a Foodborne Pathogen*

The study of *L. monocytogenes* and listeriosis officially began in 1924 following the first confirmed diagnosis in a human at the end of World War I (Rocourt and Buchrieser 2007). The genus *Listeria* was described by Pirie in 1940. Although a few cases were reported before 1950, listeriosis has emerged as a major foodborne disease only in the past 30 years (Ryser and Buchanan 2013).

At the time of writing, the genus *Listeria* comprised ten recognized species (den Bakker and others 2014). *Listeria monocytogenes* (Pirie 1940) is one of six closely related species forming a clade that also includes *L. ivanovii* (Seeliger and others 1984), *L. innocua* (Seeliger 1981), *L. welshimeri* (Rocourt and Grimont 1983), *L. seeligeri* (Rocourt and Grimont 1983), and *L. marthii*

(Graves and others 2010). Species that are more phylogenetically distant (den Bakker and others 2010a, 2010b; Graves 2010) are *L. grayi* (Errebo Larsen and Seeliger 1966) and *L. rocourtiae* (Leclercq and others 2010). Other species currently recognized within the genus include *L. fleischmannii* (Bertsch and others 2013) and *L. weihenstephanensis* (Lang Halter and others 2013). In addition, *L. cornellensis*, *L. grandensis*, *L. riparia*, *L. aquatica*, *L. floridensis* are novel non-pathogenic species that were isolated from agricultural and natural environments in Colorado and Florida (den Bakker and others 2014).

Listeria monocytogenes is a ubiquitous pathogen, and has been isolated from soil, wastewater, decaying vegetation, stale water supplies, grazing areas, poorly prepared animal feed, intestines of healthy animals, intestinal tracts of healthy humans, 17 avian species, over 42 wild and domestic mammalian species, crustaceans, fish, oysters, ticks and flies, and a wide variety of raw and ready-to-eat (RTE) foods (Sauders and Wiedmann 2007). *Listeria monocytogenes'* association with various environments is one of the factors that allows RTE foods to become contaminated at multiple points along the processing and distribution chain before consumption (Gombas and others 2003). Cross-contamination has been reported as the major source of *Listeria* in cooked or otherwise processed seafood (Jinneman and others 2007). In fact, *L. monocytogenes* in the final product might be a result of persistent ribotypes in the processing environment (Lappi and others 2004). However, raw fish contaminated in the natural environment, which could subsequently affect the final product, cannot be completely ruled out as a source of contamination in a seafood processing environment (Jinneman and others 2007). A summary of the environmental parameters that affect the growth and survival of *L. monocytogenes* is shown in Table 2.1.

Invasive listeriosis, the primary disease manifestation associated with foodborne *L. monocytogenes*, is noted for its high case-fatality rate (CFR). Traditionally, the CFR is 20-30% (Mead and others 1999; Silk and others 2013), though lower CFRs have been associated with

specific outbreaks (Cartwright and others 2013). The latest epidemiological annual estimates in the U.S. show *L. monocytogenes* as the preeminent cause of hospitalization (94%) and the third leading cause of death (15.9%) among all other major foodborne pathogens under surveillance in the U.S. (Scallan and others 2011). The 11-year hospitalization rate between 1998 and 2008 was 60% (Cartwright and others 2013) whereas the three-year hospitalization rate between 2009 and 2011 was 93% (Silk and others 2013). Under a different methodology, Mead and others reported in 1999 that *L. monocytogenes* had the highest hospitalization rate (90%) and the second highest fatality rate (20%) among foodborne pathogens. The Centers for Disease Control and Prevention (CDC) estimates that the annual number of invasive listeriosis cases is 1,662, resulting in 1,520 hospitalizations and 266 deaths in the United States (Scallan and others 2011).

Epidemiologic data suggests that most listeriosis cases are foodborne (Schlech and others 1983; Mead and others 1999; ICMSF 2002; Scallan and others 2011; Cartwright and others 2013; Silk and others 2013). RTE foods have been estimated to be the vehicles for 99% of human listeriosis cases (Hitchins and Whiting 2001; Yang and others 2006; Scallan and others 2011). Listeriosis is considered a rare disease with an average annual incidence for 2004-2009 of 0.27 cases per 100,000 population (Cartwright and others 2013) and 0.29 cases per 100,000 population for 2009-2011 (Silk and others 2013). Listeriosis is primarily a threat to specific susceptible subpopulations such as the elderly (>60 years), the fetuses of pregnant women, and immunocompromised persons (ILSI/RSI 2005). As these at-risk populations continue to grow, a greater effort may be needed to achieve the Healthy People 2020 goal of 0.20 cases per 100,000 population.

The first recognized foodborne listeriosis outbreak in North America occurred in Nova Scotia, Canada in 1981 and was traced to contaminated cole slaw (Schlech and others 1983). In the United States, the first reported listeriosis outbreak occurred in 1983 and was

associated with pasteurized milk (Fleming and others 1985). Between 1983 and 1998, outbreaks of listeriosis associated with Mexican-style cheese (Linnan and Mascola 1988) and shrimp (Riedo and others 1994) were also documented. Several outbreaks of listeriosis in the U.S. have demonstrated a positive link between this pathogen and RTE processed meat products, including "deli" meats and frankfurters (Ross and others 2009). An epidemiological investigation identified turkey frankfurters as the likely vehicle for *L. monocytogenes* in a multistate outbreak in the U.S. in 1998 and 1999 (CDC 1999). Cartwright and others (2013) reported that a total of three outbreaks (13% of total), including the largest outbreak (n=108) during 1998–2008, were associated with frankfurters. However, since 1998, listeriosis outbreaks attributed to frankfurters and deli meats have become less frequent in the United States (Cartwright and others 2013). The reduction of *L. monocytogenes* in the RTE meat and poultry category likely shows the effects of regulatory initiatives such as the 1998 mandatory HACCP regulations for the meat and poultry industry.

The documented presence of *Listeria* in seafood prompted several studies of a variety of products from different geographic regions (Jinneman and others 2007). The results of these studies have been extensively reviewed (Dillon and Patel 1992; Ben Embarek 1994; Jemmi and Keusch 1994). Although seafood was the food category most commonly linked to foodborne outbreaks in the U.S. during 1998-2007 (Smith DeWall and others 2009), there have not yet been any documented listeriosis outbreaks associated with cold-smoked salmon in the U.S. (CDC 2016). Still, product recalls of cold-smoked salmon are commonplace (Marler 2013). Lightly preserved products such as smoked fish often are contaminated with *L. monocytogenes*, demonstrating a potential high risk of transmission of listeriosis (Ben Embarek 1994; Huss and others 2000; Norton and others 2001). In addition, outbreaks associated with similar products have occurred outside of the United States. For example, cold-smoked rainbow trout was identified as the source of a listeriosis outbreak in Värmland,

Sweden (Tham and others 2000) and was also implicated in an outbreak in Finland (Miettinen and others 1999). Most recently, using whole-genome sequencing, two listeriosis outbreaks were defined in Denmark during the period 2013–15 (Gillesberg Lassen and others 2016). Although no listeriosis outbreak associated with cold-smoked salmon has been documented in the United States (CDC 2016), and although outbreaks caused by frankfurters have become less frequent, listeriosis outbreaks continue to be a public health concern. For example, a 2011 outbreak attributed to whole cantaloupes from Colorado (CDC 2011) demonstrated that large outbreaks (n=147) can still occur. In total, 33 deaths from outbreak-associated cases were reported in 2011 (CDC 2012). In 2014 a listeriosis outbreak linked to caramel apples resulted in at least three deaths and 34 hospitalizations over 12 states (CDC 2015a). In another case, a complex outbreak investigation, conducted over several years, verified that the source of the listeriosis outbreak was ice cream and various other Blue Bell brand products. In this case, three deaths and ten hospitalizations over four years were reported in four states (CDC 2015b) Most recently, a multistate outbreak of listeriosis was linked to soft cheeses, with three deaths and 28 hospitalizations reported (CDC 2015c).

2.6 *Case Study One—Frankfurters*

Sausage is one of the oldest forms of processed food (FAO 1985), having been mentioned in Homer's Odyssey as far back as the 9th century B.C. (NHDSC 2015). Frankfurters and wieners are traditional meat products classified as cooked sausages (Ward 1911 cited by Kraig 2009). Frankfurters are usually pork sausages and wieners are pork and beef sausages that originated in Germany and Austria, respectively (FAO 1985). Frankfurters were popularized in the U.S. during the late 1800s (NHDSC 2015). Schwartz and others (1988) estimated that 20 billion frankfurters are consumed each year in the U.S., an average of 60 frankfurters per person approximately. Cooked sausages, which also include bologna and a variety of luncheon meats, are prepared from mixtures of comminuted beef, pork,

chicken, and/or turkey. Frankfurters can be made of beef only, a combination of beef and pork and, more recently, from poultry meat. If the frankfurter is not all-beef, pork and/or poultry are also included in the formulation (USDA 2013). Different variations have emerged using combinations of meat, fat, spices, and preservatives. Usually the composition of frankfurters varies depending on the particular brand and where it is produced and sold. Some of the common ingredients used to manufacture frankfurters include raw ground meat and fat, hydrolyzed soy protein, water, ice, lactate, diacetate, sodium chloride, sodium erythorbate, flavoring and spices including paprika and garlic powder. The product is typically made from meat trimmings, or, leftover cuts made from meats used primarily as steaks or pork chops. This section provides a brief description of the frankfurter manufacturing process, which is depicted in Figure 2.1.

The microbiological quality of the ingredients used in hot dog production plays a role in the safety of the final product. If bacteria or other microorganisms are present on the carcass, there is a risk of spreading the microorganisms throughout the meat when it is comminuted before emulsion (Güngör and Gökoglu 2010). In the frankfurter facility visited for this study, the meat pre-blend was prepared at a separate location and shipped under refrigeration to the manufacturing site. Once received and refrigerated, the meat and other ingredients are weighed to assure the proper balance of all ingredients, which varies depending on the type of frankfurter. This study considers only all-beef frankfurters with a "standard" size and formulation.

The curing process in frankfurter manufacturing has several functions, including to enhanced the flavor and color of the final product and inhibit the growth of undesirable microorganisms. Most importantly, curing extends the shelf life of the final product by preventing spoilage and inhibiting the growth of pathogenic *Clostridium* spp. Ingredients used for curing include salt, sodium nitrite, and curing accelerators such as sodium

erythorbate and ascorbate (Durand and others 2000). A high-speed, stainless steel chopper blends meat, spices, and curing ingredients into an emulsion. This emulsion is pumped into a stuffer, which stuffs the emulsion into cellulose casings which are twisted to form sausage links. Liquid smoke is applied to the frankfurter facility immediately before the heat treatment. Frankfurters are typically cooked to an internal temperature of ≥ 70°C to coagulate the protein, fix the smoke and cured color, and destroy pathogenic and spoilage microorganisms. Frankfurters are cooked to an internal temperature of 71.1 °C inside of controlled atmosphere smokehouses (FAO 1985). Cooking frankfurters for 30 seconds at 71.1°C (160 °F) would inactivate approximately 5 logs of *L. monocytogenes* (Mazzotta and Gombas 2001). The cooking temperature reached at the center of the frankfurters collected at the visited facility showed a minimum value of 71.1°C, a most likely value of 73.3°C, and a maximum value of 75.5°C. However, the cooking time at the continuous oven was 1 hour and 35 minutes for regular frankfurters from beginning to end. Although there are four different phases in the continuous oven with differentiated temperature and time ranges, the thermal treatment was considered overwhelming.

After reaching the desired internal cooking temperature, the links pass through a cold-water shower in the oven conveyor system, where they are rinsed with cold potable water. To continue chilling the product, the frankfurters are then brined by being submerged in acidified chilled brine at the end of the oven conveyor system. A detailed explanation of these steps and the frankfurter process is provided in Chapter 4. The frankfurter casings are then removed by passing the links through a high-speed peeler including a heated blade. The cellulose casing is split, stripped off, removed, and discarded using a hot blade in a steam chamber. The individual peeled links are then conveyed into the packaging line through a series of conveyors and slip sticks. Finally, the frankfurters are moved onto a selection belt where operators make a visual inspection and remove any broken or defective frankfurters.

Potential for contamination is not inconsequential during this part of the process. In large processing plants, around 700 frankfurters are peeled per minute. In these plants, processing equipment is often very large and difficult to disassemble for thorough cleaning and sanitation (Nowak and Krysiak 2005). Bacteria can contaminate the food if employees touch contaminated surfaces and then touch the equipment, or if they touch the frankfurters themselves (Nowak and Krysiak 2005). Contamination can also occur during packaging and/or handling, especially in plants that are not properly sanitized (Pal and others 2008).

Immediately following visual inspection, the frankfurters are vacuum packaged and x-rayed for metal and bone detection. Individual vacuum-sealed packages typically contain ten frankfurters. There is no post-packaging pasteurization or other intervention at this point in the process. The vacuum-packed frankfurters are then moved to refrigerated areas or storage coolers and loaded into the refrigerated trucks for distribution. At this point, temperature and times of transportation are recorded. Typical refrigeration temperatures during transport are 0-4°C (Rotariu and others 2014). Time-temperature abuse is an issue at the consumer level, not only in terms of home storage (EcoSure 2008) but with regards to reheating of an RTE food. The adequate handling and reheating of frankfurters is particularly important for susceptible populations such as pregnant woman, the elderly, and immuno-compromised people. In the original "Quantitative Assessment of Relative Risk to Public Health from Foodborne *Listeria monocytogenes* Among Selected Categories of Ready-to-Eat Foods" study (FDA/USDA 2003), conducted to estimate the per annum and per serving relative risk of causing listeriosis cases and deaths in association with 23 RTE food categories, frankfurters were analyzed in two different categories: reheated and not reheated. That study found that non-reheated frankfurters (together with deli meats) had one the highest predicted relative risks on a per annum basis (FDA/USDA 2003).

2.7 *Case Study Two—Cold-Smoked Salmon*

The smoking of fish is a traditional preservation technique for extending the shelf life of seafood. In the cold-smoking process, the fish is "produced by subjecting it to smoke at a temperature where the product undergoes only incomplete heat coagulation" (AFDO 1991). This means that unlike frankfurters, cold-smoked salmon (CSS) does not undergo a complete thermal inactivation during the process of smoking. Instead, control is achieved through a series of partial control steps. This broad definition of the cold-smoking process implies great variability among cold-smoking processes, even within the same facility, and exact processes depend mainly on the availability of equipment, the size and composition of the fish, and the demand of customers. A flow chart of the cold-smoked salmon process is depicted in Figure 2.2. This figure explains the process of cold smoking salmon, divided into modules, based on the literature including characteristics of the typical smoked seafood facility visited. The following section provides a general summary of the cold-smoking process and some important considerations for processing.

Cold-smoked fish processors receive salmon that is either wild-caught or farm-raised. In both cases, the fish is received either refrigerated with an internal temperature of ≤40°F (4.4°C) or frozen. Delays in the icing of fresh fish are a concern with fish of the Scombridae family (tuna, mackerel, bonito) due to the possibility of histamine poisoning (Ward and Hackey 1991). Most companies use a sensory evaluation of incoming scombrotoxin-susceptible fish. If sensory analysis points to a high biogenic amine level, analytical testing is performed. The visited plant processed both scombroid and non-scombroid finfish, and implemented two different HACCP plans for each of these categories. Although salmon fall under the non-scombroid category, temperature controls and freezing at the primary production level are considered beneficial. Freezing fish is primarily needed to ensure both control of *L. monocytogenes* and prevention of histamine formation. It is also needed as a

means for controlling parasites and the growth of other pathogenic bacteria or spoiling organisms. Currently, three-quarters of total U.S. seafood imports are frozen. Approximately half of the imports are produced by aquaculture (NOAA 2011). If designated for cold-smoked processing, wild-caught fish are frozen during the process to inactivate any parasites. As a control strategy against parasites, a number of time and temperature combinations have been recommended by the U.S. government for proper storage, such as holding the fish at a temperature below -4 °F (-20 °C) for 7 days or -31 °F (-35 °C) (internal) for 15 hours (FDA 2011). Alternatively, European Union regulations require freezing the fish at a temperature of no more than -4 °F (-20 °C) in all parts of the product for not less than 24 hours in order to control parasites (FDA 2013).

Thawing, washing, and rinsing of the fish is done under sanitary conditions and temperature control. After thawing, the fish are washed and rinsed thoroughly with potable water. Fish are butchered separate from the rest of the processing area. Gutting is performed in a manner that minimizes contamination from intestinal tract contents. After butchering and evisceration, the fish are washed and rinsed thoroughly, especially the body cavities, with continuously flowing or sprayed potable water. Dry-salting involves placing fish for a certain period of time in a dry mixture of salt and other ingredients. Brining is the process by which the fish are soaked in a solution consisting of water, salt, sugar, various spices and flavorings, phosphates, and, depending on the formulation and species of fish, additives such as sodium nitrite. Fish may also be brined by injecting them with a brine solution, either by hand or machine. Salting is done as uniformly as possible, with the correct amount of salt or brine solution absorbed into each piece of fish flesh. Salting times may vary from < 1 h to 24 h. The size of the fish pieces and the appropriate duration of salting are empirically determined. Fish processed with a dry-cure mixture are held under refrigerated conditions in the salt mixture.

A number of cold-smoking procedures involve a drying stage prior to the smoking. At the cold-smoking plant visited for the study, the “drying stage” involved rinsing and draining the fish after brining, as well as racking, hanging or equilibrating it prior to cold-smoking. In drying, the product is held at a specified temperature, often refrigeration temperature, for a specified time (e.g., 4 to 5 hours in the racking/equilibrating step) before the smoke is introduced. The parameters of this initial drying component include the type or species of fish, its fat content, and its moisture level. During this time, a pellicle forms on the outside surfaces of the fish pieces. In the cold-smoking step, the fish must be arranged to allow for uniform smoke absorption, temperature exposure, and drying. The smoke can be “natural” (generated), liquid, or a combination of the two. In the United States, cold smoking is seldom performed at temperatures above 100°F. In the case of the visited facility, the reported temperature used for their process was <90°F. The duration usually ranges from 6 to 12 h, however, in the visited facility the reported duration of the cold-smoking process was between 8 and 16 hours. After cold smoking, the fish are cooled to 50°F (10°C) within 3 h, and to 38°F (3.3°C) within 12 h (AFDO 1990).

After cooling, cold-smoked fish are often sliced or cut for portioning. A processor must have well-designed and comprehensive Sanitation Standard Operating Procedures (SSOPs) and follow Good Manufacturing Practices (GMPs) during the slicing, cutting, and overall handling of the product. During the visit to the cold-smoking processing plant, two different types of equipment were used to slice the cold-smoked salmon, which were divided into two different production lines. The new slicing equipment was designed to be disassembled, whereas it was not possible to disassemble the old equipment. Thus, the degree of difficulty in following the SSOPs was greater while cleaning and sanitizing the old equipment. Likewise, there were two different types of packaging equipment used during the visit to the cold-smoking processing plant, air packaging and vacuum packaging. The air

packaging equipment was used mostly for whole fish whereas the sliced cold-smoked salmon was vacuum packed. Cold-smoked fish is packaged using film with variable oxygen transmission rates (OTRs). Gas permeability is an important parameter, and its specifications are product- and use-specific. Typically, gas permeability specifications are established at ambient temperatures under moderate humidity conditions (for example, 23°C and 50% R.H.) using a variety of testing and verification methods. If the product contains the appropriate "salt" content, scientific data supports that the storage temperature should be maintained at a maximum of 40°F (4.4°C) at all times, and that the period of storage, distribution, and marketing should be a maximum of four weeks to ensure safety (Jahncke and Herman 2001). The required storage temperatures and times are to be labeled on the product.

Table 2.1 L. monocytogenes *growth and survival limits* by environmental factors*

Environmental Factor	Limits	
	Lower	Upper
Temperature (°C)	-2 to +4	~45
Salt (% water phase NaCl)	< 0.5	13 - 16
Water activity	0.91	>0.997
pH (HCl as acidulant)	4.2	9.5
Lactic acid (water phase)	0	3.8-4.6 mM, MIC[1] of undissociated acid[2] (800-1000 mM, MIC of sodium lactate[3])
Acetic acid	0	~20 mM (MIC of undissociated acid)
Citric acid	0	~3 mM (MIC of undissociated acid)
Sodium nitrite	0	8.4 – 14.4 µM (undissociated)

[*]Adapted from Ross and others 2009 summarized from Ryser and Marth 1991; ICMSF 1996; and Augustin and Carlier 2000.
[1]MIC: minimum inhibitory concentration, i.e., the minimum concentration that prevents growth.
[2] from Tienungoon 1998;
[3] from Houtsma and others 1993.

Figure 2.1 Frankfurters Process Flow Chart

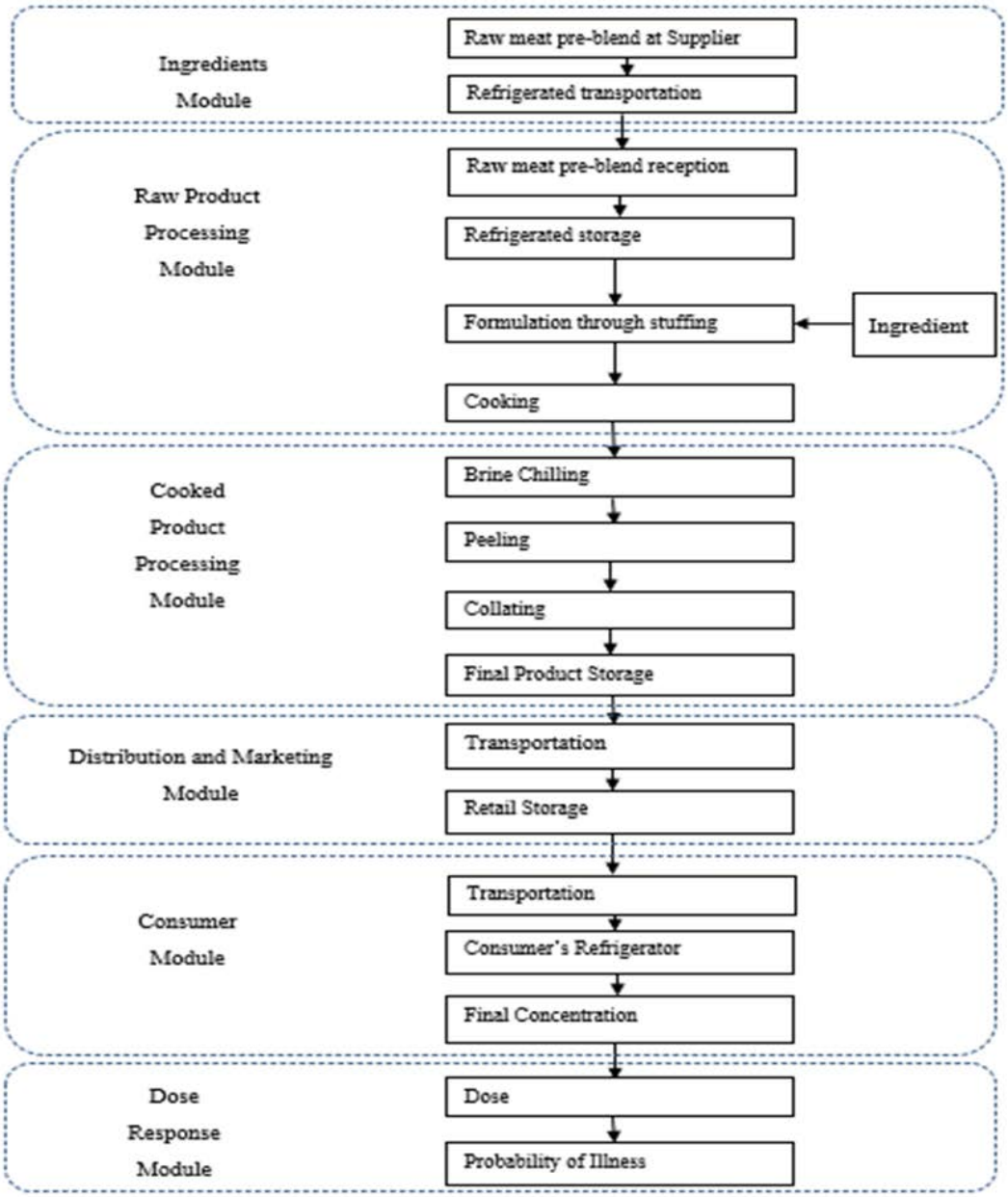


Figure 2.2 Cold-Smoked Salmon Process Flow Chart

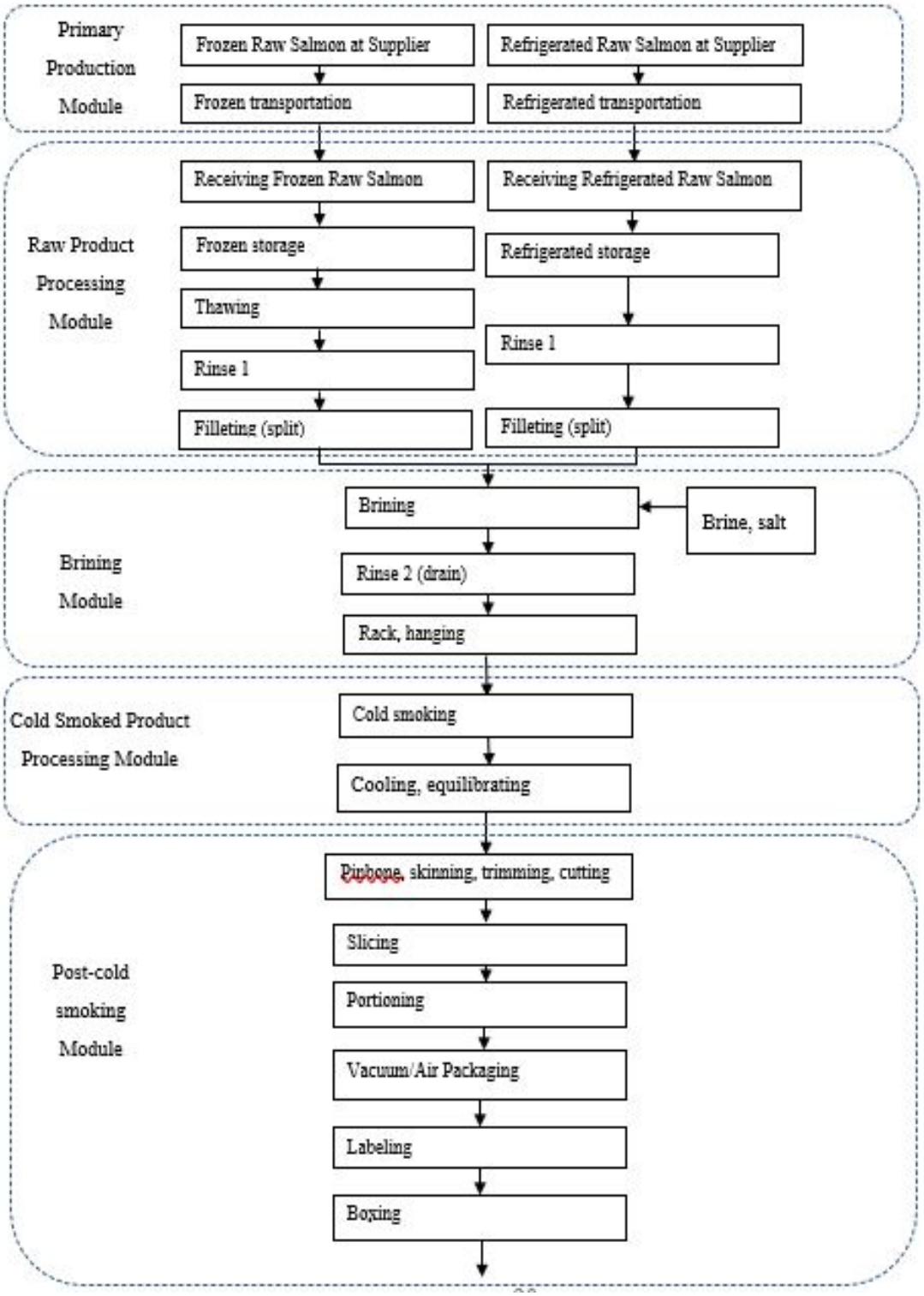
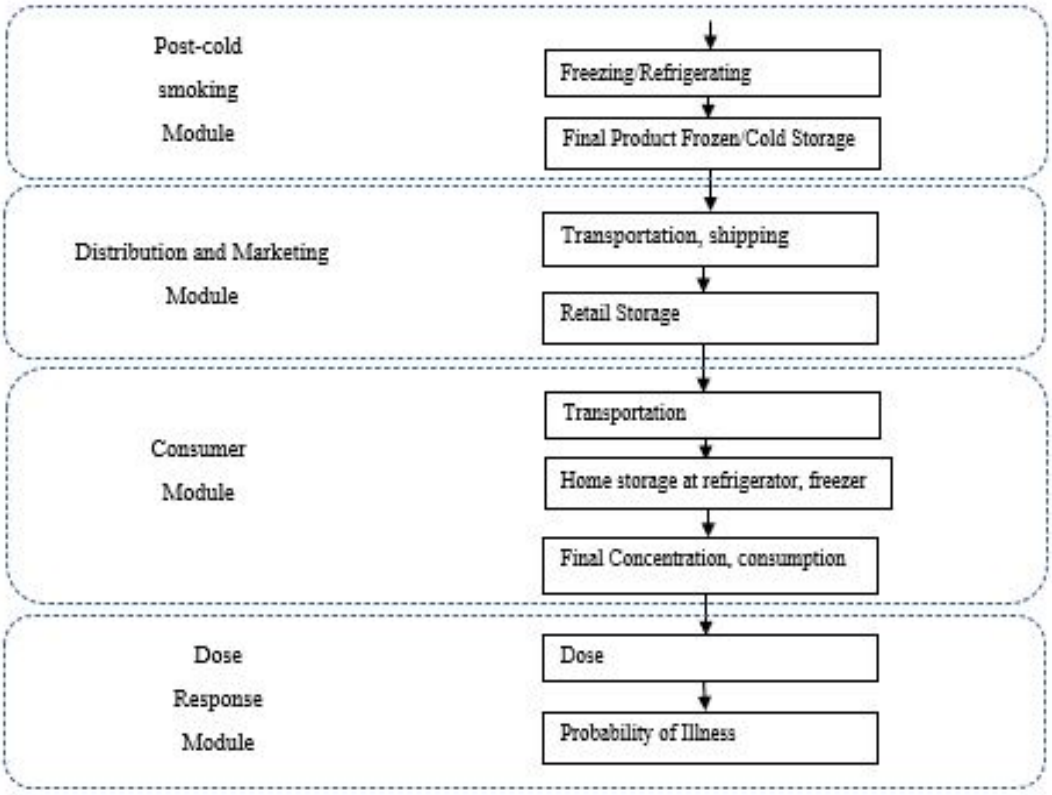


Figure 2.2 Cold-smoked Salmon Process Flow Chart (Continued)



Chapter 3: Methodology and Procedures

3.1 *Introduction to Research Methodology*

The development of risk assessment models followed the general framework of Codex Alimentarius (CAC 2015) and employed a modular approach based on a Product Pathogen Pathway (PPP) analysis (Buchanan 2001, 2008) and the Modular Process Risk Model (MPRM) (Nauta 2001, 2008). These models were then used to quantitatively assess the risk management decisions using the International Commission on Microbiological Specifications for Foods (ICMSF) conceptual equation for food safety risk management (ICMSF 2002) presented in Chapter 1. The ICMSF equation was used to consider the performance of both the overall food safety system and individual subsystems (i.e., unit operations and production modules). This chapter provides an overview of the process modeling approach, incidence data selection procedures, data collection procedures, and quantitative analyses procedures (e.g., sensitivity analysis, what-if scenario analyses).

3.2 *Overview of Process Modeling Approach*

Models to quantitatively describe the pathogen pathways throughout different kinds of processes in the food production chain are increasingly being used within the quantitative microbial risk assessment (QMRA) framework (Whiting and Buchanan 1997; Bemrah and others 1998; Cassin and others 1998a; Harnett and others 2001; Nauta 2001; Lindqvist and others 2002; Nauta 2008). These models may cover only specific parts of the "farm to fork" continuum or the whole pathway. Pathways are often complex and may require different modeling approaches. Cassin and others (1998b) developed the Process Risk Model, which was subsequently improved by Nauta (2001, 2008) who introduced a modular approach through the Modular Process Risk Model (MPRM). This methodology has been applied in several food chain risk assessments (Nauta 2001, 2002, 2005; Nauta and others 2005).

Equivalently, the Product Pathogen Pathway (PPP) analysis is "an evaluation of the entire process from start to finish that examines the risk of adverse effects within a population and models specific combinations of pathogens and products" (USDA 2016). The PPP analysis has also been described as a technique that can be used to examine factors that contribute to risk over the course of a particular segment of the path from farm to fork. The procedure makes it possible to "quantify the importance of contamination sources, the effectiveness of interventions, the comparative effectiveness of different control measures, the likely effect of performance criteria or standards, and the importance of complying with the criteria" (Buchanan 2008). The PPP analysis is an approach used in the *Vibrio parahaemolyticus* risk assessment developed by the FDA (FDA 2001, 2005; Buchanan 2008) and provides a systematic evaluation of the factors affecting the pathogen in the product and the sequence of events leading to consumer illness. The approach used in the current study was to develop modules for the different phases of product manufacturing, distribution, marketing, and consumption using a combined MPRM/PPP analysis, with the "Modular Product Pathogen Pathway (MPPP)" risk assessment as a framework to incorporate the ICMSF equation (ICMSF 2002) in a standardized way for each module. Previous researchers have used the ICMSF conceptual equation (Walls and Buchanan 2005; van Schothorst 2009; Zwietering 2010) for several purposes, but none appear to have taken full advantage of the Food Safety Objective and Performance Objective concepts as they relate to the HACCP system. The ICMSF equation provides the opportunity not only to link the HACCP system with public health outcomes but also to do so in a quantitative manner. Using the MPPP framework to incorporate the ICMSF equation in food processes in order to develop risk-based HACCP plans represents one of the contributions of this study. This study builds upon past research by using the ICMSF equation as a framework to determine critical control points. Each of the components of the ICMSF equation, such as initial contamination level (H_0), kinetic inactivation ($\sum R$), and growth ($\sum I$), were modeled at each module of the process and the highest variability was determined through

sensitivity analysis. The modules/steps with the highest variability were identified as CCPs. In addition, "what-if" scenario analyses were generated and run within this framework to quantitatively determine the CLs.

3.2.1 Modular Product Pathogen Pathway (MPPP) Analysis

The description of the pathway from production to consumption by means of a PPP analysis (Buchanan 2001, 2008) included in the exposure assessment section accounts for *Listeria monocytogenes* along the food pathway. The food pathway was represented as a chain of modules, similar to the approach used in the Modular Process Risk Model (MPRM). As previously mentioned, this combined approach was called a "Modular Product Pathogen Pathway (MPPP)" risk assessment in which the key steps or unit operations, from the primary production through processing to the point of consumption, were modeled. Each food process was divided into modules that provided manageable and flexible subsystems for developing a quantitative microbial risk assessment (QMRA) model for the overall system. The modules followed the pathway of the pathogen. The predicted levels of *L. monocytogenes* in frankfurters and cold-smoked salmon were determined at the end of each module along the pathway.

A schematic representation of the modules for the frankfurters and cold-smoked salmon processes are shown in Figures 3.1 and 3.2, respectively. For example, in the frankfurters case study (Figure 3.1), the Ingredients Module considered the factors influencing the incidence of *L. monocytogenes* in the meat and other ingredients up to the time of reception. The Raw Product Processing Module recognized factors associated with the handling and processing of frankfurters up to the thermal treatment. The Cooked Product Processing Module took into account factors associated with the handling and processing of frankfurters up to the point of packaging and storage of the final product before it leaves the manufacturing facility. The Distribution and Marketing Module accounted for factors

associated with the handling and storing under refrigeration of frankfurters until the product is purchased by the consumer. The Consumer Module took into consideration factors between the purchase and consumption of the product such as in-home storage and methods of preparation (e.g., reheating frankfurters prior to consumption). Similarly, the cold-smoked salmon process was divided into a number of modules: Primary Production, Raw Product Processing, Brining, Cold-Smoked Product Processing, Post-cold Smoking, Distribution and Marketing, and Consumer. Each of the modules are illustrated in Figure 3.2.

The MPRM approach identifies each module as one of six basic processes providing a unified and structured approach to food chain exposure assessment (Nauta 2008). These six fundamental processes or events may affect the transmission of "any microbial hazard in any food process" (Nauta 2008). There are two "microbial" basic events in the process and four "food-handling" events. Both microbial and food-handling events will be briefly addressed below. In addition, a section on dose-response models was also included.

3.2.1.1 Basic Microbial Events

There are two microbial basic events in a food process: growth and inactivation. These microbial events strongly depend on the characteristics of the microbial hazard and directly relate to the ICMSF equation. In addition, a variety of models can be applied for each basic event. For example, for microbial growth, the modified Gompertz equation in combination with a Response Surface Model (Buchanan and Phillips 2000) were used to describe the growth of *L. monocytogenes* in frankfurters. The modified Gompertz model (1994) is described by the following equation: $\text{Log } x(t) = A + C \exp\{-\exp[-B(t-M)]\}$. The Buchanan and Phillips (2000) quadratic response surface models (RSM) for the aerobic growth of *L. monocytogenes* along with the variable ranges for its parameters are included in Table 3.1. This RSM is, in part, the basis for the widely used USDA Pathogen Modeling

Program. In the case of cold-smoked salmon, the Hwang (2009) model was used to estimate the growth of *L. monocytogenes*. The Hwang (2009) model was selected because it is specific to cold-smoked salmon and is a simple polynomial model which includes the effect of smoke components (phenol), temperature, and NaCl on the growth of *L. monocytogenes*. Both "microbial" basic events, growth and inactivation, were considered during the processes. In assessing the inactivation of *L. monocytogenes* in frankfurters during processing, a thorough literature search of models of thermal inactivation for *Listeria* spp. was performed. Numerous researchers have described the thermal inactivation of *L. monocytogenes* in different substrates. D-values and models for the thermal inactivation of *Listeria* spp. in various foods were reviewed and graphed to identify the most pertinent model for thermal inactivation for this organism. As a result, it was determined that Murphy and others (2002) was the most pertinent model, as it was specific to frankfurters. It is important to note that D-values are substrate and temperature specific. The Murphy model was not only developed for the exact substrate but also had the closest temperatures to the frankfurter process at the visited facility. The parameter of estimates for log10D versus heating temperatures and the z-values in frankfurters are shown in Table 3.2. By contrast, with regards to the inactivation of *L. monocytogenes* in cold-smoked salmon due to the phenolic compounds applied during the cold-smoking process, distributions were generated based on available data published by Porsby and others (2008) and Montazeri and others (2013).

3.2.1.2 Food-handling Events

Nauta (2008) differentiated four food-handling events: mixing, partitioning, removal, and cross-contamination. In the frankfurter making process two key handling events, the mixing or blending of ingredients before emulsification and the partitioning or separation into individual frankfurters, occur before thermal treatment. Two of the other classes of food-handling events, removal and cross-contamination, occur during and after the thermal

treatment, respectively. In cold-smoked salmon processing, two key food-handling events are cross-contamination and partitioning. Both of these events can occur at different steps of the process, for example, during slicing and portioning of the cold-smoked salmon before packaging.

In the case of cross-contamination, models for the two ready-to-eat products of interest were identified in the literature. For frankfurters, the cross-contamination model only included the post-heat treatment parts of the process. For this purpose, the *Listeria* cross-contamination model of Schaffner (2004) was used. In the case of the cold-smoked salmon, the used model was that of Aarnisalo and others (2007) for *L. monocytogenes* during slicing of 'gravad' salmon. This model was selected for its specificity to salmon and salmon fillet composition.

There are two instances in which cross-contamination and removal occur almost simultaneously in the frankfurter process. The first takes place at the peeling step, which is a form of "removal" or elimination (Nauta 2008). The equipment, consisting of a heated blade and steamed peeler chamber, is generally used to peel off the frankfurter casings. In the past, nonheated blades were used to peel frankfurters at processing facilities. It is noteworthy that peeling frankfurters using an unheated blade has been implicated as a cause of contamination (Wenger and others 1990) in an outbreak of *L. monocytogenes* previously mentioned in Chapter 2. Therefore, the peeling step is not only a removal event but is also a potential cross-contamination event. The second instance of combined removal and cross-contamination occurs during the discarding of broken frankfurters at the end of the line just before packaging. Rejection of frankfurters that do not pass visual inspection is performed by operators wearing stainless steel mesh gloves. As a result, the removal of broken frankfurters can also be considered an indirect transmission cross-contamination via the mesh gloves of the food operators. These stainless steel cut-resistant mesh gloves, which also have the

function of protecting the fingers of food operators, inevitably come into contact with some frankfurters located nearby the broken ones as they pass visual inspection. The different modalities of cross-contamination in this operation may include a combination of several basic processes/events; however, there is not enough data available on the transmission dynamics of the hazard to include a detailed analysis. These are characteristics of complex steps that may be regarded as black boxes, in which case the transmission is to be represented by linear models on a log scale (Nauta 2008).

Similarly, the MPRM implies formalities that while time consuming may not have enough impact on the overall system to be relevant to the present QMRA. However, they provide a clear structure for modeling food processes using a modular approach. The "food-handling" events were simplified, whenever possible, using assumptions in the context of a MPPP risk analysis. For example, because the overwhelming thermal treatment in the frankfurter process will most likely eliminate the hazard under investigation, certain steps prior to the thermal inactivation were simplified.

3.2.1.3 Dose-Response Model

Dose-response or hazard characterization has been described as the qualitative and/or quantitative evaluation of the adverse health effects associated with the hazard (CAC 1999; 2015). For the purpose of the present Quantitative Microbial Risk Assessment (QMRA) studies, the hazard of concern is *L. monocytogenes.* Many dose-response models have been developed for foodborne pathogens, for example, Log-Normal, Log-Logistic, Simple Exponential, Flexible Exponential, Beta-Poisson, Beta-Binomial, Weibull-Gamma, and Gompertz. These models have been summarized by FAO/WHO (2004) and adapted from Holcomb and others (1999). A section on dose-response model was included since the original Process Risk Model (PRM) developed by Cassin and others (1998a) is divided into two parts. The first part describes the behavior of the microorganism during processing,

handling, and consumption of the food, including exposure assessment. The second part corresponds to the dose-response model, which estimates the health risk associated with consuming a food.

The present study used the FAO/WHO (2004) exponential dose-response model, which was connected at the end of the exposure assessment to estimate the probability of illness for *L. monocytogenes* as part of the risk characterization component of the risk assessment. The FAO/WHO (2004) model approach was based on two population groups: a susceptible population and a healthy population. It was assumed that 80% of the total population was healthy and would thus be much less likely to acquire listeriosis after exposure. Accordingly, 20% of the population was assumed to be at high risk for listeriosis. The exponential dose-response model requires appropriate r-values and a dose that is dependent on the serving size. The r-value used for the susceptible population was 1.06×10^{-12}. The 5^{th} and 95^{th} percentiles for this r-value were 2.47×10^{-13} and 9.32×10^{-12}, respectively. The healthy population r-value used was 2.37×10^{-14} with 5^{th} and 95^{th} percentiles of 3.55×10^{-15} and 2.70×10^{-13}, respectively. Thus, the probability of a single ingested bacterial cell causing listeriosis was 1 to 2 orders of magnitude greater for the susceptible population than for the healthy population. The serving sizes needed in this exponential equation for the two RTE products were based on the distributions provided by the FDA/USDA (2003) risk assessment.

3.3 *Data Collection Procedures*

Three data collection visits were made to a frankfurter facility on the following dates:

Visit 1: Wednesday, October 28 to Friday, October 30, 2009.

Visit 2: Monday, January 11 to Friday, January 15, 2010.

Visit 3: Monday, January 17 to Friday, January 21, 2011.

A one-day visit to a cold-smoked salmon facility was made:

Visit 4: Wednesday, August 27, 2012.

Visit 4 to the CSS facility was not a data collection visit and consisted of a detailed explanation of the process in situ.

Regarding visits to the frankfurter facility, detailed explanations of the process *in situ* were provided during each visit, and company improvements made to the frankfurter processing line between 2009 and 2011 were noted. During the first visit, one-week's worth of data per season over a one-year period was requested. Four week's worth of data on the frankfurter process was randomly selected and the files were provided. The same files corresponding to the year 2009 through October (winter data obtained from the beginning of 2009) were made available on each data collection visit. Each daily file contained abundant material that was processed by reviewing and typing the data into a new electronic file. The management decided to add a pasteurization step for the brine at the beginning of 2010. Because of this variation in the process, it was decided to use data collected prior to this major change (i.e., data collected in 2009).

This study was observational in nature, and was accomplished, in part, based on three data collection visits to a frankfurter processing plant and one visit to a cold-smoked salmon facility. The data collection procedure for the frankfurter study included a random selection of one week of all data collected by the processing facility per season (i.e., one week of summer, fall, winter, and spring for a total of four weeks of data). This data was obtained and collected in situ. A list requesting particular data was given to the processing plant prior to the first visit. A summary of the data collection survey can be found in Appendix 1.

3.4 *Incidence Data Selection Procedures*

Scientific literature on the incidence of *L. monocytogenes* in raw ground beef, frankfurters, raw salmon, and cold-smoked salmon was reviewed and available studies were consolidated, as appears in Tables A2.2, A2.3, A2.4, and A2.5, respectively. To determine the estimation of incidence from the literature, we employed a scheme for weighting the relative importance of the studies based on several criteria. This allowed maximization of literature data while considering changing practices in the meat and seafood industries. The criteria for inclusion or exclusion of studies from the literature for estimating the weighted incidence of *L. monocytogenes* and the rationale for the incidence data selection procedures are explained in Appendix 2.

3.5 *Quantitative Analyses*

This section includes Monte Carlo simulations and Latin Hypercube Sampling, Sensitivity Analysis, What-if scenario analysis, Statistical analysis, and Fit-for-purpose analysis.

3.5.1 Monte Carlo Simulations and Latin Hypercube Sampling

In the present study, the risk assessment models were simulated using Latin Hypercube Sampling techniques with @Risk 7.5 (Palisade Corporation, Ithaca, NY). @Risk 7.5 was used to run the frankfurters and the cold-smoked salmon model simulations. Previously published reports (Pradhan and others 2010; Sanaa and others 2004) pertinent to this study used 100,000 iterations. Therefore, all models were simulated with a minimum of 100,000 iterations per scenario. To more accurately estimate the number of iterations needed, however, the convergence function was used. The level of convergence range was between 1 and 5%. After running the simulations using @Risk for each level of convergence, the number of iterations was obtained. With the established information, a graph using the

number of iterations as the dependent variable "y" was generated. The convergence is generally an exponential function. With the convergence function and its corresponding R^2, the number of iterations needed to reach a specific level of convergence was calculated.

3.5.2 Sensitivity Analysis

Sensitivity analysis (SA) is the assessment of the impact of changes in input values on model outputs (Cullen and Frey 1999). Thus, SA can include the study of how uncertainty in the output of a model can be apportioned to different sources of uncertainty in the model inputs (Saltelli and others 2000). SA is used to increase the confidence in the model and its predictions by providing an understanding of how the model dependent response variables respond to changes in the independent variables inputs. In addition, SA can be helpful in verification of a model. It can be used to evaluate the robustness of risk estimates and management strategies to model input assumptions, and can aid in identifying data collection and research needs (Frey and Patil 2002). In this research, SA was used to determine the Critical Control Points of two distinct food processes. Using data from the simulations, sensitivity analysis showed the relationships among the inputs to help account for output variation. This same type of analysis was applied at the subsystem level to estimate the variation of the outputs at different stages of the process.

There are three different methodologies for SA: 1) mathematical, which includes the Nominal Range Sensitivity Analysis and the Differential Sensitivity Analysis (DSA); 2) statistical, which includes Sample and Rank Correlation Coefficients, Regression Analysis, Rank Regression, Analysis of Variance, Classification and Regression Tree, Sobol's Indices, and Fourier Amplitude Sensitivity Test (FAST); and 3) graphical, which includes scatter plots and conditional sensitivity analysis (Frey and others 2004).

This study used the software @Risk 7.5 for the frankfurters and the cold-smoked salmon model as an add-in for Excel. The methods employed in @RISK are multivariate

stepwise regression and rank order correlation. Under circumstances where regression performs poorly (e.g., low R^2 value), correlation is the preferred method to study the relations between input variations and the effect on (explanation of) the output variations. Spearman's correlation coefficients indicate the relative relationship among the inputs in order to determine the strength of the explanatory relationship. For model parameters, the same distribution used in the model simulation was also used in the SA. Different values for the parameters (input values) were sampled from the corresponding distributions and the effect of the inputs was evaluated on the risk of listeriosis and at the subsystem level to determine CCPs.

When coefficients are small, i.e., closer to zero than 0.1 in absolute value, they contribute little to the output variation and their significance is negligible. The SA ranking indicated which inputs have the greater impact at the top of the list (tornado graph). Lesser impact inputs were ranked lower or at the bottom of the tornado graph. Thus, the rankings show relative impact. From the present study perspective, the inputs listed at the top of the tornado chart had a net bigger impact on the output if they were inputs that could be influenced in some way. If they were controlled inputs, making changes to them would produce a bigger resulting impact on the output variation and value as compared to items further down on the tornado chart.

Beginning with the latest version of @RISK, in addition to the "Inputs ranked by effect on output mean" tornado, @RISK 7.5's has been updated with a "Contribution to variance" tornado graph capability that helps explain how much of the variance in the output variable is attributable to each individual input (Palisade Corporation 2017). This is directly related to the working hypotheses explained in Chapter 2. Both of these specific two types of tornadoes were selected for each module of the RTE products with the research hypothesis in mind. Following the logic of the present research, the ability to find the most critical or top-

ranked RB-CCPs would depend on the criteria used to determine low relative risk for listeriosis. In this research, the criteria used were drawn from Carrington and others (2004). Thus, the low risk category was defined as <1 case per billion servings.

3.5.3 "What-if" Scenario Analysis

"What-if" scenario analysis was used to help inform decisions related to establishing critical limits. This was based on the components of the ICMSF equation. Different scenarios were calculated regarding initial contamination (H_o), reduction of microbial populations ($\sum R$), and potential increases ($\sum I$) of *L. monocytogenes* at different stages of the processes of interest. To achieve specific POs and the final PO determined by the required FSO, the component of the ICMSF equation that could have the greatest impact in achieving the final PO was determined based on the sum of all kinetic inactivation ($\sum R$). Therefore, several simulations of the models were run to estimate the likely effect and impact of mitigation strategies by changing one input parameter at a time (keeping all other factors fixed) and measuring the change in the model output. For example, in the case of the frankfurter process, two different scenarios were modeled, frankfurters reheated and frankfurters non-reheated by the consumer. By contrast, twenty-five scenarios, including examples of intervention strategies based on the most important factors found by the sensitivity analysis, were developed for the cold-smoked salmon process.

3.5.4 Statistical Analyses

Standard statistical tests used included Analysis of Covariance (ANCOVA), one-way Analysis of Variance (ANOVA), and Tukey's test. Correlation among means was detected using a two-tailed Pearson correlation test. Statistics were analyzed using SAS for Windows (version 9.1, Cary, NC). Statistical significance was defined as $P \leq 0.05$. Statistical analysis

using SAS was conducted to process the data collected from the frankfurter processing facility.

3.5.5 Fit-for-Purpose Assessment

The main purpose of developing the frankfurter and cold-smoked salmon QMRA models was to create risk-based HACCP plans for these processes, in other words, to determine critical control points using sensitivity analysis and establish critical limits on public health impacts as determined by what-if scenario analysis. Baseline models, having as a goal for each product the obtaining of incidence distributions similar to the ones calculated from the literature, were developed. Many simulations of each baseline model and different scenarios were performed. Fit-for-purpose analysis encourages application of a level of rigor commensurate with the intended purpose and use of an assessment (Meek and others 2013). To that end, the input data, probability distributions, and mathematical models for the frankfurter and cold-smoked salmon processes were meticulously selected with the main purpose in mind, to develop risk-based HACCP plans. In addition, verification of the achievement of food safety performance metrics through the establishment of Microbiological Criteria was determined (FAO/WHO 2004; Whiting and others 2006; Zwietering and others 2010).

Table 3.1 *Quadratic response surface models for generation time and lag phase duration for the aerobic growth of* Listeria monocytogenes

Ln(GT) = 21.2574 – 0.2643T + 0.00404S – 5.2054 P + 0.0189 N + 0.00709TP – 0.00252PN + 0.00265T^2 + 0.000129S^2 + 0.3746P^2

Ln(LPD) = 26.0899 – 0.1901T + 0.0545S – 6.3831P + 0.0167N + 0.000201TS + 0.0000232TN – 0.00729SP – 0.00229PN + 0.0019T^2 + 0.000098S^2 + 0.4784P^2

Variable Ranges (Aerobic Growth)

Temperature (T): 4° – 37 °C

pH (P): 4.5 – 7.5

Sodium Nitrite (N): 0 – 150 μg/g

Sodium Chloride (S): 0.5 – 10.5%

Water Activity: ≥0.92

Source: Buchanan and Phillips (2001)

Table 3.2 Parameter of estimates for log10D versus heating temperatures and the z-values of Listeria innocua *in frankfurters at 55° to 70°C*

Intercept	8.7625
Slope	-0.1318
r^2	0.99
z (°C)	7.59

Source: Murphy and others (2002)

Chapter 4: Baseline Quantitative Microbial Risk Assessment (QMRA) for *Listeria monocytogenes* on Frankfurters

A baseline model for frankfurters was developed for the purpose of incorporating quantitative risk assessments into the HACCP plan. This chapter describes the baseline model for *Listeria monocytogenes* in frankfurters including a detailed explanation of each step of the process, following the modular product pathogen pathway (MPPP) described in Chapter 3. This chapter details the risk modeling for the frankfurter process and *L. monocytogenes*, including estimates of the initial level of contamination in raw meat pre-blend, models of *L. monocytogenes* growth and inactivation, and estimates of *L. monocytogenes* contamination levels in frankfurters. The description of the manufacturing process of frankfurters was based on a review of manufacturing specifications and augmented by the literature and by visits to a frankfurter processing facility as described in Chapter 3. The baseline model for this product was based largely on the specific practices of the particular facility, which was visited as an example of a generic frankfurter process. This processing plant provided a HACCP plan for frankfurters which was used to compare the results obtained in this research. When applicable, the baseline model used a set of simplified steps and unit operations throughout the different modules of the processes. The scope of the risk modeling and HACCP application spanned from post-slaughter to consumption, allowing determination of "risk-based" critical control points (CCPs) and critical limits. The results from sensitivity analysis and what-if scenario developed from the baseline model, including consideration of the intervention strategy of reheating frankfurters at the consumer level to reduce the risk of *L. monocytogenes*, are covered in Chapter 6.

4.1 *Exposure Assessment for the Baseline QMRA Modular Product Pathogen Pathway (MPPP) for Frankfurters*

The frankfurters process was divided into modules that provide manageable and flexible subsystems for developing a quantitative microbial risk assessment (QMRA) model for the overall system. This QMRA begins with an introductory worksheet (Figure 4.1) which summarizes key information necessary to follow the sequence of the model. The frankfurter process was divided into six modules: (1) ingredients, (2) raw product processing, (3) cooked product processing, (4) distribution and marketing, (5) consumer, and (6) dose-response. Each module is described below and illustrated in Figure 4.2. The exposure assessment followed the pathway of *L. monocytogenes* through the first five modules of the frankfurter process using tools such as predictive microbiology and the risk assessment MPPP model framework described in Chapter 3. The frankfurter conceptual model worksheet and its associated calculations is provided in Figure 4.3. Distributions used for each module throughout the worksheets were consolidated in tabular form and included in Table 4.4.

4.1.1 Ingredients module

The ingredients module represents the earliest stage in the production of frankfurters. This module consists of two steps: (1) raw meat pre-blend provided by a supplier, and (2) refrigerated raw meat pre-blend transported from the supplier to the manufacturing facility. The baseline model assumes refrigerated conditions with potential growth of *L. monocytogenes*. The ingredients module worksheet is illustrated in Figure 4.4.

4.1.1.1 Raw meat pre-blend at supplier

The first stage describes the main ingredient—raw meat—in the frankfurters' formulation. When the raw meat pre-blend leaves the supplier, the current baseline model starts. The initial level of *L. monocytogenes* in the raw ground meat, including fat tissue,

represents the main potential source of this pathogen in the incoming ingredients. We provide further details on estimating the initial levels of *L. monocytogenes* in raw ground meat in the next section (4.1.1.1.1).

In addition to beef pre-blend, other frankfurter ingredients include hydrolyzed soy protein, water, ice, and food-grade granulated salt blend (including sodium chloride, sodium nitrite, sodium erythorbate, sodium lactate, potassium lactate, flavoring, and spices such as paprika and garlic powder). Although it was assumed that the raw ground meat pre-blend was the main source of *L. monocytogenes*, it is important to recognize that any of these ingredients could be a potential source of this pathogen. For example, hydrolyzed soy protein could be a potential source of *L. monocytogenes* because *Listeriae* are relatively common in soil, and therefore it is possible for these organisms to find their way into soy processing factories (FDA 2015) and soy proteins products as a post-pasteurization contaminant. Estimating initial levels of *L. monocytogenes* in other frankfurter ingredients is covered in more detail in section 4.1.1.1.2.

4.1.1.1.1 Estimating initial levels of *L. monocytogenes* contamination in raw meat pre-blend

To produce *Listeria*-free raw meats is a challenge since *Listeria* spp., including *L. monocytogenes*, are virtually endemic to slaughterhouse environments (Kornacki and Gurtler 2007). To accurately estimate the initial overall level of contamination of the main ingredient of the frankfurters' raw meat pre-blend, one ideally would need know the exact composition of the raw meat and the results from microbiological testing for *L. monocytogenes*. In addition, it would be helpful to know whether any antimicrobials or bacteriocin-producing cultures were added at the supplier level since these treatments could reduce the levels of *L. monocytogenes*. For example, Buchanan and Klawitter (1992) tested *Carnobacterium piscicola*, reclassified as *Carnobacterium maltaromaticum* (Mora and others 2003), and found that the strain LK5 was most effective when the background microflora of the foods

was low. In sterile raw ground beef, strain LK5 inactivated the organism at 5°C and prevented its growth at 19°C. Bacteriocin-producing lactic acid bacteria have also been used in frankfurters to control the growth of *L. monocytogenes* (Berry and others 1991; Amézquita and Brashears 2002). Specific microbiological data related to the meat pre-blend were not available at the facility level, and as a result, the distribution of *L. monocytogenes* in ground beef was developed using literature data. The risk assessment was restricted to "all beef" frankfurters. The initial level of contamination with *L. monocytogenes* in ground beef was estimated based on the USDA Baseline Survey Results for Ground Beef and the estimated national mean levels of selected bacteria in raw ground beef produced under federal inspection (USDA 1996). The USDA baseline survey used a total of 563 samples of raw ground beef from which 99 samples were found to contain *L. monocytogenes*, indicating an incidence of 18%. Further quantitative analysis of the ground beef indicated that of the 99 samples, 90.4% had fewer than 30 cfu/g and overall only 3 of the 563 samples had greater than 110 *L. monocytogenes* per gram, which was the upper limit of detection used for testing (USDA 1996; ICMSF 2002). A best fit distribution was developed based on this data and is shown in Table 4.1. This distribution was used as the initial level of contamination of the raw meat.

Since the impact of the level of contamination of incoming material on product safety is dependent on the design of the subsequent food processes (Zwietering and others 2010), and since frankfurter processing includes an overwhelming thermal treatment, the model indicated that the degree of contamination associated with the raw ingredients would have a minimal impact on the levels of *L. monocytogenes* after the thermal treatment. Thus, baseline risk assessment was primarily a function of the post-thermal processing recontamination of the product. A more detailed explanation is forthcoming in section 4.1.2.5.2.

4.1.1.1.2 Estimating initial levels of *L. monocytogenes* contamination in other ingredients

As previously mentioned, the main ingredient in the frankfurter formulation is raw ground meat pre-blend. The other ingredients are hydrolyzed soy protein, water, ice, sodium chloride, sodium nitrite, sodium erythorbate, flavoring, and the spice pre-blend that includes paprika and garlic powder. The spice pre-blend and sodium nitrite are discussed further in this section. Since the ingredients are comminuted to form the frankfurter emulsion, and subsequently heated, *L. monocytogenes* survival is unlikely.

While the specific spice blend can vary from formulation to formulation, it typically contains paprika and garlic powder. The spice pre-blend used for the frankfurter emulsion in the visited processing facility was reported to be irradiated. Irradiation of spices, which has been shown to be an effective method of reducing microbiological populations, is especially recommended for processes without a thermal treatment. Normal irradiation treatments provide nearly commercially sterile spices (Tainter and Grenis 2001). Since the spice pre-blend used for the frankfurters at the facility was irradiated, no contamination with *L. monocytogenes* was assumed. It is worth noting that most of the primary recalls of spices identified by the FDA between 1969 and 2003 were due to *Salmonella* contamination (Vij and others 2006; FDA 2013). The only recall associated with *L. monocytogenes* occurred in fresh bay leaves (Hogan 2011), an ingredient usually not included in frankfurter formulations. Hence, spices do not appear to be a likely source of *L. monocytogenes*. In addition, some spices may inhibit or even inactivate selected pathogenic bacteria and fungi. For example, mace, bay, and nutmeg extracts at levels less than 125 ppm have been shown to inhibit *C. botulinum* toxin production in turkey frankfurter slurries (Hall and Maurer 1986). Clove, mustard, garlic, and onion added at 0.5% (Bahk and others 1990), and rosemary added at 1% (Pandit and Shelef 1994), were found to have inhibitory effects on *L. monocytogenes*. In addition, cilantro (6%), sage (1%), oregano (0.1 to 0.7%), thyme (0.1%), and cinnamon

(0.1%) showed listericidal activity (Aureli and others 1992; Hefnawy and others 1993; Gill and others 2002). The specific spices in the spice pre-blend were not known, as they were proprietary. The spice pre-blend was assumed to have no inhibition on *L. monocytogenes.*

Another ingredient added during the formulation step is sodium nitrite, which is frequently used to preserve meat and fish (Lado and Yousef 2007). Sodium nitrite has more than one function in frankfurters; it influences the formation of the characteristic color and flavor of the frankfurter, and also has a bacteriostatic effect on spoilage organisms and *Clostridium botulinum*. This curing agent slightly inhibits growth of *L. monocytogenes* (Buchanan and others 1989). Lag phase and generation time increases as the nitrite concentration rises from 0 to 150 ppm in nutrient broth. Inhibition increases when the addition of nitrite is combined with low temperature, pH, or oxygen level, or when the concentration of sodium chloride in the medium increased (Buchanan and others 1989). Nitrite antilisterial activity has mainly been reported at pH $\leq$5.5. At pH 6.3, combining 103 ppm sodium nitrite and 3.5% sodium chloride in meat did not control growth of *L. monocytogenes* at 32°C (Glass and Doyle 1989). The mechanism of nitrite action against *L. monocytogenes* in processed food is unclear. The mechanism of nitrite inhibition of *S. aureus* has been hypothesized to involve a disruption of glucose catabolism (Buchanan and Solberg 1978; Fang and others 1985). Studies focused exclusively on the mechanism of nitrites against *L. monocytogenes* were not found in the literature. However, Ngutter and Donnelly (2003) have documented nitrite injury of *L. monocytogenes* in frankfurters. Nitrite could also be combined with other antimicrobials. For example, Nitrite (30 ppm) did not increase the listeriostatic activity of sodium diacetate in turkey slurries (pH 6.2) (Schlyter and others 1993). Viability of *L. monocytogenes* at 4°C decreased up to 3.7 logs in 12 days in BHI (pH 5.5) supplemented with nitrite (125 ppm) and one or several of the following compounds: sorbate (0.3%), lactate (4%), nisin (400 IU/mL), and polyphosphate (0.5%) (Buncic and

others 1995). The nitrite concentration generally used in frankfurters was 50 µg/g approximately and was represented within the model by a distribution. The assumption used in the model was that the contamination with *L. monocytogenes* originated from the raw ground beef meat pre-blend (i.e., the main ingredient), and not from other ingredients.

4.1.1.2 Refrigerated transportation

"Refrigerated transportation" describes the shipment of raw meat pre-blend from the supplier to the processing facility at refrigeration temperature. The level of *L. monocytogenes*, when present in the raw meat pre-blend, depends upon the storage temperature and duration, pH, and salt concentration, among other factors. The duration of this transportation from the supplier to the processing facility was approximately seven hours and was represented by a distribution. The baseline model assumes refrigerated conditions with a potential growth of *L. monocytogenes*. Figure 4.5 shows, at the end of the ingredients module, the output of the level of *L. monocytogenes* (Log CFU/g) modeled in Excel using @Risk.

4.1.2 Raw product processing module

The raw product processing module consists of the following four steps: (1) receive refrigerated meat pre-blend, (2) place in refrigerated storage, (3) formulate through stuffing, and (4) cook the product. Each of these four steps is described below. The baseline model consists of refrigerated product at the beginning of the module, representing some growth of the pathogen, which was then mitigated by cooking at the end of the module. The raw product processing module worksheet is presented in Figure 4.6.

4.1.2.1 Receiving refrigerated meat pre-blend

The reception of the refrigerated meat pre-blend at the processing facility describes the arrival of the incoming refrigerated meat pre-blend from the supplier. At the visited facility, domestically procured meats were only purchased from federally inspected facilities. Shipments from suppliers arrived in sealed trailers to ensure integrity. Internal temperatures

of raw materials were checked. Other facilities may include imported meat or meat pre-blends.

4.1.2.2 Refrigerated storage for meat pre-blend

The refrigerated storage encompasses the time between receipts of the raw meat pre-blend through formulation of the final emulsion. The processing facility visited controlled their room temperatures during this step of the process. The most likely temperature and duration of the refrigerated storage was 5°C and 48 hours. These values were represented by distributions within the model. Temperature and duration served as inputs to model the growth of *L. monocytogenes*. A description of the growth model used for this *L. monocytogenes* follows.

4.1.2.2.1 Modeling *L. monocytogenes* growth under refrigeration

Various primary and secondary growth models for *L. monocytogenes* were evaluated. A primary model for microbial growth aims to describe the kinetics of the process with as few parameters as possible while still accurately defining the distinct stages of growth. In contrast, secondary models describe the effect of environmental conditions (e.g., physical, chemical, and biotic features) on the values of the parameters of a primary model. This distinction is consistent with the widely accepted terminology introduced by Whiting and Buchanan in 1993 (McKellar and Lu 2004). One commonly used example of a primary model is the Gompertz equation (Gompertz 1815, 1825), which was originally developed to describe human mortality as a function of age (Causton 1977 cited by Li and others 2007). The Gompertz equation was first utilized to fit microbial growth curves by Gibson and others (1987). Over the last two decades, the Gompertz model has been modified and expanded in such a way that new bacterial growth curve models have been developed based on this modification. The Buchanan Three-Phase Linear Model (Buchanan and others 1997a) was used to describe the three phases for *L. monocytogenes*: lag phase, exponential growth phase,

and stationary phase. In addition, the modified Gompertz model (Gibson and others 1987) was used to estimate the growth of *L. monocytogenes*. The secondary model utilized was the modified quadratic response surface model (RSM) by Buchanan and Phillips (2000). This RSM is, in part, the basis for the widely used USDA Pathogen Modeling Program. This RSM model was used to estimate the generation time (GT) and lag phase duration (LPD) for the growth of *L. monocytogenes*. The aerobic RSM model along with the variable ranges for its parameters was included in Table 3.1.

4.1.2.3 Formulation through stuffing refrigeration

Once received and stored under refrigeration, the meat and other ingredients are weighed to assure the proper balance of all ingredients depending on the style and formulation of the frankfurters. Only the "standard" formulation for all-beef frankfurters produced at the visited facility was studied. A high-speed, stainless steel chopper blends meat, spices, and curing ingredients into an emulsion. This emulsion is pumped into a stuffer, stuffed into cellulose casings, and twisted to form sausage links. These steps were simplified and represented within the raw product processing module.

4.1.2.4 Cooking

The thermal treatment associated with the frankfurter process using data collected at the processing facility, in accordance with the protocols from Chapter 3 and standard thermal processing calculations (Marks 2010), provided quantitative estimates of the degree of protection achieved by the cooking step, thereby showing that the risk reduction obtained from the thermal treatment of frankfurters is enough to ensure the effective elimination of *L. monocytogenes* contamination under both normal and extreme conditions immediately after the cooking step at the processing facility. In fact, the thermal treatment parameters used by the facility were overwhelming. The oven pertinent to the data for this study consisted of equipment for continuous, conveyorized thermal processing. The temperatures and duration

of the treatment were recorded following the protocol described in Chapter 3 and using the survey instrument developed prior to the visit (Appendix 1). The parameters used in the model were described as distributions. Frankfurters are typically cooked to an internal temperature of ≥ 70°C to coagulate the protein, fix the cured color, and destroy pathogenic and spoilage microorganisms. Cooking effectively renders the sausage free of *L. monocytogenes* immediately after cooking. Post-processing contamination, however, can still occur and, in rare instances, has led to outbreaks discussed earlier in Chapter 2. In fact, epidemiological data from the Centers for Disease Control and Prevention (CDC) showing an apparent association between listeriosis and undercooked frankfurters has prompted several studies examining the thermal resistance of *L. monocytogenes* in this type of sausage. Zaika and others (1990) prepared frankfurters from a sausage emulsion inoculated to contain ~10^8 CFU/g of *L. monocytogenes*. After stuffing, all frankfurters were thermally processed (without smoke) according to a standard commercial heating schedule. The USDA found that *L. monocytogenes* populations decreased approximately 1000-fold in frankfurters that were heated to an internal temperature of 71.1°C (160°F). Based on this data, cooking frankfurters to an internal temperature of 71.1°C would probably eliminate this level of *L. monocytogenes* (<10^3 CFU/g) that could possibly occur in raw frankfurter emulsions (Farber and others 2007). Compliance guidelines for meeting lethality performance standards for certain meat and poultry products (USDA 1999a) detail the cooking requirements to attain the prescribed lethality. In the processing facility visited, frankfurters were fully cooked on controlled wet/dry cycles until they reached an internal temperature of 73.3°C (164°F) on average, resulting in an overwhelming heat treatment. The results obtained reinforced the assumption that concerns related to *L. monocytogenes* in frankfurters are associated with post-thermal processing contamination (FDA/USDA 2003). This model also served as the basis for further consideration of recontamination points and other factors contributing to the association of *L. monocytogenes* with frankfurters.

4.1.2.4.1 Modeling *L. monocytogenes* inactivation in frankfurters

A thorough literature search was performed for models of thermal inactivation of *L. monocytogenes*. Numerous researchers have described the thermal inactivation of *L. monocytogenes* in different substrates. D-values and models for the thermal inactivation of *Listeria* spp. in many foods were reviewed and graphed to identify the most pertinent model for thermal inactivation of this organism (data not shown). It was determined that the model of Murphy and others (2002) was the most pertinent model as it was specific for frankfurters. It is important to note that D-values are substrate specific and temperature specific. The Murphy model was not only developed for the exact substrate but also had the closest temperatures to the process in the visited facility. Although the Murphy and others (2002) model used *L. innocua* instead of *L. monocytogenes*, it was determined that as a surrogate of this pathogen, *L. innocua*, had a similar heat resistance (Doyle and others 2001). The results obtained by Gaze and others (1989) and Murphy and others (2000) supported the use of this surrogate strain. The parameters for $\log_{10}$ D-values versus heating temperatures and the derived z-values for *L. innocua* in frankfurters at 55°C to 70°C are depicted in Table 4.3.

4.1.2.4.2 Smoking

Although not absolutely required, frankfurters and other similar sausages are frequently hung in smoking rooms either before or after cooking. Alternatively, commercially available liquid smoke products can be added to the sausage emulsion or applied directly to the surface of frankfurters before or during heating (Farber and others 2007). The visited facility applied liquid smoke to the frankfurters immediately before the heat treatment. In any event, besides imparting a pleasant smoked flavor to the finished product, some smoke components (i.e., formaldehyde, acetic acid, creosote, and phenols with high boiling points) are actually bacteriostatic and/or bactericidal toward many microbial contaminants (Farber and others 2007). Lingbeck and others (2014) indicated that liquid smoke is an effective antimicrobial in food systems and has several advantages over traditional smoking techniques

including ease of application, speed of smoking process, and omission of hazardous polycyclic aromatic hydrocarbons. Liquid smoke may be used as a component of a hurdle system for food preservation. Table 4.4 shows the antimicrobial effect of liquid smoke against *L. monocytogenes* in frankfurters. As described earlier, the processing facility visited showered the surfaces of the frankfurters with liquid smoke immediately before administering the overwhelming heat treatment. Liquid smoke was applied to the surface of the links through drenching in a closed environment, including a pre-rinse liquid smoked drench and a drip off. Although there might be residual effects from the application of liquid smoke, it was assumed that smoking would not affect the levels of *L. monocytogenes* in frankfurters within the present baseline model. It is worth noting that the subsequent peeling step of the process would facilitate removal of much of the residual smoke from the surface of the frankfurters. In a standard frankfurter process, the smoking step would usually occur before or during the thermal treatment (Farber and others 2007) with the purpose of fixing the smoke to the frankfurters. However, liquid smoke could also be applied after the thermal treatment. For example, Martin and others (2010) sprayed liquid smoke on frankfurters after the peeling step, thus providing an additional 1-log reduction of *L. monocytogenes* on the inoculated frankfurters. Martin and others (2010) further concluded that liquid smoke was an effective antimicrobial for frankfurters as it also suppressed growth during the extended shelf life of the product. The effect of the smoking step in the process is further discussed in Chapter 6.

4.1.2.4.3 Impact of cooking on frankfurters on levels of *L. monocytogenes*

Frankfurters are cooked smoked sausages. Applying a thermal treatment has more than one purpose including (1) formation of crust or a dense layer of coagulated protein, as shown in Figure 4.8, (2) inactivation of pathogens and most spoilage organisms, and (3) fixation of characteristic cured meat (Farber and others 2007). In frankfurters, all ingredients are finely blended to form the sausage emulsion, which is then stuffed into artificial casings

to form the links. Regardless of the equipment used (e.g., continuous or stationary) to achieve uniform thermal transfer, strings of frankfurter links are organized into neat columns hanging from a stick prior to entering the oven.

The strings of frankfurter links are cooked to achieve an internal minimal temperature of 71.1°C (160°F) as required by USDA. At the processing facility visited, product temperatures reached on average 73.3°C (164°F) during the thermal treatment using the continuous equipment. The cooking temperatures were carefully monitored and verified by trained/authorized personnel. After the overwhelming thermal treatment, the frankfurters model was truncated.

4.1.3 Cooked product processing module

The cooked product processing module occurs immediately after the thermal treatment and has the following five steps: (1) chilling, (2) peeling, (3) collating, (4) packaging, and (5) storing the final product. Each of these steps is explained below. The cooked product processing module worksheet is shown in Figure 4.9.

4.1.3.1 Chilling

After cooking, frankfurters are usually carefully cooled, packaged, and shipped to wholesale and retail markets, during which time they are refrigerated (Farber and others 2007). Cooling or chilling in the facility visited occurred in two phases: (1) cold water rinse and (2) acid brine chill. It is important to note that cooking by itself does not satisfy U.S. regulations; chilling the products is also required (Waters 2010). The detailed chilling requirements to attain the prescribed lethality are described in the literature (USDA 1999b).

4.1.3.1.1 Cold water rinse

After cooking, the links pass through a shower where they are rinsed with cold, potable water. The potable water use for the final rinse was assumed to be not contaminated

with *L. monocytogenes*. To continue chilling the product until it reaches 30°F, the frankfurters are then brined for at least 14 minutes.

4.1.3.1.2 Acid brine chilling

The second phase of chilling, acid brine chilling, is performed for at least 14 minutes and is used to reduce the temperature of the product to 30°F. In the visited frankfurter facility, the chilled brine was acidified with citric acid. The facility originally used recycled or recirculated non-pasteurized brine with citric acid at >0.5M. The brine was the environment in which *L. monocytogenes* had to survive at very low temperatures. Although relatively high concentrations of citric acid (>0.5 M) contain listeriostatic activity, lower concentrations can be utilized by *L. monocytogenes* depending on the pH and water activity of the brine (Young and Foegeding 1993; Buchanan and Golden 1994). In addition, bactericidal activity of acid increases with temperature (USDA PMP 2003 cited by Lado and Yousef 2007). When listeriostatic doses of organic acids are used, storage at refrigeration temperature is essential to prevent further growth of the pathogen (Islam and others 2002). Under listericidal conditions, however, refrigeration diminishes acid lethality. The growth rate of *L. monocytogenes* in the presence of organic acids varies markedly with the type and concentration of acid and pH. For example, acetic and lactic acids (50 mM) inhibited growth of the pathogen at 37°C when the pH was 4.7, but not when it was 6.0 (Young and Foegeding 1993).

4.1.3.2 Peeling

Skinless frankfurters are produced by mechanically peeling the casing from the sausage after cooking (Farber and others 2007). After the frankfurters leave the brining area of the oven conveyor system, they are transported to the peeling area. Specifically, the product is removed from the oven hooks, dropped onto a stainless steel table, and fed into a casing peeler where the cellulose casing is slit and removed from the exterior of the product.

The frankfurter casings are removed by passing the links through a high-speed peeler. The cellulose casings are split, removed, and discarded using a hot blade in a steam chamber. In the past when a hot blade and steam chamber peeler were not used, contamination occurred in the processing environment, specifically at the peeling step, i.e., after the cook step, which would inactivate *L. monocytogenes*.

Environmental investigations of a turkey frankfurter plant, whose product was linked to a case of listeriosis, found that contamination of the majority of frankfurters occurred at a single point during the peeling step of the process, prior to packaging (Wenger and others 1990). In addition, the presence of *L. monocytogenes* of the same serotype and isoenzyme type as the case strain in ready-to-ship products during the four-month period after the initial occurrence suggested persistence or reintroduction of this isoenzyme type in the processing plant (Wenger and others 1990). The presence and persistence of *L. monocytogenes* in processing areas has proven to be a considerable challenge to regulators and the food industry in the United States, as evidenced by the contamination of ready-to-eat meat and poultry products after preparation of the finished product but before packaging. Such contamination led to three large, multistate listeriosis outbreaks over 12 years (Farber and others 2007). Most recently, the recall of over 270,000 pounds of frankfurters (USDA 2016) as a precautionary measure to prevent listeriosis, shows that *L. monocytogenes* in frankfurters is still an issue that needs to be addressed at the facility level through a robust HACCP plan. In addition, a sound process design and good hygienic practices (GHP) are key elements to assure the safety of this type of ready-to-eat product in the United States.

4.1.3.3 Collating

After the peeling step, the individual peeled links are collated and channeled along a series of in-feed conveyors and slipsticks to the packaging line. For the purpose of this research, potential cross contamination with *Listeria* from contaminated food contact surfaces

was considered to be transferred on the surface of the frankfurters. The total surface area of the equipment in direct contact with the frankfurters was calculated. Potential growth under refrigeration was also considered during this step.

4.1.3.4 Packaging

Packaging is the step in which the frankfurters, while being divided into individual packages of approximately one pound, are (1) visually inspected, and (2) vacuum-sealed packed. Packages include approximately nine frankfurters in each and some exudate. In addition, the packaging step of the process included (3) labeling, (4) x-ray screening of the packages soon after they were sealed, and (5) boxing of the packages. A detailed description of these five sections follows.

4.1.3.4.1 Visual Inspection

Frankfurters are processed products that are treated listericidally by heat but are subject to potential recontamination during subsequent handling. This is particularly true during the visual inspection and manual removal of the visually non-compliant product. Immediately before the frankfurter packages are sealed, the frankfurters are moved to a selection belt where operators visually inspect and remove any broken or defective frankfurter, touching them with special protective stainless steel wire mesh gloves to assure the packages to be sealed contain the correct number of visually compliant frankfurters.

4.1.3.4.2 Vacuum-sealed packaging

Packaging creates a barrier with the environment, and therefore, it can accurately be said that the safety of a product is only as good as the safety, wholeness, and integrity of the packaging that contains it. This relates to the packaging technology as well as the permeability and reliability of the materials used. Processing plants use either vacuum or air packaging for frankfurters and most RTE products. For the baseline model, vacuum packaging was used. A review of the available literature on vacuum packaging of frankfurters

showed that packaging per se cannot eliminate or reduce the hazard of interest. In fact, growth of *L. monocytogenes* is not inhibited in food that has been packaged under vacuum (Hudson and Mott 1993). On the contrary, vacuum packaging has been found to increase the level of *L. monocytogenes* in several studies. For example, Glass and Doyle (1989) reported that *L. monocytogenes* at ~0.01 cfu/g can proliferate on vacuum-packed, artificially contaminated retail frankfurters during storage at 4.4°C, and increase by 2 to 5 logs after 4 weeks. It is noteworthy that these samples were found organoleptically acceptable after the 4 weeks, which may imply that the packaging used in this study was preventing the growth of spoilage organisms. Indeed, the CO_2-enriched atmosphere that is created within a meat pack can inhibit the normal spoilage flora and select for organisms such as lactic acid bacteria (Farber 1991). This may imply, in the absence of spoilage organisms and low levels of lactic acid bacteria, a potential Jameson effect which may be favorable for *L. monocytogenes* and could potentially explain the growth that occurs under such conditions. It is noteworthy that *L. monocytogenes* grows well under aerobic and anaerobic conditions and at refrigeration temperatures (Jinneman and others 2007).

Concerns have been raised about the ability of *L. monocytogenes* to outgrow the normal spoilage flora on modified atmosphere (MA-) packaged foods. In addition, the relatively long shelf life of MA-packaged foods can allow psychrotrophic foodborne pathogens such as *L. monocytogenes* to grow to high levels (Mano and others 1995). These properties make *L. monocytogenes* a potential threat to the safety of foods packaged under vacuum or modified atmospheres (Church and Parsons 1995). However, combining different hurdle technologies in addition to vacuum packaging with specific characteristics (e.g., 100% CO_2) may suppress the growth of *L. monocytogenes* (Szabo and Cahill 1999). It was assumed that there was no recontamination of the product with *L. monocytogenes* during packaging

itself which occurred after the visual inspection and handling. However, growth under refrigeration was considered.

4.1.3.4.3 Labeling

The packaging itself could include “code dated” labeling or the product could be transported to an area where the label, including lot number and expiration date, is applied to each package. No growth in this step of the process was assumed. However, labeling could represent a bottleneck in some instances and delays could occur at this step of the process. It was assumed that the time at which the product was held at refrigeration temperatures was too short to permit the growth of *L. monocytogenes*. It is important to recognize that the packages must contain a “keep refrigerated” statement (e.g., “Important, keep refrigerated until used.”) on the finished product to ensure that temperature controls are applied during the distribution and consumer phases. Frankfurters represent a possibly serious hazard to young children, and therefore, in addition to all required labeling, it would be useful to consider adding a special warning label for children.

4.1.3.4.4 X-Ray screening

After the packages are fed into vacuum packaging equipment and sealed in a consumer sized package (immediate container), they are transported in a conveyor belt and passed through an x-ray unit to undergo further screening for metal particles, bones, and other hard foreign materials that might have unintentionally ended up in the frankfurter packages. The x-ray screening reduces the probability of consumers finding physical hazards in the final product by detecting pertinent foreign material and discarding non-compliant packages prior to boxing. No growth of *L. monocytogenes* in this step of the process was assumed.

4.1.3.4.5 Boxing

After each package is x-rayed, the finished product packages are then checked for correct labeling and quality attributes and packed into boxes. These boxes are coded with the

appropriate labeling information (i.e., lot number, expiration date). The boxes are then transported to a refrigerated warehouse or final refrigerated storage where they are palletized and identified with a handling unit tag. It was assumed that no growth of *L. monocytogenes* took place during boxing because the duration at which the product was held under refrigeration was too short to permit the growth of this pathogen. However, delays could occur at this step of the process, and therefore it is important that boxes contain a "keep refrigerated" statement to ensure that temperature controls are applied throughout retail distribution.

4.1.3.5 Final product storage

In the baseline model, it was assumed that the final product was stored in a refrigerator and that growth of *L. monocytogenes* was possible during the period of final refrigerated storage. Individual packages containing nine frankfurters (approximately one pound per package) are vacuum-sealed, packaged, and boxed. The boxes with packaged frankfurters are moved to the final storage coolers where they remain until they are shipped to retail distribution centers or wholesale markets.

4.1.3.5.1 Estimating levels of *L. monocytogenes* contamination on frankfurters

The distribution of frequency and levels of *L. monocytogenes* contamination in frankfurters was estimated from studies and surveys carried out worldwide over the last 20 years (Appendix 2). These studies (see Table A2.3) indicated a substantial variability in both the frequency and extent of contamination, with incidence rates ranging from 0% to 45%. Several studies revealed incidence of *L. monocytogenes* in frankfurters, with most ranging from 0 to 12.5% (Ng and Seah 1995; Samelis and Metaxopoulos 1999; Levine 2001; Wallace and others 2003). The prevalence of *L. monocytogenes* in processed RTE meats is of greater concern than contamination of raw meats. Several studies have investigated the incidence of *L. monocytogenes* in sausages. Among those, the study by Wallace and others (2003) was

considered to be the most relevant source of data for vacuum-sealed packages of frankfurters since the number of samples collected (n=32,800) was sufficient and data was volunteered and collected specifically for frankfurters and *L. monocytogenes* from 12 U.S. commercial manufacturers over a two-year period. The 12 producers included nine large and three small plants located in 10 states. In total, 532 of 32,800 (1.6%) packages of frankfurters tested positive for *L. monocytogenes*. This incidence included all of the individual processing facilities with a minimum incidence of 0 and maximum incidence of 16% (plant 133) (Wallace and others 2003). The cumulative frequency for the incidence of *L. monocytogenes* on frankfurters based on Wallace and others (2003) is displayed in Appendix 2 (Figure A2.1). The literature for the incidence of *L. monocytogenes* on frankfurters was reviewed and available studies were consolidated in Appendix 2 (Table A2.3).

The calculated total incidence of *L. monocytogenes* from all other studies including Wallace and others (2003) was 1.95% which is slightly higher than the incidence calculated from the study by Wallace and others (2003). This calculated value is higher because the other studies are older, and, as explained in the previous chapter, there has been a reduction in the occurrence of *L. monocytogenes* in the meat industry including frankfurters. Studies other than Wallace and others (2003) report a minor effect in the overall incidence due to their combined low number of samples (see Appendix 2). It is noteworthy that Wallace and others (2003) is not a quantitative study; other studies were key references to determine the distribution of the levels (CFU/g) of *L. monocytogenes* in frankfurters (e.g., Wang and Muriana 1994). The probability distribution for the level of *L. monocytogenes* in contaminated frankfurters is reported in Appendix 2 (Figure A2.2).

4.1.4 Distribution and marketing module

The distribution and marketing module was divided into two steps, (1) refrigerated transportation from the processing facility to the retail distribution center, and (2) refrigerated retail storage. The distribution and marketing module worksheet is depicted in Figure 4.10.

4.1.4.1 Refrigerated transportation to retail

The frankfurters are shipped refrigerated from the processing facility to retail markets. Since this ready-to-eat product was assumed to be refrigerated, growth of *L. monocytogenes* was a concern at this level; thus, adequate refrigerated transportation would be critical to avoid temperature abuse during the distribution process. In addition to adequate temperature controls, the safe design and maintenance of vehicles and transportation equipment would be helpful. The refrigerated transportation step assumed potential growth of *L. monocytogenes*.

4.1.4.2 Retail refrigerated storage

The refrigerated storage conditions during retail refrigerated storage were modeled using data from Ecosure (2008) in the form of distributions. The refrigerated retail storage step assumed growth of *L. monocytogenes* under refrigeration. Control mechanisms at this level are useful to ensure that refrigerated commercial temperatures for frankfurters are maintained throughout retail distribution.

4.1.4.2.1 Retail display

Frankfurters are usually displayed refrigerated. The refrigerated storage conditions during retail display were modeled using data from Ecosure (2008). It was assumed that growth of *L. monocytogenes* within a package could occur, that the packaging of the product remained intact and that there was no recontamination or cross-contamination of the product with *L. monocytogenes*.

4.1.5 Consumer module

The consumer level has been shown to be a key component in the farm-to-fork continuum. Consumers constitute the final step in the food chain and their behavior is critical in minimizing the risk of foodborne disease, including listeriosis (Yang and others 2006). The consumer model includes three sections: (1) transportation of the frankfurters from the retail store to the consumer's refrigerator, (2) home refrigerated storage by the consumer, and (3) the final level of *L. monocytogenes* after preparation (e.g., reheating) of the product prior to consumption. These steps were modeled in the consumer module worksheet depicted in Figure 4.11.

4.1.5.1 Transportation by consumer

Growth of *L. monocytogenes* due to potential temperature and time abuse is possible during the transportation of frankfurters by the consumer. A distribution was created within the baseline model to represent this step of the product pathogen pathway. During transportation by the consumer, growth of *L. monocytogenes* was assumed. Potential temperature abuse was considered using Ecosure (2008) temperatures. The variability of the system was accounted for by including all pertinent distributions.

4.1.5.2 Home storage by consumer

Growth due to potential temperature and time abuse was assumed during the refrigerated storage of frankfurters at the consumer level. A distribution was created within the baseline model including the mean product temperature (38.2°F or 4.33°C) for home refrigerators published by Ecosure (2008).

4.1.5.3 Final level of L. monocytogenes

After refrigerated storage at the consumer level, it is assumed that frankfurters non-reheated (FNR) are immediately served prior to consumption without any thermal treatment. No growth of *L. monocytogenes* was assumed at this level. This step was modeled as the

output within the consumer module worksheet (FNR) and represents the final concentration of *L. monocytogenes* immediately prior to consumption for the baseline model.

4.2 *Hazard Characterization*

Human listeriosis is recognized as a disease primarily caused by ingestion of *L. monocytogenes* in food (ICMSF 2002). Although hazard characterization could include a broader scope considering sequelae and severity assessment (Buchanan and Lindqvist 2000), it is often described as dose-response assessment within the food safety community. In this study, the hazard characterization was addressed as the dose-response module.

4.2.1 Dose-response module

The dose-response module constitutes the final module in the quantitative risk model for frankfurters. In 2004, the FAO/WHO established a respected dose-response study for *L. monocytogenes*. This model was used for the present QMRA baseline model. The dose-response module was divided in two sections: (1) dose or serving size and (2) probability of illness. These steps were modeled within the dose-response module worksheet for non-reheated frankfurters (baseline model) depicted in Figure 4.12.

4.2.1.1 Dose

The dose ingested, or serving size, of frankfurters was described by the empirical distribution RiskCumul(57,285,{114,171,285},{0.75,0.95,0.99}) in grams of food eaten per serving. This distribution shows the 50th (median), 75th, 95th and 99th percentiles of the weighted distributions of serving size. These percentiles for frankfurters are 57, 114, 171, and 285 grams per serving, respectively. This distribution indicates that half of the servings were less than 57 grams and 95% of the servings were less than 171 grams. This distribution was based on data used by the 2003 FDA/USDA Risk Assessment (see Table 4.5). These values were also used by other *Listeria* risk assessments (FDA/USDA 2001; FAO/WHO 2004)

which used equivalent distributions. It should be noted that the original values used to generate the distribution of serving sizes of frankfurters in the simulation model were obtained from the Continuing Survey of Food Intakes by Individuals undertaken in the U.S. (CSFII) and the National Health and Nutrition Examination Survey (NHANES). It is important to recognize the limitations of existing data throughout the exposure assessment. For example, the serving size was represented with the same distribution for frankfurters reheated and non-reheated. This serving size distribution was included within the dose-response module worksheet.

4.2.1.1.1 Probability of illness

The probability of illness (listeriosis) was mathematically represented by the exponential dose-response model from FAO/WHO (2004) described in Chapter 3. The predictions generated by this dose-response model for the susceptible population, part of the FAO/WHO risk assessment of *L. monocytogenes* in ready-to-eat foods, were compared by Hoelzer and others (2013) with other selected published models for *L. monocytogenes* developed using different data sets. The FAO/WHO (2004) model showed acceptable performance. This exponential model was based on the dose-response relationship between exposure estimates and infection rates (Buchanan and others 1997b). The probability of illness was calculated using the exponential dose-response model including the R distribution for susceptible populations provided by FAO/WHO (2004). The dose-response model was combined with the serving size distribution, and the modeled contamination level data, to predict probability of illness. The distributions used for the dose-response module worksheet are included in Table 4.4 at the end of this chapter.

4.3 *Risk Characterization*

The quantitative risk characterization summarizes all previous steps within a probabilistic risk assessment. In other words, it represents the integration and interpretation of

the previous steps by combining the dose-response model with the exposure assessment model. The hazard characterization of the frankfurters model aimed to describe the relationship between the level of *L. monocytogenes* resulting from the exposure assessment, and the probability of subsequent development of listeriosis and adverse health outcomes on susceptible populations. Chapter 4 estimates risk predicted by the frankfurter model using cases of listeriosis per million servings. While risk estimates of listeriosis caused by consumption of frankfurters could be calculated for different populations, it was deemed unnecessary since the susceptible population represents the vast majority of listeriosis cases. Furthermore, the primary goal of the risk assessment was to identify through sensitivity analyses the critical control points in the frankfurter process. Risk estimates for the susceptible population using the FAO/WHO (2004) exponential model were considered sufficient.

4.3.1 Risk Assessment Model Estimate

The baseline model for frankfurters non-reheated (FNR) predicts for the susceptible population, as described the FAO/WHO (2004) R-values, a median probability of listeriosis of 4.17×10^{-9} which represents 0.0041 cases of listeriosis per million servings. FDA/USDA (2003) estimated a total median number of cases of listeriosis per serving for FNR of 6.5×10^{-8}. ICMSF (2002) estimated for frankfurters that the probability of the high-risk population acquiring listeriosis from frankfurters containing high levels of *L. monocytogenes* would be 8.3 cases of listeriosis per serving. It is noteworthy that because of the combination and pooling of data from many diverse sources, the risk estimates may not accurately represent the situation for every frankfurter processing facility. Furthermore, since numerous assumptions have been made and reliable data are lacking, it is impossible to validate this estimate or calculate attendant uncertainties (ICMSF 2002). A scenario for reheated

frankfurters (FR) at the consumer level was developed and its results are discussed in Chapter 6.

Table 4.1 Distribution to estimate the initial level of L. monocytogenes *in the raw meat pre-blend destined for frankfurter production*

Level in the Process	Symbol	Unit	Distribution	Reference
Initial Level or Concentration of *L. monocytogenes* in raw meat pre-blend	IC	(CFU/g)	RiskGamma(0.015386,67.306,RiskShift(0.09861))	Best Fit distribution developed from USDA Baseline Survey Results for Ground Beef, Ground Turkey, and Ground Chicken (Nationwide Federal Plant Raw Ground Beef Microbiological Survey, August 1993 – March 1994) Source: USDA 1996; USDA 1996 cited by ICMSF 2002.

Table 4.2 Chemical properties and efficacy of commercial liquid smokes as an antimicrobial against Listeria monocytogenes *in frankfurters*

Liquid smoke (LS) tested	**Manufacturer**	**Liquid smoke concentration**	**Titrable acidity acetic acid % (wt/wt)**	**pH**	**Phenol content (mg/ml)**	**Carbonyl content (g/100 ml)**	**Processing parameters**	**Strain**	**Result**	**References**
CharSol10	Red Arrow Company	100%	10.5-12	2.1 - 2.6	10-15	12-13	Frankfurters were inoculated, dipped in LS, vacuum packed and stored at 4°C for 72h	*L. monocytogenes* LCDC 4b	1 log initially and undetectable after 72 h	Messina and others 1988
Zesti Smoke	Mastertaste, Inc.	Formulated into product at 10,5, 2.5% (wt/wt)	Not listed	Not listed	Not listed	Not listed	Frankfurters were inoculated, vacuum packed and stored at 4°C for 12 weeks	*L. monocytogenes*	0.5-2 log CFU/ml reductions	Morey and others 2012

Table 4.3 Values used to generate the distribution of serving size of frankfurters used in the quantitative risk assessment model for L. monocytogenes

Serving Size (g)	Cumulative Probability
0	0.00
57	0.50
114	0.75
171	0.95
285	0.99

Source: FDA/USDA 2003

Table 4.4 Distributions Used in the Frankfurters Quantitative Microbial Risk Assessment (QMRA) Model

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Ingredients Module	C9	Best Fit distribution developed from USDA Baseline Survey Results (ICMSF 2002)	=RiskGamma(0.015386,67.306,RiskShift(0.019861))
Ingredients Module	B6	Duration of transportation of raw meat pre-blend from the meat supplier to the processing facility in hours (ttr1)	=RiskTriang(6.5,7,10)
Ingredients Module	C6	Initial level of *Listeria monocytogenes* (Lm) in raw meat pre-blend in Log CFU/g (IC)	=LOG(C9,10)
Ingredients Module	D6	Growth of Lm in raw meat pre-blend during transportation from the meat supplier to the processing facility in Log cfu/g (Gtr1)	='RSM Aerobic Processing'!$K$19*(B6-0)
Ingredients Module	E6	Output (Log cfu/g) Log IC+ Log Gtr1= LogC1	=RiskOutput()+Log_IC+'RSM Aerobic Processing'!$K$19*(B6-0)
Raw Prod. Proc Module	A6	Time for the refrigerated storage of raw meat pre-blend after reception in the processing facility in (ts1)	=RiskTriang(8,48,144)
Raw Prod. Proc Module	E22	Intercept (b) (Murphy and others 2002)	8.742786
Raw Prod. Proc Module	E9	Temperature of cooking at the processing facility level in °C (Tck1)	=RiskTriang(71.1,73.3,75.5)
Raw Prod. Proc Module	E20	Slope (m) (Murphy and others 2002)	-0.131438
Raw Prod. Proc Module	FG20	Log D (Murphy and others 2002)	=($E$20*E9)+$E$22

Table 4.4 Distributions Used in the Frankfurters Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section Worksheet (Tab)	Location (Cell)	Description or Step in the Process	Distribution Formula
Raw Prod. Proc Module	F22	Decimal reduction time in minutes (D value)	=10^F20
Raw Prod. Proc Module	E6	Time of cooking at the processing facility level in minutes (tck1)	=RiskTriang(15,15.2,20)
Raw Prod. Proc Module	F6	Reduction of Lm in frankfurters during cooking at the processing facility level in Log CFU/g (Log Rck1)	=($E$6/$F$22)
Raw Prod. Proc Module	C6	Level of Lm in raw meat pre-blend after refrigerated storage (S1) in the processing facility in Log CFU/g (Log C2)	='Ingredients Module'!$E$6+B6
Raw Prod. Proc Module	G6	Level of Lm in cooked frankfurters immediately after cooking (ck1) in oven (including casing) in Log CFU/g (Log C3)	=RiskOutput("Log C3")+$D$6-$F$6 Output (Truncated Distribution)
Cooked Prod Proc Module	A9	Reference distribution after cooking (C3)	='Cooked Franks Reference Distrib'!$E$28
Cooked Prod Proc Module	C9	Concentration of Lm in cooked frankfurters after peeling of the artificial casing using a heated blade (C5)	=RiskMakeInput((A9+10^RiskNormal(-2.43,0.69)*RiskLognorm(0.0005,0.1)),RiskName("Peeling"))
Cooked Prod Proc Module	D9	Time during collate before packaging in hours (tcol)	=RiskPert(2,6,12)
Cooked Prod Proc Module	E9	Growth of Lm in cooked and peeled frankfurters during collate before packaging (Gcol)	='RSM Aerobic Processing'!$K$20*(D9-0)
Cooked Prod Proc Module	L9	Time for the refrigerated final product storage of packaged frankfurters in the processing facility in hours (ts2)	=RiskTriang(2,8,24,RiskName("Duration of refrigerated storage at facility level"))

Table 4.4 Distributions Used in the Frankfurters Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section Worksheet (Tab)	Location (Cell)	Description or Step in the Process	Distribution Formula
Cooked Prod Proc Module	I9	Concentration of Lm in cooked frankfurters after collate/transportation throughout the equipment (i.e., peeler table, chute, drake [elevated conveyor, in-feed conveyor, collator chain, inspection conveyor]) (C6)	=RiskMakeInput((C9*F9+10^RiskNormal(-0.28,0.2)*RiskLognorm(0.0005,0.1)),RiskName("Collating"))
Cooked Prod Proc Module	M9	Growth of Lm in packaged frankfurters during refrigerated final product storage in the processing facility (Gs2)	RiskGoalSeekChangingCell (0.00228921876932926)
Cooked Prod Proc Module	K9	Level of Lm in cooked frankfurters during packaging which includes contact with gloves during visual inspection (C7)	=RiskMakeInput(I9+10^RiskNormal(-4.96,0.37)*RiskLognorm(0.0005,0.1),RiskName("Handling at Packaging"))
Cooked Prod Proc Module	O9	Concentration of Lm after the final product storage (C8)	=RiskOutput("C8")+K9*N9 ='Cooked Prod Proc Module'!$O$9
Distribution and Marketing Module	B9	Duration of transportation of packaged frankfurters from the processing facility to retail in hours (ttr2)	=RiskTriang(8,24,72,RiskName("Duration of Transportation to Retail"))
Distribution and Marketing Module	F9	Duration of the refrigerated storage of packaged frankfurters at retail (back and front) in hours (ts3)	=RiskTriang(24,48,96, RiskName("Duration of Refrigerated Storage during Retail"))
Distribution and Marketing Module	C9	Growth of Lm in packaged frankfurters during transportation from the processing facility to retail (Gtr2)	='RSM Anaerobic Retail Distrib.'!$K$20*(B9-0) =IF(C9=0,1,C9)

Table 4.4 Distributions Used in the Frankfurters Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section Worksheet (Tab)	Location (Cell)	Description or Step in the Process	Distribution Formula
Distribution and Marketing Module	G9	Growth of Lm in packaged frankfurters during refrigerated storage at retail (back and front) (Gs3)	='RSM Anaerobic Retail Distrib.'!$K$20*(F9-0)
Distribution and Marketing Module	E9	Concentration of Lm in frankfurter packages after refrigerated transportation (tr2) from the processing facility to retail (C9)	='Cooked Prod Proc Module'!O9*D9
Distribution and Marketing Module	I9	Concentration of Lm in frankfurter packages after refrigerated retail storage (C10)	=RiskOutput("C10")+E9*H9
Consumer Module Frankfurter non-reheated (FNR)	A9	Duration of transportation of packaged frankfurters from retail to consumer's refrigerator in hours (ttr3)	=RiskTriang(0.25,1.167,2.333, RiskName("Duration of Transportation from Retail to Consumer"))
Consumer Module (FNR)	E9	Duration of refrigerated storage of frankfurters at consumer level in hours (ts4)	=RiskTriang(0.1*24,2*24,5*24,RiskName("Duration of Refrigerated Consumer Storage"))
Consumer Module (FNR)	C9	Growth of Lm in packaged frankfurters during transportation from retail to consumer's refrigerator (Gtr3)	='RSM Anaerobic Consumer Storage'!$K$20*(A9-0)
Consumer Module (FNR)	F9	Growth of Lm in frankfurter during consumer's refrigerated storage (Gs4)	='RSM Anaerobic Consumer Storage'!$K$20*(E9-0)
Consumer Module (FNR)	D9	Level of Lm in frankfurter packages after refrigerated transportation (tr3) from retail to consumer's refrigerator (C11)	='Distribution and Marketing Mod'!I9*C9

Table 4.4 Distributions Used in the Frankfurters Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Consumer Module (FNR)	G9	Final Level of Lm in frankfurter packages after refrigerated (S4) consumer storage (C12)	=RiskOutput("C12")+D9*F9
Dose-Response Module (FNR)	A9	Serving Size (SS)	=RiskCumul(57,285,{114,171,285},{0.75,0.95,0.99},RiskName("Serving Size"))
Dose-Response Module (FNR)	B9	Dose or number of biological agent consumed (CFU/serving) [D(FNR)](output of consumer module*SS)	=RiskOutput()+'Consumer Module (FNR)'!G9*A9
Dose-Response Module (FNR)	J9	Susceptible Population r-value (WHO/FAO 2004)	=0.00000000000106
Dose-Response Module (FNR)	K9	Probability of Illness ($P=1-e^{-R*D}$)	=RiskOutput()+1-EXP(-J9*B9)
Dose-Response Module (FNR)	L9	Cases of Listeriosis per one million servings	=RiskOutput("Cases per Million Servings")+K9*1000000

Figure 4.1 Introductory worksheet for the quantitative risk assessment model for *L. monocytogenes* on frankfurters

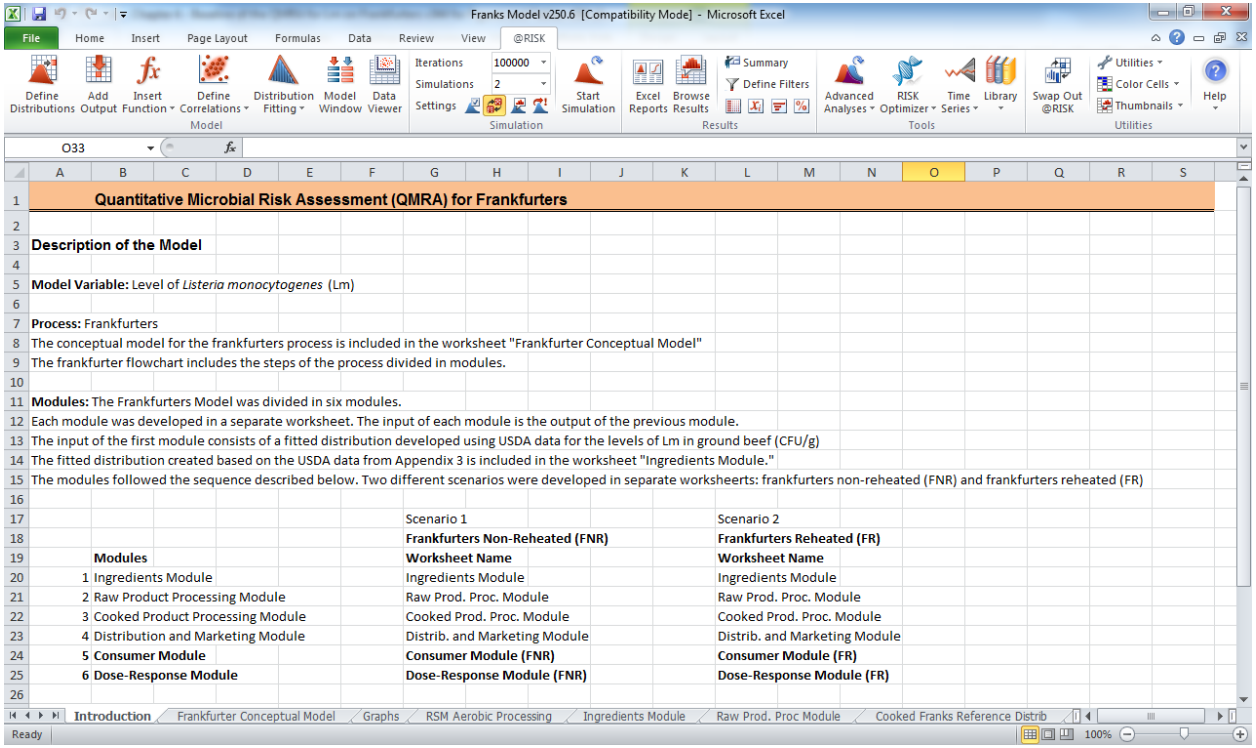


Figure 4.2 Conceptual model for the frankfurters Modular Product Pathogen Pathway (MPPP) for *L. monocytogenes*

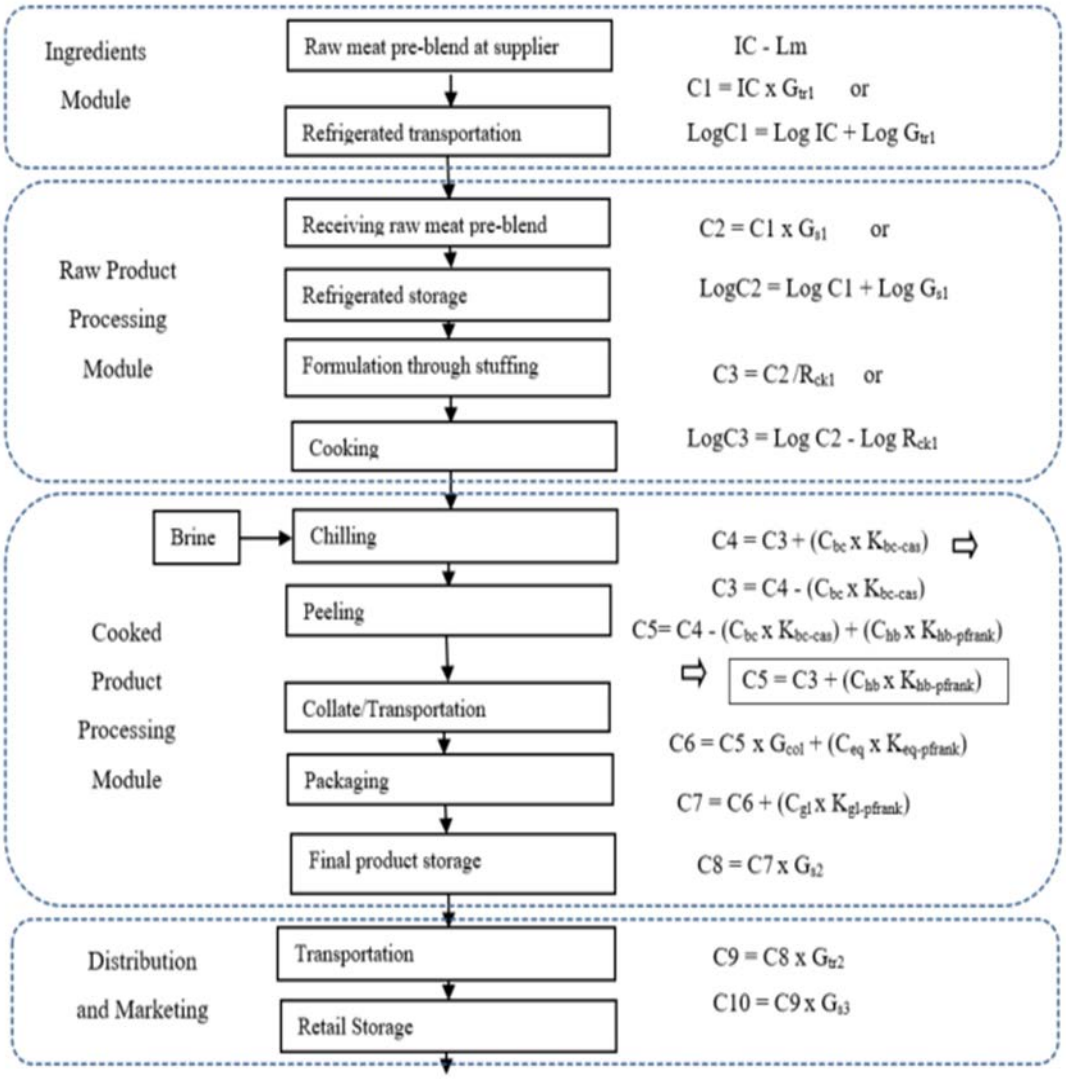


Figure 4.2 Conceptual Model for the Frankfurters Modular Product Pathogen Pathway (MPPP) for *L. monocytogenes* (Continued)

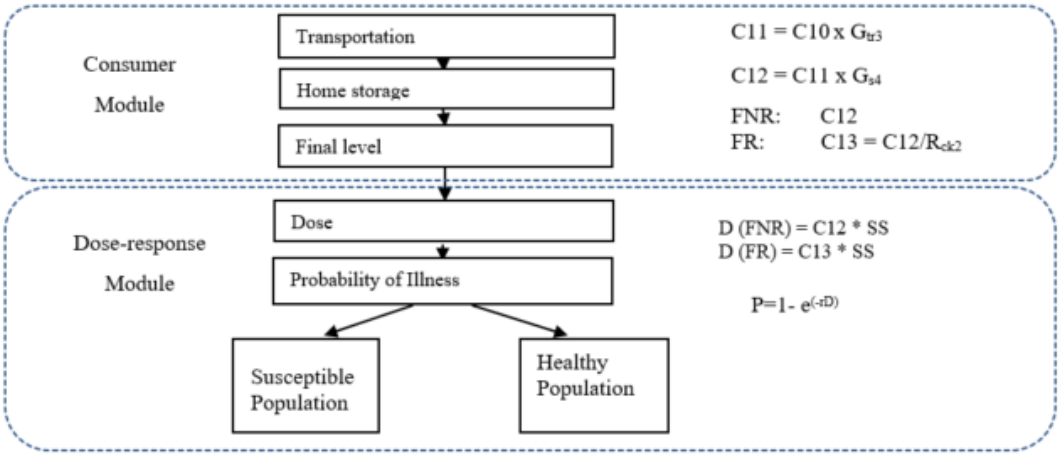


Figure 4.2 Conceptual Model for the Frankfurters Modular Product Pathogen Pathway (MPPP) for *L. monocytogenes* (Continued)

Where:
IC or IC-Lm = Initial level of *Listeria monocytogenes* (Lm) in raw meat pre-blend
C = Level of Lm in raw or cooked meat at subsequent steps of the process (C1, C2,…,C13)
C_{bc} = Level of Lm in the brine chill (bc)
C_{hb} = Level of Lm in heated blade (hb)*
C_{eq} = Level of Lm in equipment (eq)
C_{gl} = Level of Lm on gloves (gl)** as food contact surfaces during packaging
G = Growth of Lm in raw or cooked meat at different steps of the process (G_{tr1}, G_{s1},…,G_{tr3}, G_{s4})
G_{tr1} = Growth of Lm in raw meat pre-blend during transportation (tr_1) from the meat supplier to the processing facility
G_{tr2} = Growth of Lm in packaged frankfurters during transportation (tr_2) from the processing facility to retail
G_{tr3} = Growth of Lm in packaged frankfurters during transportation (tr_3) from retail to consumer's refrigerator
G_{s1} = Growth of Lm in raw meat pre-blend during refrigerated storage (s_1) in the processing facility immediately after reception
G_{s2} = Growth of Lm in packaged frankfurters during refrigerated final product storage (s_2) in the processing facility
G_{s3} = Growth of Lm in packaged frankfurters during refrigerated storage (s_3) at retail (back and front)***
G_{s4} = Growth of Lm in frankfurter during consumer's refrigerated storage (s_4)
G_{col} = Growth of Lm in cooked and peeled frankfurters during collate (col) before packaging
R_{ck1} = Reduction of Lm in frankfurters during cooking (ck_1) at the processing facility level
R_{ck2} = Reduction of Lm in frankfurters during reheating (cooking—ck_2) at the consumer level
K = Transfer rates of Lm at key steps of the process ($K_{bc\text{-}cas}$, $K_{hb\text{-}pfrank}$, …, $K_{gl\text{-}pfrank}$)
$K_{bc\text{-}cas}$ = Transfer rate of Lm from brine chill (bc) to cooked casing (cas) during brining
$K_{hb\text{-}pfrank}$= Transfer rate of Lm from heated blade (hb)* to peeled frankfurter (pfrank) during peeling
$K_{eq\text{-}pfrank}$= Transfer rate of Lm from equipment (eq) in direct contact with peeled frankfurters (pfrank) to pfrank
$K_{gl\text{-}pfrank}$= Transfer rate of Lm from the contact between gloves (gl)** and peeled frankfurters (pfrank)
FR = Frankfurter reheated (consumer module)
FNR = Frankfurter non-reheated (consumer module)
SS = Serving size
P = Probability of listeriosis
e = Base of the natural logarithm
r = Constant specific for Lm that helps define the shape of the dose-response curve
D = Dose or number of Lm consumed (CFU)
*The baseline model uses a heating blade (hb) which produces a superficial longitudinal cut barely noticeable. If the blade is not heated then a potentially contaminated "peeling blade" could inoculate frankfurters with *L. monocytogenes* during the peeling step of the process.
**The gloves used were stainless steel wire mesh gloves
***The back part of the retail store generally has a lower temperature than the front part of it.

Figure 4.3 Frankfurters conceptual model worksheet including modules

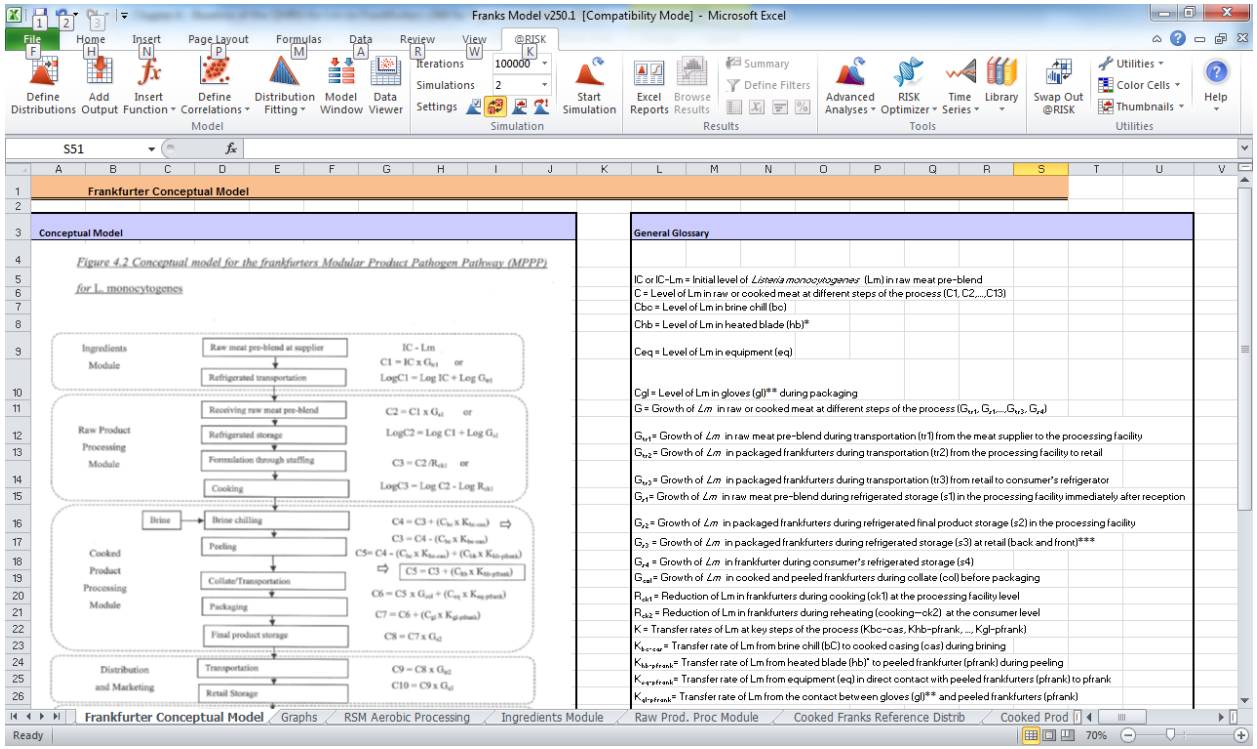


Figure 4.4 Ingredients module worksheet

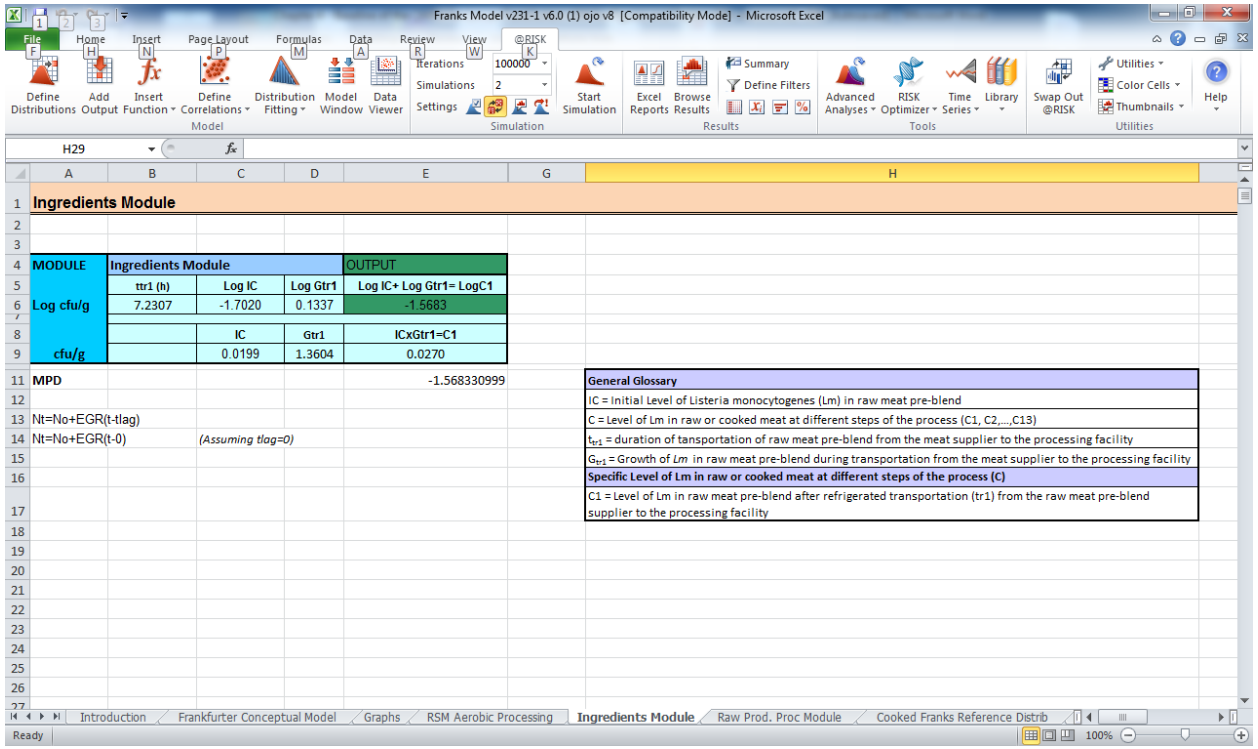


Figure 4.5 Level of *L. monocytogenes* in incoming meat pre-blend based on incidence and reference distributions

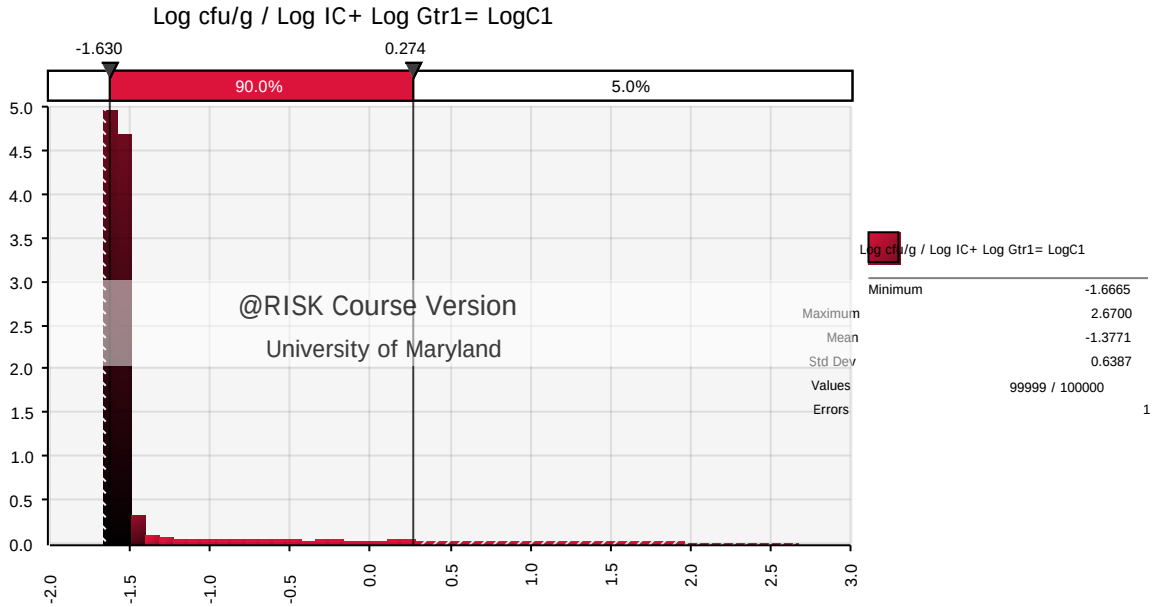
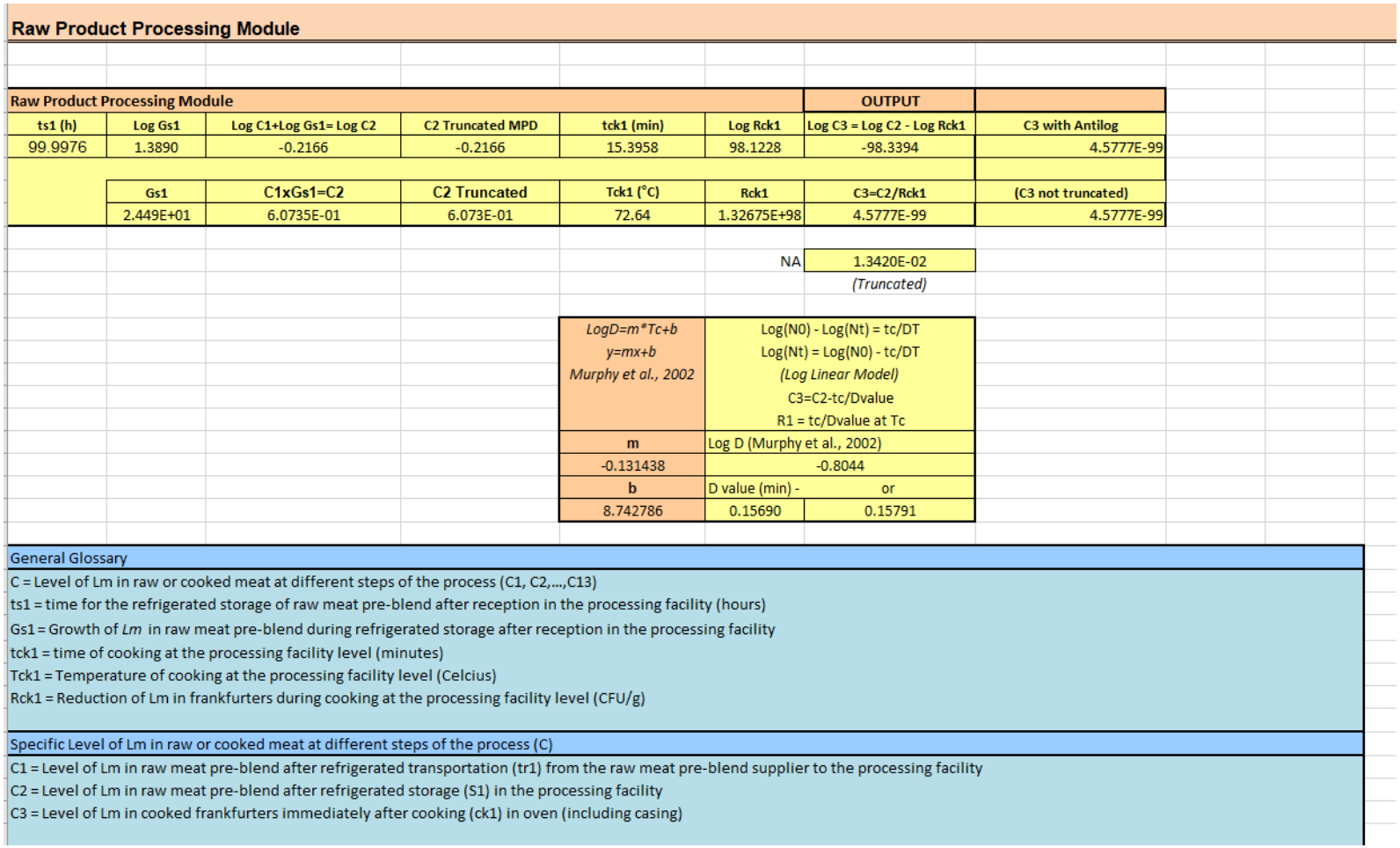


Figure 4.6 Raw product processing module worksheet



Raw Product Processing Module

Raw Product Processing Module						OUTPUT	
ts1 (h)	Log Gs1	Log C1+Log Gs1= Log C2	C2 Truncated MPD	tck1 (min)	Log Rck1	Log C3 = Log C2 - Log Rck1	C3 with Antilog
99.9976	1.3890	-0.2166	-0.2166	15.3958	98.1228	-98.3394	4.5777E-99
	Gs1	C1xGs1=C2	C2 Truncated	Tck1 (°C)	Rck1	C3=C2/Rck1	(C3 not truncated)
	2.449E+01	6.0735E-01	6.073E-01	72.64	1.32675E+98	4.5777E-99	4.5777E-99
					NA	1.3420E-02	
						(Truncated)	

*LogD=m*Tc+b*	Log(N0) - Log(Nt) = tc/DT
y=mx+b	Log(Nt) = Log(N0) - tc/DT
Murphy et al., 2002	*(Log Linear Model)*
	C3=C2-tc/Dvalue
	R1 = tc/Dvalue at Tc
m	Log D (Murphy et al., 2002)
-0.131438	-0.8044
b	D value (min) - or
8.742786	0.15690 0.15791

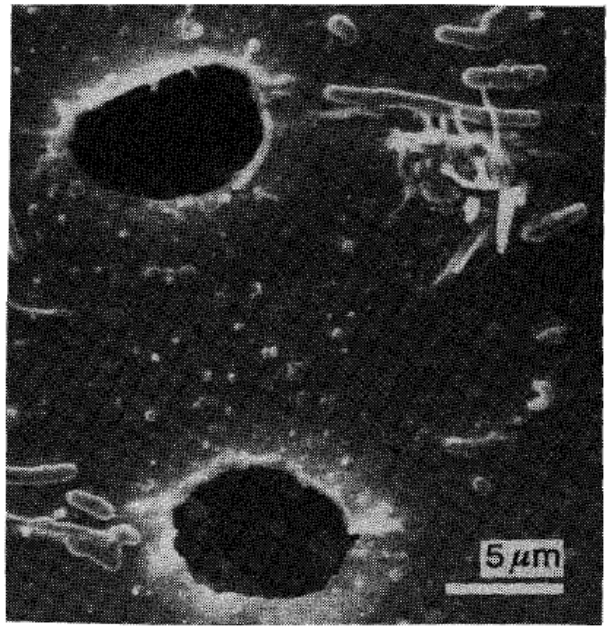
General Glossary

C = Level of Lm in raw or cooked meat at different steps of the process (C1, C2,...,C13)
ts1 = time for the refrigerated storage of raw meat pre-blend after reception in the processing facility (hours)
Gs1 = Growth of *Lm* in raw meat pre-blend during refrigerated storage after reception in the processing facility
tck1 = time of cooking at the processing facility level (minutes)
Tck1 = Temperature of cooking at the processing facility level (Celcius)
Rck1 = Reduction of Lm in frankfurters during cooking at the processing facility level (CFU/g)

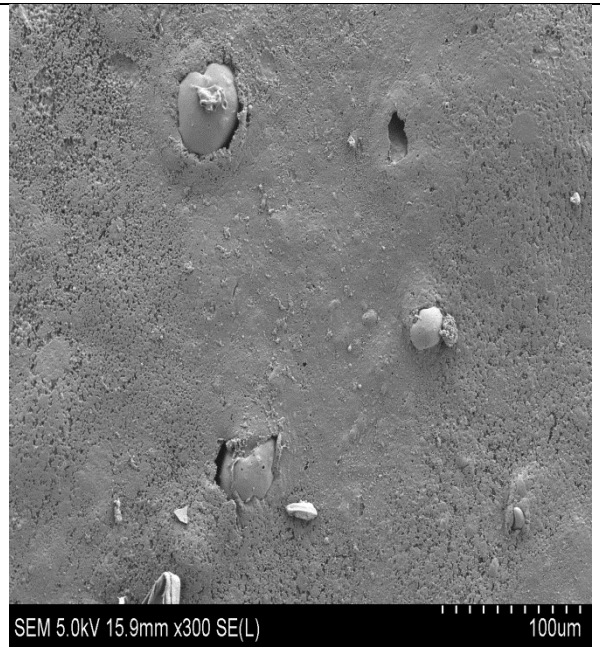
Specific Level of Lm in raw or cooked meat at different steps of the process (C)

C1 = Level of Lm in raw meat pre-blend after refrigerated transportation (tr1) from the raw meat pre-blend supplier to the processing facility
C2 = Level of Lm in raw meat pre-blend after refrigerated storage (S1) in the processing facility
C3 = Level of Lm in cooked frankfurters immediately after cooking (ck1) in oven (including casing)

Figure 4.7 Scanning electron microscopy close-up images of surface regions of frankfurters



(a) Scanning electron micrograph (SEM) of the "surface of a control wiener showing the dense layer of coagulated protein, surface lesions, and indigenous microflora (probably lactobacilli)" (McKellar 1994).



(b) SEM of the surface of a peeled beef frankfurter showing the layer of coagulated protein, fat globules of different sizes embedded within the coagulated skin of the frankfurter emulsion, and a surface lesion (probably produced by the detachment of a fat globule from the frankfurter surface). No indigenous microflora was found in the control samples selected (Williams 2011).

Figure 4.8 Cooked product processing module worksheet

Cooked Product Processing Module

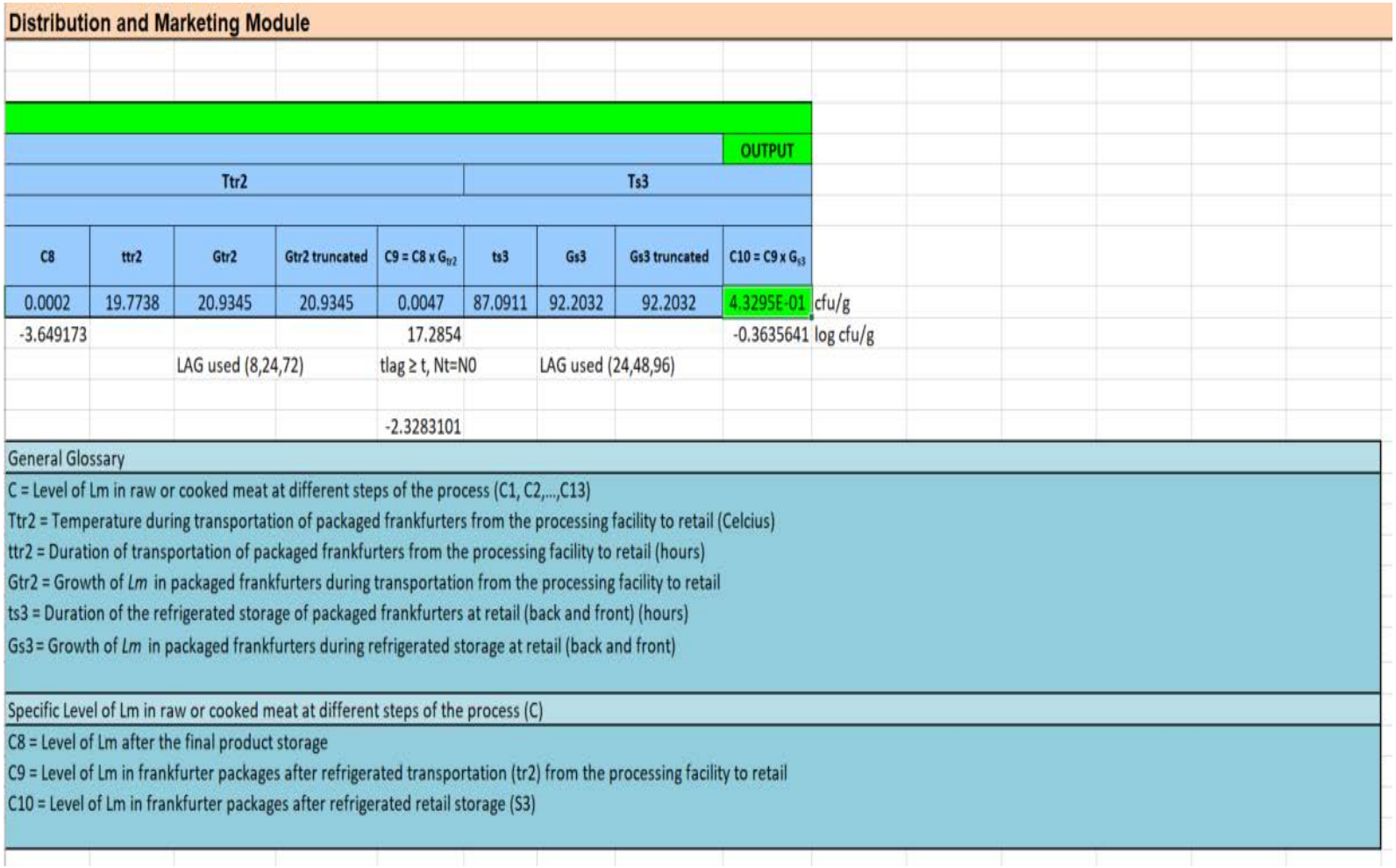
From after Brine Chill and Peeler and using the truncated to zero cfu/g

			Tcol								
Level after Cooking (C3)	(Chb*Khb-frank)	C5=C3 + (Chb*Khb-frank)	tcol (h)	Gcol	Gcol Truncated	(Ceq x Keq-pfrank)	C5 x GcolT	C6 = C5 x GcolT + (Ceq x Keq-pfrank)	(Cgl x Kgl-pfrank)	C7 = C6 + (C_{gl} x $K_{gl\text{-}pfrank}$)	ts2
0.02	6.73151E-08	0.0200	2.7300	2.8837	2.9	8.47937E-08	0.057673336	0.0577	8.7095E-12	0.0577	14.8376
-1.6990	-7.1719	-1.6990				-7.0716	-1.2390	-1.2390	-11.0600	-1.2390	
				LAG used (2,6,12)			tlag ≥ t, Nt=N0				
Distrib. Normal	-2.164565809					-0.11117688			-4.733792867		
Distrib. Normal Antilog	0.002528151					0.855238283			1.31753E-05		

General Glossary

C = Level of Lm in raw or cooked meat at different steps of the process (C1, C2,...,C13) CFU/g
Tcol = Temperature during collate before packaging (°C)
tcol = time during collate before packaging (hours)
Gcol = Growth of Lm in cooked and peeled frankfurters during collate (before packaging)
GcolT=Gcol truncated so that when tlag ≥ t, Nt=N0
Ts2 = Temperature for the refrigerated final product storage of packaged frankfurters in the processing facility (°C)
ts2 = time for the refrigerated final product storage of packaged frankfurters in the processing facility (hours)
Gs2 = Growth of Lm in packaged frankfurters during refrigerated final product storage in the processing facility

Figure 4.9 Distribution and marketing module worksheet



Distribution and Marketing Module

								OUTPUT
Ttr2					Ts3			
C8	ttr2	Gtr2	Gtr2 truncated	C9 = C8 x G_{tr2}	ts3	Gs3	Gs3 truncated	C10 = C9 x G_{s3}
0.0002	19.7738	20.9345	20.9345	0.0047	87.0911	92.2032	92.2032	4.3295E-01 cfu/g
-3.649173				17.2854				-0.3635641 log cfu/g
		LAG used (8,24,72)		tlag ≥ t, Nt=N0		LAG used (24,48,96)		
				-2.3283101				

General Glossary

C = Level of Lm in raw or cooked meat at different steps of the process (C1, C2,...,C13)

Ttr2 = Temperature during transportation of packaged frankfurters from the processing facility to retail (Celcius)

ttr2 = Duration of transportation of packaged frankfurters from the processing facility to retail (hours)

Gtr2 = Growth of *Lm* in packaged frankfurters during transportation from the processing facility to retail

ts3 = Duration of the refrigerated storage of packaged frankfurters at retail (back and front) (hours)

Gs3 = Growth of *Lm* in packaged frankfurters during refrigerated storage at retail (back and front)

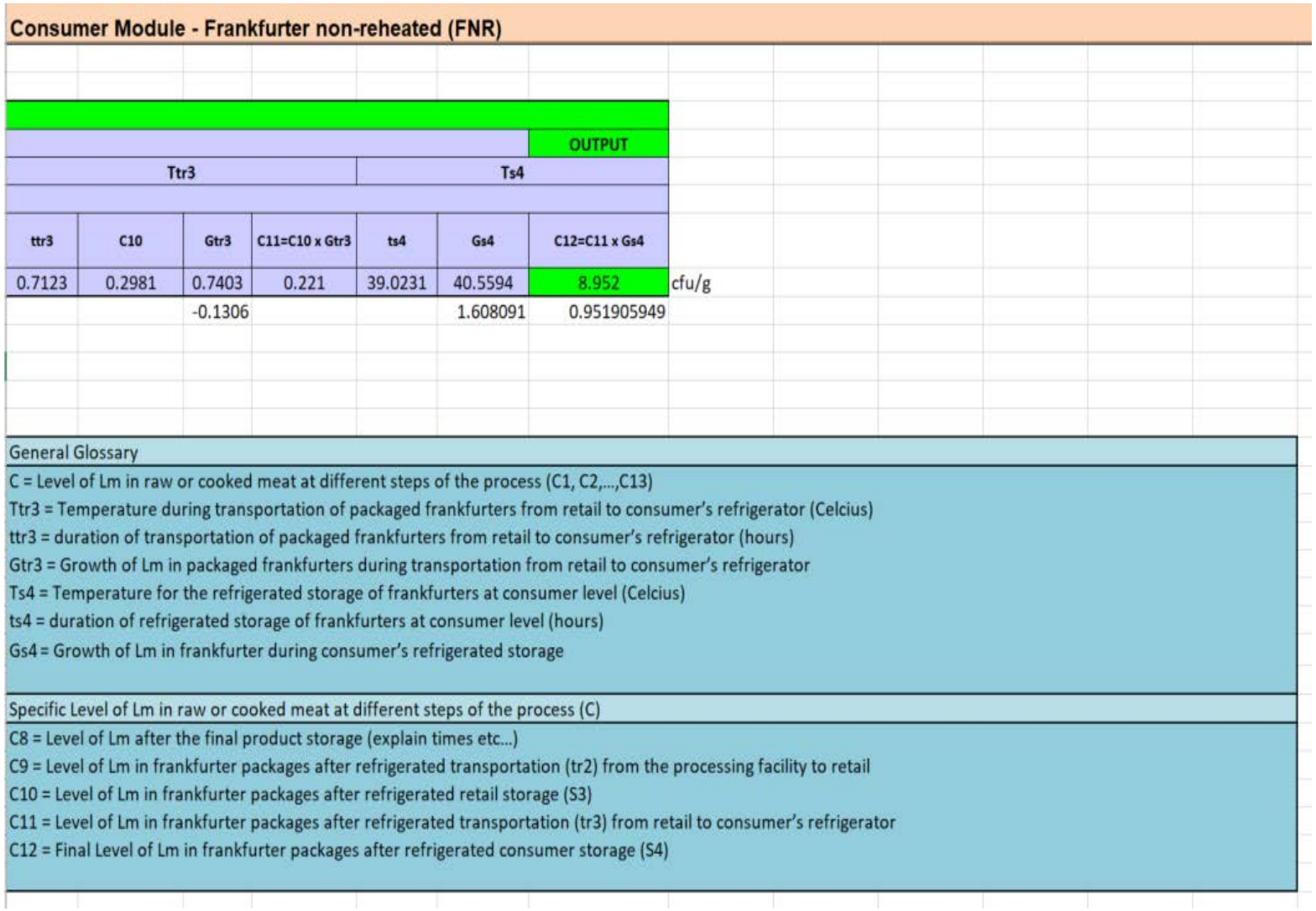
Specific Level of Lm in raw or cooked meat at different steps of the process (C)

C8 = Level of Lm after the final product storage

C9 = Level of Lm in frankfurter packages after refrigerated transportation (tr2) from the processing facility to retail

C10 = Level of Lm in frankfurter packages after refrigerated retail storage (S3)

Figure 4.10 Consumer module worksheet for non-reheated frankfurters



Consumer Module - Frankfurter non-reheated (FNR)

						OUTPUT	
Ttr3				Ts4			
ttr3	C10	Gtr3	C11=C10 x Gtr3	ts4	Gs4	C12=C11 x Gs4	
0.7123	0.2981	0.7403	0.221	39.0231	40.5594	8.952	cfu/g
		-0.1306			1.608091	0.951905949	

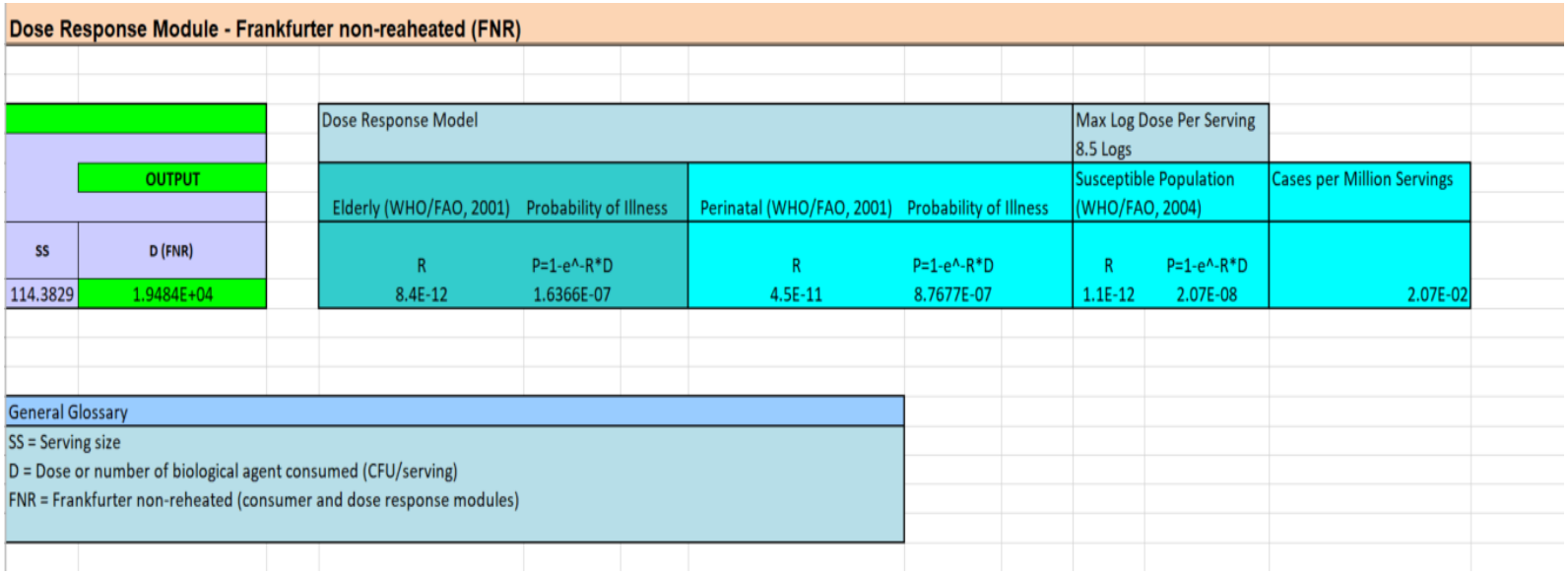
General Glossary

C = Level of Lm in raw or cooked meat at different steps of the process (C1, C2,...,C13)
Ttr3 = Temperature during transportation of packaged frankfurters from retail to consumer's refrigerator (Celcius)
ttr3 = duration of transportation of packaged frankfurters from retail to consumer's refrigerator (hours)
Gtr3 = Growth of Lm in packaged frankfurters during transportation from retail to consumer's refrigerator
Ts4 = Temperature for the refrigerated storage of frankfurters at consumer level (Celcius)
ts4 = duration of refrigerated storage of frankfurters at consumer level (hours)
Gs4 = Growth of Lm in frankfurter during consumer's refrigerated storage

Specific Level of Lm in raw or cooked meat at different steps of the process (C)

C8 = Level of Lm after the final product storage (explain times etc...)
C9 = Level of Lm in frankfurter packages after refrigerated transportation (tr2) from the processing facility to retail
C10 = Level of Lm in frankfurter packages after refrigerated retail storage (S3)
C11 = Level of Lm in frankfurter packages after refrigerated transportation (tr3) from retail to consumer's refrigerator
C12 = Final Level of Lm in frankfurter packages after refrigerated consumer storage (S4)

Figure 4.11 Dose-response module worksheet for non-reheated frankfurters



Dose Response Module - Frankfurter non-reaheated (FNR)

		Dose Response Model				Max Log Dose Per Serving 8.5 Logs		
	OUTPUT	Elderly (WHO/FAO, 2001)	Probability of Illness	Perinatal (WHO/FAO, 2001)	Probability of Illness	Susceptible Population (WHO/FAO, 2004)		Cases per Million Servings
SS	D (FNR)	R	P=1-e^-R*D	R	P=1-e^-R*D	R	P=1-e^-R*D	
114.3829	1.9484E+04	8.4E-12	1.6366E-07	4.5E-11	8.7677E-07	1.1E-12	2.07E-08	2.07E-02

General Glossary

SS = Serving size

D = Dose or number of biological agent consumed (CFU/serving)

FNR = Frankfurter non-reheated (consumer and dose response modules)

Chapter 5: Baseline Quantitative Microbial Risk Assessment (QMRA) for *Listeria monocytogenes* on Cold-Smoked Salmon

A baseline model for cold-smoked salmon was developed for the purpose of incorporating quantitative risk assessments into the HACCP equation. This chapter describes the baseline model for *Listeria monocytogenes* in cold-smoked salmon including a detailed explanation of each step of the process, following the modular product pathogen pathway (MPPP) described in Chapter 3. This chapter describes the details of the cold-smoked salmon and *L. monocytogenes* risk modeling, including sections on estimating the initial level of contamination, modeling *L. monocytogenes* growth in cold-smoked salmon, and the impact of the steps in the process contributing to the partial control of *L. monocytogenes* in cold-smoked salmon. The description of the cold-smoked salmon process is based mainly on reviewing manufacturing descriptions and augmented by a visit to the smoked salmon facility as described in Chapter 3. The process description was supported by information generously provided by Dr. Barbara Blakistone, an expert in the seafood industry. It was also supported by reference documents such as the Association of Food and Drug Officials' Guidance for Processing Smoked Seafood in Retail Operations (AFDO 2004a), which has been adopted by reference (Wiedmann and Gall 2008) in the Association of Food and Drug Officials' Cured, Salted and Smoked Fish Good Manufacturing Establishments GMPs (AFDO 2004b). In addition, the document "Processing Parameters Needed to Control Pathogens in Cold-Smoked Fish," a report of the Institute of Food Technologists for the Food and Drug Administration of the U.S. Department of Health and Human Services (IFT 2001) in response to Task Order 2 (IFT/FDA 2001), as well as a summary update of the IFT (2001) report from the International Smoked Seafood Conference Proceedings (Jahncke 2008), were key references. Lastly, the Fish and Fishery Products Hazards and Control Guidance (FDA 2011)

was consulted. When applicable, the baseline model, used simplified steps and unit operations during the various modules of the processes. The scope of risk modeling and HACCP application considered extended from primary production through consumption, allowing determination of "risk-based" critical control points (CCPs) and critical limits. The results from sensitivity analysis and what-if scenarios developed from this baseline model, including consideration of some intervention strategies to reduce the risk of *L. monocytogenes* in cold-smoked salmon, are covered in Chapter 6.

5.1 *Exposure Assessment for the Baseline QMRA Modular Product Pathogen Pathway (MPPP) for Cold-Smoked Salmon*

The cold-smoked salmon (CSS) process was divided into modules that provided manageable and flexible subsystems for developing a quantitative microbial risk assessment (QMRA) model for the overall system. This QMRA begins with an introductory worksheet (Figure 5.1) that summarizes key information needed to follow the sequence of the model. The modules followed the pathway of *L. monocytogenes* for the cold-smoking process, using tools such as predictive microbiology and the risk assessment MPPP model framework described in Chapter 3. The CSS process was divided into eight modules: Primary Production, Raw Product Processing, Brining, Cold-Smoked Product Processing, Post-cold Smoking, Distribution and Marketing, Consumer, and Dose-response. Each module is described below and illustrated for the baseline model in Figure 5.2. In addition, the detailed cold-smoked salmon flowchart worksheet, including calculations, is depicted in Figure 5.3. The distributions used for each module throughout the worksheets were consolidated in a table format and included in Table 5.4.

5.1.1 Primary Production Module

The primary production module represents the earliest stages in the production of CSS. In developing this module, it was assumed that the bulk of raw salmon used in the production of CSS is acquired from distant suppliers, and as such, is frozen at point of harvest and shipped to the manufacturing site in a frozen state. Thus, the module consists of two steps: frozen raw salmon at the supplier, and frozen transportation from supplier to the processing facility. The primary production module worksheet is portrayed in Figure 5.4.

5.1.1.1 Frozen raw salmon at supplier

The "frozen raw salmon at supplier" refers to the baseline model used to describe the state of the raw product before being shipped from a primary production facility (fisheries) to the processing facility. This describes the level of *L. monocytogenes* in the raw fish which is dependent upon the storage temperature, storage duration, good aquaculture practices (if applicable), sanitary conditions, and the temperature at which the fish was maintained immediately after it was caught or harvested source until it is transformed into its frozen state. It is assumed that the salmon is eviscerated by the supplier prior to freezing. The method of freezing and keeping the raw product frozen, maintaining the cold chain throughout this module until the product is received by the processing facility, is important for achieving a safety final product. The presence of *Listeria* spp. in the frozen fish at the suppliers' facilities reflects the adequacy of the suppliers' hygienic and temperature controls.

5.1.1.1.1 Estimating initial levels of *L. monocytogenes* contamination in raw salmon

The initial level of *L. monocytogenes* contamination in raw salmon was estimated from pertinent incidence studies found in the literature and compiled in Appendix 2.3.1 (Table A2.3). These studies revealed a relatively high but variable incidence of *L. monocytogenes.* Appendix 2.3.1 summarizes the incidence rates for raw salmon used as an input for the distribution generated for the cold-smoked salmon model. Weights associated

with the year of publication of the data found in the literature and geographical locations were applied. The incidence of *L. monocytogenes* in raw salmon varied from 0 to 100% with a weighted mean of 20.34% and a weighted standard deviation of 20.83. The calculations to obtain these values are explained in Appendix 2.1 (Table A2.1). The initial reference distribution created for the incidence of *L. monocytogenes* in raw salmon and other distributions pertinent to this model are included in Table 5.4. Figure 5.5 shows the raw salmon reference distribution worksheet for the concentration of *L. monocytogenes* at the primary production level.

Although *L. monocytogenes* does not occur naturally in oceans, aquatic environments may become contaminated from human or animal sewage or runoffs (FAO 2004). It is noteworthy that raw salmon is often contaminated with *L. monocytogenes* to a greater extent than cold-smoked salmon (Guyer and Jemmi 1990; Chitlapilly-Dass 2011). In addition, there are indications that one source of *L. monocytogenes* contamination of the final processed products is incoming frozen or fresh raw salmon (Eklund and others 1995; Fonnesbech Vogel and others 2001; Chitlapilly-Dass and others 2010b; Chitlapilly-Dass 2011). It is also key to consider the important role that in-house *L. monocytogenes* contamination plays during processing as an important source of contamination (Autio and others 1999; Fonnesbech Vogel and others 2001; Medrala and others 2003). One of the greatest challenges in cold-smoked salmon processing facilities is the control and prevention of *L. monocytogenes* harborage in specific niches in the processing environment (Lappi and others 2004; Chitlapilly-Dass 2011).

In general, the available incidence studies of *L. monocytogenes* contamination in raw salmon are not quantitative with regard to the levels of *L. monocytogenes*. To describe the concentration of this pathogen in raw salmon, a binomial distribution was assumed to direct the sampling. The incidence of *L. monocytogenes* in raw salmon reported in the literature

(Appendix 2.3.1) was used to deduce the concentration that would most likely lead to the rate of detecting the reported number of positive samples of *L. monocytogenes* in raw salmon. To account for the weights and the variability in the incidence data, the weighted mean and weighted standard deviation of the *L. monocytogenes* incidence in raw salmon found in the literature were embedded in a lognormal distribution within the binomial distribution. Then, an alternative lognormal distribution was assigned to the positive samples for the purpose of obtaining a defined tailed distribution. The alternative lognormal distribution allows defining a normal distribution with three percentiles. In this case, the 5^{th}, 50^{th}, and 95^{th} percentiles were used and the values 0.04, 0.1 and 0.4 CFU/g were assigned based on the lower limit of detection for *L. monocytogenes* being 0.04 CFU/g. The negative samples were given a value half the value of the lower limit of detection, 0.02 CFU/g, a conservative approach to assigning a concentration value to negative detection data. This approach was selected after careful consideration of the process and the desirability of establishing quantitative means for the modeling. Figure 5.6 shows the concentration of *L. monocytogenes* in incoming raw salmon based on incidence and reference distributions.

5.1.1.1.2 Estimating initial levels of *L. monocytogenes* contamination in other ingredients

Cold smoking is a mild process in which only a few ingredients are added to the fish. In the risk assessment, the focus was on the processing of Atlantic salmon (*Salmo salar)*, the primary species used in commercial cold-smoked salmon. Although the ingredients for this process may vary, they generally consist of salt or brine, smoke, and spices that vary according to the particular region where the cold-smoked salmon is produced. Depending on the cold-smoking procedures, salt granules may be applied directly to the salmon fillets. In this case, contamination of the salt granules with *L. monocytogenes* is unlikely. However, salting can also be done by brine injection or bath brining, which could potentially contaminate the product as explained in the brining step of the process (Section 5.1.3). In

addition to salt, brines may contain other ingredients, depending on the formulation. Brine usually consists of water, salt, and various spices and flavorings. In addition, it may contain sugar, phosphates, citric acid, and, depending on the recipe and species of fish, sodium nitrite. In the case of spices, some have antimicrobial effects on plant and human pathogens (Brandi and others 2006). The essential oils of spices often have antimicrobial activity. Application of essential oils on the surface of whole fish could inhibit *L. monocytogenes* as well as natural spoilage flora (Hayouni and others 2008). However, the antibacterial effect of essential oils may be reduced due to the high fat content of some fish, such as salmon (Tajkarimi and others 2010). The processing facility visited did not use spices for their standard cold-smoked salmon product; subsequently, no variation in the concentration of *L. monocytogenes* was assumed. Smoke is an ingredient added later in the process during the cold-smoking step. Smoke can be produced from different wood varieties or can be added as liquid smoke. Liquid smoke formulations and concentrations may vary depending on the supplier. Montazeri and others (2013) tested three commercially refined fractions and a full strength liquid smoke against *Listeria innocua* ATCC 33090 (surrogate to *L. monocytogenes*) in cold-smoked salmon, observing a 2 log reduction in the concentration of *L. innocua* after 14 days of application. The effects of liquid smoked will be covered in more detail in section 5.1.4.1.

Sodium nitrite ($NaNO_2$) is sometimes added to seafood products to preserve them. Its use is permitted in some types of smoked fish, such as smoked salmon (Nyachuba and others 2007). The antimicrobial activity of $NaNO_2$ toward foodborne pathogens, including *L. monocytogenes*, is enhanced in conjunction with NaCl, pH, and temperature (Buchanan and others 1989; McClure and others 1991). $NaNO_2$ can induce injury in *L. monocytogenes*; however, this pathogen can repair and grow to high levels over extended refrigerated storage. Therefore, special attention is required as $NaNO_2$ induced injury may mask detection of *L. monocytogenes* in RTE seafood products containing nitrite (Nyachuba and others 2007).

5.1.1.2 Frozen transportation

This step refers to the shipment of the frozen fish from the primary production supplier to the processing facility. The frozen transportation could be either domestic production or imported salmon. The baseline model assumed frozen conditions were monitored and maintained to keep the product frozen during its transport to the processing facility.

5.1.2 Raw Product Processing Module

This module consists of the following five steps in the process: receipt of frozen raw salmon, frozen storage, thawing, rinse 1, and filleting or splitting. Each of these steps is included in the raw product processing module worksheet in Figure 5.7.

5.1.2.1 Receiving frozen raw salmon

The reception of frozen raw salmon at the processing facility refers to the incoming frozen raw product from the supplier. At this step, it is helpful to know the source (e.g., harvest waters, certified supplier) of the product. Verifying that the internal temperature is lower than 4.4°C is particularly important when receiving refrigerated raw fish, which should be kept in an appropriate refrigerated or iced condition. If the product is received frozen, facilities can refer to a number of time and temperature combinations recommended by the U.S. government, as explained in Chapter 2. Monitoring temperature charts from the supplier, including records of temperatures from the harvest source up to the reception for each lot of product, may be recommended. Lots should be accompanied by documentation certifying proper time and temperature handling of the salmon.

5.1.2.2 Frozen storage

The duration of the storage was assumed to be less than 14 days (<0.5 cfu/g growth of *L. monocytogenes*). Although freezing (at -18 °C, -0.4 °F) causes a one log reduction of *L. monocytogenes* in buffer (El-Kest and others 1991), the lipids present in fatty fish protect

bacteria against freezing damage (IFT 2001). Salmon is a fatty fish, and even after cold-smoked processing it preserves its fatty composition as shown by scanning electron microscopy (Figure 5.17). Thus, it was assumed that there is no meaningful growth or decline in *L. monocytogenes* levels during frozen storage.

5.1.2.3 Thawing

Frozen products are thawed under refrigeration at or below 41°F (5°C). If the frozen product is thawed in water to accelerate the process, it needs to be placed in clean flowing water, with the water temperature below 70°F (21°C) until thawing is complete (AFDO 2004a). In the case of the visited facility, the temperature of potable water was reported to be 65°F; the temperature of equilibrium of the system (i.e., fish and water) was 40°F at night and 55°F by the next day with an average of less than 48°F. The thawing time was 12 hours (overnight). In addition, the salmon was thawed in vats with set rather than flowing water. It can be reasonably assumed that at least a portion of the original *L. monocytogenes* population is redistributed during thawing, leading to a more normalized spatial distribution. This statement is supported by ICMSF (2002), which emphasized that, although in practice microorganisms may not often be randomly distributed, they are randomly distributed in mixed liquid samples. However, during food processing, microorganisms are commonly exposed to multiple potential lethal or sublethal stresses sequentially or simultaneously (Tiganitas and others 2009).

In the visited facility, frozen product was received 90% of the time and refrigerated product was received only 10% of the time for special orders. In the baseline model only frozen fish was considered. The visited facility reported using a calcium hydroxide solution for all incoming product. Food-grade calcium hydroxide $Ca(OH)_2$ is a GRAS additive that can be used to reduce *L. monocytogenes* contamination on headed and gutted (H&G) salmon (Yonker 2002; Himelbloom and others 2003; Jahncke and others 2004). Studies at the

University of Alaska used a water solution of calcium hydroxide (pH 12.9) for 3, 6, and 9 hours to reduce the counts of *L. monocytogenes* in H&G salmon. Raw salmon were inoculated at two different levels (~10^4 CFU/cm^2 and ~10^6 CFU/cm^2) and after being held in limed water for the specified times, the numbers of *L. monocytogenes* were reduced. For example, at the lower inoculum, the numbers of *L. monocytogenes* were reduced to less than 10^1 CFU/cm^2 after 6-9 hours. At a higher inoculum, *L. monocytogenes* were reduced to 10^3 CFU/cm^2 after 9 hours in limed water (Jahncke and others 2004). At the facility visited the calcium hydroxide solution was used overnight (12 hours) which implies a more stringent antimicrobial effect depending on the pH of the solution. Depending on whether or not an antimicrobial treatment is applied, very different and variable outcomes are possible at this step of the process. For example, the numbers of *L. monocytogenes* can be reduced if a $Ca(OH)_2$ solution is used, or, alternatively, the numbers may increase if the time and temperatures are abused when thawing with water. Although thawing could imply an increase or reduction in the numbers of *L. monocytogenes* on the surface, as explained, the baseline model assumed no increase or decrease of *L. monocytogenes* when thawing raw frozen H&G salmon. Furthermore, penetration of *L. monocytogenes* into intact flesh via the vascular system did not occur when frozen, headed, and eviscerated fish were thawed for 20 hours in water inoculated with 44 *L. monocytogenes* organisms per ml (Eklund and others 1995). This implies that it is likely that, when present, *L. monocytogenes* will remain on the surface of the product, which might facilitate its reduction when using a $Ca(OH)_2$ solution. In addition, the thawing step is followed by a rinse step (rinse 1) which it was assumed that could potentially remove injured *L. monocytogenes* cells remaining on the surface of the product.

Hardening of the salmon flesh might be another benefit of using calcium hydroxide; however, its main utility may be to reduce the microbial burden. The mechanisms of antimicrobial activity of calcium hydroxide were studied by Siqueira and Lopes (1999) for

non-food applications. Most recently, Starliper and Watten (2013) researched the use of elevated pH levels in different fish-associated organisms and found that a bacterial growth medium having the pH adjusted with sodium hydroxide to pH 10.0-12.0 proved to be an inhospitable environment for a variety of Gram-negative and Gram-positive bacteria. Although some Gram-positive bacteria are relatively tolerant to increased pH, e.g., *E. faecalis*, the effect of elevated pH usage on *Bacillus* sp. showed a 6 log reduction in 12 hours at a pH 10-11 and a 7 log reduction at pH 12 under the specified conditions of the study (Starliper and Watten 2013). Grabow and others (1978) showed reductions of 99.98% in total coliforms, 97.11% reduction in Enterococci, and 100% in enteric viruses with a retention time of 50 minutes at pH 11.1 in a wastewater reclamation plant. Starliper and Watten (2013) researched the inactivation of *Aeromonas salmonicida* subsp. *salmonicida* isolated from Atlantic salmon (*Salmo salar*) and found between 4 and 6 log reduction after 4 hours at pH 10-11 and complete inactivation after 4 hours at pH 12 as well as after 12 hours or more at pH 10-12. Considering the fact that refrigerated and frozen product are received at the visited processing facility with aerobic plate counts (APC) of 10^6 cfu/g and 10^2 cfu/g, respectively, thawing with a solution of $Ca(OH)_2$ at pH 10-12 could be considered a potential treatment to reduce the microbial burden in raw materials depending on the particular conditions of each process. One additional potential treatment for raw materials is washing raw fish with water containing chlorine (Jahncke and others 2004).

5.1.2.4 Rinse 1

After the frozen product is thawed, potable water is used to rinse the product thoroughly, which constitutes rinse 1 within the raw processing module. The potable water used in rinse 1 was assumed to be not contaminated with *L. monocytogenes.*

5.1.2.5 Filleting or splitting

After the first rinse at the processing facility, the salmon is split along the back from tail to head in two halves using a knife. A recontamination of the fish was assumed from the knife to the salmon. It was assumed that there was no additional cross-contamination with other utensils, equipment, or workers in regards to *L. monocytogenes* at the filleting level. In practice, the raw processing area should be separate from the rest of the processing areas to avoid cross-contamination and introduction of pathogens to the smoked product environment, and finished product should be separate from raw incoming product, which may be contaminated with *L. monocytogenes*. The degree of cross-contamination at the filleting level is likely small in comparison to the cross-contamination during the next step of the process, brining.

5.1.3 Brining Module

This module consists of the following three steps of the process: brining, rinse 2, and racking/hanging or equilibrating. The brining module worksheet summarizing the steps considered in this module is presented in Figure 5.8.

5.1.3.1 Brining

Brining is the process by which the fish is soaked in a solution consisting of water, salt, sugar, various spices and flavorings, phosphates, and, depending on the recipe and species of fish, additives such as sodium nitrite. Dry-salting involves placing fish for a certain period of time in a dry mixture of salt and other ingredients. Fish also may be brined by injecting the fish with a brine solution, either by hand or machine. Salting should be as uniform as possible, with the correct amount of salt or brine solution absorbed into each piece of fish flesh (IFT 2001). Salting times are empirically determined and may vary depending on the size of the fish pieces. For example, in the facility visited it was reported that fish weighing 2-3 pounds were salted for 24 hours. Fish processed with a dry-cure mixture are

typically held under refrigerated conditions in the salt mixture and are flipped after 24 hours. In the case of the processing facility visited, dry-salting and brining were performed under refrigeration prior to cold smoking.

In the brining step, recontamination of the product could occur from a contaminated sodium chloride solution, e.g., recycled or recirculated brine (non-pasteurized). In fact, studies have isolated *L. monocytogenens* from brine and fish flesh injected with contaminated brine (Eklund and others 1995). In the present model, it was assumed that *L. monocytogenes* present during bath brining was transferred to the fish surface. It was also assumed that the organism was recovering from injury from previous steps in the processing environment and had not completed its lag phase during this survival phase. Therefore, no growth was assumed.

Salt levels (salt in water phase) in CSS final product range from 3% to, in a few cases, as high as 12%, although salt levels typically range from 3.5% - 5% (Jørgensen and others 2000). This level of salt (3.5% - 5%) has no inhibitory effect on *L. monocytogenens* (Peterson and others 1993). Although levels above 6% NaCl with a low initial inoculum could prevent the growth of *L. monocytogenes* at 5°C (Peterson and others 1993), this level of salt is generally too high for consumer preferences. High levels of salt (>5.5%) may also significantly delay the growth of lactic acid bacteria, thus reducing their potential inhibitory effect against *L. monocytogenes* (Himelbloom, Nilsson, and Gram 2001).

5.1.3.2 Rinse 2

The potable water use for rinse 2 was assumed to be not contaminated with *L. monocytogenes*. For the purpose of considering the impact of potential antimicrobial rinses/treatments, this additional step was incorporated in the brining module. However, when running the baseline model this step was “not active” and did not influence the outcome of the module.

5.1.3.3 Racking or hanging

No cross-contamination with the equipment or workers was assumed. However, potential growth under refrigeration was considered during this step.

5.1.3.3.1 Modeling the growth characteristics of *L. monocytogenes* and native microflora in cold-smoked salmon

Hwang and Sheen (2009) indicated that the growth rate of *L. monocytogenes* and native microflora in cold-smoked salmon, regardless of product formulation, are similar. The native microflora in the smoked salmon in the Hwang and Sheen (2009) study were mainly lactic acid bacteria (LAB). The dominant microflora frequently isolated from cold-smoked salmon are LAB (Truelstrup Hansen and others 1997). LAB such as *Streptococcus*, *Leuconostoc*, *Lactobacillus*, and *Carnobacterium* belong to the normal microbiota of healthy fish (Ringø and Gatesoupe 1998). *Carnobacterium* are the dominant species found on cold-smoked salmon (Paludan-Muller and others 1998). *Carnobacterium* [*piscicola*] (now *Carnobacterium maltaromaticum*) (Mora and others 2003) may in some cases prolong the shelf life of this product (Leroi and others 1996) and inhibit the growth of *L. monocytogenes* in refrigerated food products (Buchanan and Klawitter 1991; Buchanan and Bagi 1997; Paludan-Muller and others 1998; Duffes and others 1999; Nilsson and others 1999; Lovdal 2015). For example, Nilsson and others (1999) successfully used high cell numbers of particular strains of *C. [piscicola] maltaromaticum* to control the growth of *L. monocytogenes* in cold-smoked salmon. The Nilsson and others (1999) study largerly observed decreases in *L. monocytogenes* levels during storage. Buchanan and Bagi (1997) investigated the interaction between *L. monocytogenes* and antilisterial strains of *C. [piscicola] maltaromaticum*, and found that suppression of *L. monocytogenes* was not always due to production of antilisterial compounds but could be partially attributed to nutrient depletion. It was concluded that the extent of suppression of *C. [piscicola] maltaromaticum* on *L. monocytogenes* was a function of the relative growth rates of the two bacteria, with

culture conditions that favor the growth of *L. monocytogenes* decreasing the suppression by *C. [piscicola] maltaromaticum*. Data on the growth characteristics of *L. monocytogenes* and the native microflora, and the growth relationship between the two in smoked salmon, are limited. More recently, Mejlholm and Dalgaard (2015) concluded that more studies are needed to understand the quantitative effects of microbial interactions, including the significance of different food matrices and their microbial substrates.

The Hwang and Sheen (2009) model was selected to estimate the growth of L. monocytogenes in cold-smoked salmon. Hwang and Sheen (2009) developed models to describe the growth characteristics of *L. monocytogenes* and native microflora specifically in smoked salmon at refrigerated and abuse temperatures. Mathematical equations were developed to describe the lag phase duration (LPD), exponential growth rate (EGR), and maximum population density (MPD) of *L. monocytogenes* and native microflora as a function of storage temperature in smoked salmon (Hwang and Sheen 2009). The EGR of *L. monocytogenes* in smoked salmon, after a square root transformation, is a linear function with temperatures proposed by Ratkowsky and others (1983). The linear relationship between the EGR and growth temperatures allows predictions for temperatures below the maximum growth temperature, 40°C (Ratkowsky and others 1983). These models describe the growth characteristics of *L. monocytogenes* and native microflora in smoked salmon as affected by storage temperatures at 4 to 16°C, and enable the estimation of LPD, EGR, and MPD of *L. monocytogenes* and native microflora in smoked salmon. The LPD and EGR values of *L. monocytogenes* and native microflora with low inoculum level were not significantly different ($P > 0.05$) from those with high inoculum level at each storage temperature (Hwang and Sheen 2009). This indicates that the growth rates (LPD and EGR) of *L. monocytogenes* and native microflora in smoked salmon at storage temperatures of 4 to 16°C were similar and not affected by the initial population levels of *L. monocytogenes* in smoked salmon. The

MPD of *L. monocytogenes* was 4.9 to 6.9 $\log_{10}$ CFU/g at 4 to 16°C, whereas the MPD of native microflora was approximately 8.6 $\log_{10}$ CFU/g. The MPD of *L. monocytogenes* was significantly higher at 16°C than those at 4, 8, and 12°C. Therefore, the growth of *L. monocytogenes* appeared to be more active and more competitive against the native microflora at higher storage temperatures. Similar MPD values were predicted by Mejlholm and Dalgaard (2015) for *L. monocytogenes* with microbial interactions.

5.1.4 Cold-Smoked Product Processing Module

This module is comprised of two steps, smoking and cooling. Smoking has traditionally been used to preserve fish. There are two different kinds of smoking processes based on the temperature used, hot smoking and cold smoking. The present study only considers cold smoking. Cooling is the step immediately after smoking in which the smoked fish is transferred to cold rooms to reach equilibrium at standard refrigeration temperatures. These two steps were modeled in the cold-smoking processing module worksheet depicted in Figure 5.9.

5.1.4.1 Cold smoking

In the United States, the temperature of the smoking chamber for cold-smoked seafood must either 1) not exceed 90°F (32.2°C) during a period of 20 hours or less or 2) not exceed 50°F (10°C) for a period of 24 hours or less (AFDO 2004a). According to Rørvik (2000), cold smoking of fish in Norway is usually performed at temperatures below 21°C (69.8°F). These variations in temperatures are dependent on different regions and specific plant requirements in which the cold-smoking process takes place. Studies indicate that short term cold-smoking (<24 h), as recommended by the Association of Food and Drug Officials guidelines (AFDO 1991), reduces rather than increases the number of *L. monocytogenes* (Eklund and others 1995; IFT 2001). The cold-smoking process uses liquid smoke, or wood smoke, produced by the pyrolysis of wood chips under controlled conditions. The visited

smoking facility used natural wood smoke. Wood smoke is considered a natural flavor, whether it is applied as a gas from smoldering wood chunks or chips or as liquid smoke (Toledo 2008). In the cold-smoking step, the fish must be arranged to allow for uniform smoke absorption, temperature exposure, and drying.

5.1.4.1.1 Drying during smoking

Wood smoke or liquid smoke may be used as a component of a hurdle system for food preservation. A number of cold-smoking procedures include a drying stage prior to smoking of the product which could be considered another hurdle strategy by lowering the water activity of the product. The product is held at a specified temperature, often refrigeration temperature, for a specified amount of time before the smoke is introduced. The parameters of this initial drying depend on the type or species of fish, its fat content, and humidity levels. During this time, a pellicle forms on the outside surfaces of the fish pieces. It should be noted that smoke must be applied to the product before the surface dries, otherwise *L. monocytogenes* will be embedded under the pellicle where the effect of smoke is markedly reduced (IFT 2001).

5.1.4.1.2 Potential substrate effect

Traditionally, phenolic compounds present in smoke combined with other factors have been associated with listericidal or listeriostatic effects. For example, the combination of 20 ppm phenols and 4% NaCl was inhibitory at 4 to 12°C in nutrient broth (Membré and others 1997). However, similar studies in smoked salmon found no inhibition of *L. monocytogenes* using even higher phenolic concentrations (Cornu and others 2006). These discrepancies may be explained by a difference between the behavior of phenolic compound in broth versus fish substrate. It is important to recognize that the solubility of phenolic compounds in the water phase of a fatty fish is unknown (Cornu and others 2006). Figure 5.17 provides a SEM close-up of the cold-smoked fatty fish surface. Suñen and others (1998)

found that liquid smoke extracts differed considerably not only in composition and relative concentrations but also in their ability to inhibit the growth of pathogens. Based on this study, IFT (2001) concluded that the listericidal effect of liquid smoke will depend on the particular product in use. In addition, the potential inhibitory properties of liquid smoke will vary depending on type of wood, method of preparation, and target organism (IFT 2001).

5.1.4.1.3 Potential smoke components antimicrobial effect

Different varieties of wood generate different levels of phenols, carbonyls, and organic acids upon pyrolysis, which affects their antimicrobial properties (Lingbeck and others 2014). Smoked salmon contains generally 2 to 15 ppm phenolic compounds. Studies published in the United States (Yoon and others 2004; Burnett and others 2005) used 6 ppm phenolic content. In contrast, European studies such as Giménez and Dalgaard (2004) and Lakshmanan and Dalgaard (2004) used smoked salmon with higher concentrations of phenolic compounds, i.e., 12.6 ppm and 14.6 ppm, respectively. Other studies from the United States and Europe used even higher phenol content, for example, Vitt and others (2001) and Porsby and others (2008), which used 18-25 ppm and 22-26 ppm, respectively. However, phenolic concentration is probably not sufficient to assess the antimicrobial activity of smoke (Cornu and others 2006). For example, Suñen (1998) found that the fraction with the strongest antimicrobial properties not only contained high levels of phenols (21 mg/kg) but also the highest concentration of acids (34 mg/kg). Further studies suggest that the antimicrobial properties of liquid smoke are not attributable to their phenolic composition. Specifically, Suñen and others (2001) found that the fraction with most antimicrobial activity was lowest in phenol concentration (23 mg/Kg) but highest in acid concentration (23 mg/Kg), while the least effective fraction contained high levels of phenols (99 mg/Kg). One should note that carbonyls and acids can also have a wide spectrum of antibacterial activity even at low levels of phenols (Milly 2003; Milly and others 2005; Montazeri and others 2013b;

Toledo 2008). In particular, organic acids have shown the most antimicrobial activity among the functional components of smoke (Milly and others 2005; Toledo 2008). Similarly, organic acids have also been considered the main factor causing inactivation of *L. monocytogenes* in most studies of non-thermal inactivation of this pathogen (Buchanan and others 1993; Buchanan and Golden 1994, 1998; Golden and others 1995). Most recently, Mejlholm and Dalgaard (2015) used benzoic and sorbic acid, which are more soluble in lipids than acetic and lactic acids, to prevent the growth of *L. monocytogenes* in mayonnaise-based salads.

5.1.4.1.4 Efficacy of commercial liquid smokes against Listeria spp. in cold-smoked salmon

Montazeri and others (2013) investigated the antilisterial properties of liquid smoke against *L. innocua*. In vitro assays showed strong inhibition for most commercial liquid smokes at 1% (vol/wt.) when inoculated with *L. innocua* at 3.5 log CFU/g, vacuum packaged, and stored at 4°C. Although the liquid smoke did not completely eliminate *L. innocua*, it provided approximately 2-log reduction by day 14, with no growth up to 35 days of refrigerated storage. This could be considered similar to the reduction of *L. monocytogenes* from 10^3 to 10-10^2 log cfu/cm^2 immediately after cold smoking salmon reported by Porsby and others (2008). Montazeri and others (2013) found that the application of liquid smoke is an effective antilisterial additive for cold-smoked salmon. In like manner, Lingbeck and others (2014) concluded that liquid smoke is an effective antimicrobial in food systems and has several advantages over traditional smoking techniques including ease of application, speed of smoking process, and omission of hazardous polycyclic aromatic hydrocarbons. In this study, it was assumed that natural wood smoke had the same effect on *L. monocytogenes* as liquid smoke. Table 5.1 shows the antimicrobial effect of liquid smoke against *L. monocytogenes*. A cold-smoking empirical distribution estimating the log reduction achieved by this step was developed for the model based on the findings by Montazeri and others

(2013), including the variability found by published studies (see Table 5.1). It was assumed that the actual reduction in the levels of *L. monocytogenes* caused by the liquid smoke happened immediately after the cold-smoking step of the process. It is noteworthy that Rørvik (2000) reported that 54% of 200 samples of salmon were positive for *L. monocytogenes* just before smoking, whereas only 9.5% were positive after smoking. The specific antimicrobial effects of smoke on *L. monocytogenes* were studied by Guilbaud and others (2008). This study suggested that liquid smoke affects the synthesis of the cell membrane, reduces the hemolytic activity of *Listeria*, and may reduce its virulence.

5.1.4.2 Cooling

After smoking, the cooling step helps equilibrate the smoke throughout the fish prior to slicing. The fish must be cooled to 70°F (21°C) or less within 2 hours of the smoking process and from 70°F (21°C) to 41°F (5°C) or less within 4 hours. The cooling time should not exceed 6 hours from start of cooling (AFDO 2004a). It was reported during the visit to the smoking facility that cold-smoked salmon reached 25°C within 2 hours. The cooling took place in a refrigeration unit that had the capacity to assure the temperature was maintained at 41°F (5°C). AFDO (2004a) recommends maintaining potentially hazardous foods at temperatures at or below 38°F (3.3°C). Also, cooling involves minimal contact of the product by workers, so no recontamination was assumed at this level. However, growth of *L. monocytogenes* under refrigeration during cooling was considered.

5.1.5 Post-cold Smoking Processing Module

This module includes removal of the pinbone and subsequent slicing and portioning. After slicing and portioning, the products are vacuum-packed, labeled, boxed, and generally stored frozen until they are distributed. These steps were modeled in the post-cold smoking processing module worksheet depicted in Figure 5.9.

5.1.5.1 Pinbone

This step is usually done by vacuuming the skin and bones. Optional trimming or cutting prior to slicing may also be included. Potential recontamination was considered.

5.1.5.2 Slicing

For over two decades, slicers have been identified as a source for contamination and its spread through food lots in ready-to-eat products. For example, an examination of retail meat slicers revealed a contamination rate of 13% with *L. monocytogenes* (Humphrey and Worthington 1990). More recently, a survey of retail operations in the state of New York found *L. monocytogenes* in the environment in 60% of these establishments; however, it was only detected in 3% (5/183) of the slicers sampled (Hoelzer and others 2012). This shows an improvement that could be attributed to better sanitary design of equipment (i.e., slicers), better sanitation procedures and practices, stringent regulations, etc. However, there is also the possibility that the pathogen in the equipment sampled was not detected. There are a number of factors that influence detection rates, such as the biological state of the bacterium, the specific location and surface structure within equipment (e.g., areas of difficult access during and after cleaning and sanitation which may be in contact with the product), and the material (e.g., stainless steel grade) and friction coefficients associated with different areas of the equipment sampled (e.g., blades). In addition, the moisture, fat, and/or protein residues that may be found around the selected sampled areas; the different conditions at which equipment was sampled prior, during, or after processing; the area sampled; and the nature and intensity of the contact during sampling may all impact detection rates. These factors may also influence the transfer coefficients of bacteria from the slicer to the product. There are many other conditions that will affect the transfer of *Listeria* during the slicing step of the process, including the physicochemical nature and composition of the food product itself. Fish is a protein rich food and, in the case of salmon, there is also high fat content. In

addition, another factor that may affect not only the detection or recovery of *L. monocytogenes* but also its ability to transfer from slicers to the product is the sanitary design, especially if the equipment is not allowed to be disassembled. Some slicers, due to their challenging design and the inherent difficulty of cleaning and sanitizing them, cannot be disassembled, and thus become a potential source of recontamination of the final product. A calculation of transferring of *Listeria* cells under normal conditions assumes that *Listeria* species are evenly distributed across food contact surfaces and *L. monocytogenes* are evenly distributed in a food product batch. The model of Aarnisalo and others (2007) for *L. monocytogenes* during slicing of "gravad" salmon was considered with an exponential equation: $Y = a*e^{(-x/b)}$ of transfer as a function of slice number (x). This exponential model is specific for salmon and it accounts for the roughness of the slide blade as well as the composition of the salmon fillets. It only considers the transfer from slicer machine to slices. Transfer from the fish to the slicing machine and then to the slices was not considered. This model was graphed within the post-cold smoking processing module worksheet. However, the processing facility visited was equipped with large scale slicers with several blades. The facility had two different kinds of large scale slicers. The newer equipment was disassembled on a daily basis whereas the older equipment did not have this capability. Equipment that cannot be disassembled for cleaning further complicates the cleaning and sanitation procedures and is anticipated to have greater variability in contamination.

5.1.5.3 Portioning

A division into portion sizes was performed by workers using gloves and weighting the products in small trays right before vacuum packaging.

5.1.5.4 Packaging

Packaging is the step in which the product, after being divided into portions, is sealed individually creating a barrier from the environment. Processing plants use either vacuum or

air packaging for cold-smoked salmon. The safety of the product is as good as the wholeness and integrity of the packaging. However, traditional packaging per se would not be assumed to effectively reduce or eliminate *L. monocytogenes* in this risk assessment. This pathogen grows well under aerobic and anaerobic conditions and at refrigeration temperatures (Jinneman and others 2007). These properties make *L. monocytogenes* a potential threat to the safety of foods packaged under vacuum or modified atmospheres (Church and Parsons 1995). Growth of *L. monocytogenes* was not inhibited in food that has been packaged under vacuum (Hudson and Mott 1993). However, combining different hurdle technologies in addition to vacuum packaging with specific characteristics (e.g., 100% CO_2) may suppress the growth of *L. monocytogenes* (Szabo and Cahill 1999). It was assumed that no recontamination of the product occurred during packaging.

5.1.5.5 Labeling

No growth in this step of the process was assumed. However, labeling could represent a bottleneck in some instances and delays could occur at this step of the process. It was assumed that the time at which the product was held at refrigeration temperatures (prior to freezing) was too short to permit the growth of *L. monocytogenes*. Packages must contain a "keep refrigerated" statement (e.g., "Important, keep refrigerated until used.") for finished product to ensure that temperature controls are applied throughout distribution and at the consumer level.

5.1.5.6 Boxing

No growth in this step of the process was assumed. However, delays could occur at this step of the process. It was assumed that the product was put in boxes prior to freezing and that the time during which the product was held under refrigeration was too short to permit the growth of *L. monocytogenes*. Boxes must contain a "keep frozen" (e.g., "Important, keep

frozen until used, thaw under refrigeration immediately before use") statement to ensure that temperature controls are applied throughout distribution as intended.

5.1.5.7 Freezing

Controls should be in place to ensure that cold-smoked salmon is immediately frozen after processing. Depending on the efficiency of the freezing method, the time to achieve <0°C at the center of a package needs to be considered. The time to achieve such temperatures is dependent on whether they are frozen before or after boxing. The time to freezing is substantially shorter if initial freezing of individual packages occurs prior to boxing instead of after boxing, as this minimizes the duration of refrigeration temperatures at this level. It was assumed that the freezing step was done immediately after processing, labeling, and boxing. It was assumed that the time at which the product was held at refrigeration temperatures during the freezing process was too short to permit the growth of *L. monocytogenes*. Therefore, no growth of this pathogen was assumed.

5.1.5.8 Final product storage

Although frozen storage is preferred over refrigerated storage for cold-smoked salmon, this product could be stored refrigerated or frozen depending on many factors, including the duration of the shipment and required shelf life based on consumption patterns. In the baseline model, it was assumed that the product was stored frozen and that no growth of *L. monocytogenes* was possible during the period of frozen storage.

5.1.6 Distribution and Marketing Module

The distribution and marketing module was divided into three steps: the frozen transportation from the processing facility to the retail market, the retail frozen storage, and the thawing and subsequent refrigerated retail display.

5.1.6.1 Transportation to retail

The frozen cold-smoked salmon is transported from the processing facility to retail markets. Since this ready-to-eat product was assumed to be frozen, growth of *L. monocytogenes* was not a concern at this level; however, if the final product was refrigerated, adequate refrigerated transportation would be critical to avoid temperature abuse during the distribution process.

5.1.6.2 Retail frozen storage

Frozen storage during transport and frozen retail storage would not be expected to support the growth of *L. monocytogenes*. Controls should be in place to ensure that frozen commercial temperatures for cold-smoked salmon are maintained throughout distribution.

5.1.6.3 Retail display

Although retail display could benefit from keeping the product frozen, cold-smoked salmon is usually displayed refrigerated. These refrigerated storage conditions during retail display after thawing were modeled using data from Ecosure (2008). Though growth within a package would occur, it was assumed that the packaging of the product remained intact and thus there was no recontamination or cross-contamination of the product.

5.1.6.4 Estimating levels of L. monocytogenes *contamination in cold-smoked salmon*

The distribution of frequency and levels of *L. monocytogenes* contamination in cold-smoked salmon were estimated from studies and surveys carried out worldwide during the last 20 years (Appendix 2.3.2). These studies (see Table A2.5) indicated substantial variability in both the frequency and extent of contamination, with incidence rates ranging from 0% to 78.7%. Several studies revealed high incidence of *L. monocytogenes* in smoked finfish, with most ranging from 15 to 20% (Johansson and others 1999; Inoue and others 2000; Dominguez and others 2001; Gombas and others 2003; Besse and others 2004;

Nakamura and others 2004; Van Collie and others 2004; Beaufort and others 2007; Latorre and others 2007, Uyttendaele and others 2009). More recently, Rotariu and others (2014) reported that fifty-six percent of the Scottish smoked salmon processors (mostly large and medium size companies) tested the final product for *L. monocytogenes* and found an incidence of 0 to 12%. This implies that active testing results in a substantially lower incidence rate than smaller processors that do not test. The cumulative frequency for the incidence of *L. monocytogenes* in cold-smoked salmon is depicted in Figure A2.3. In addition, the probability distribution for the level of *L. monocytogenes* in contaminated cold-smoked salmon is reported in Figure A2.4. This probability distribution represented the levels of *L. monocytogenes* at the end of the process or during retail distribution. For example, Jørgensen and Huss (1998) followed the change in numbers and incidence of contamination with *L. monocytogenes* during the normal shelf life (50 days) of cold-smoked salmon. They found a progressive increase in both the number of positive samples and the mean concentration level in those samples (Table 5.2). The data presented in Table 5.2 suggest that up to a further 30% of samples positive at the point of processing may be incorrectly identified as not contaminated with *L. monocytogenes*. This might be due to limitations in the detection of *L. monocytogenes* at low concentrations. It is noteworthy that food products are usually contaminated at low levels and there is still a need for improvement of *Listeria* enumeration methods, particularly at low levels of concentration (Auvolat and Besse 2016).

5.1.7 Consumer Module

The consumer level has been shown to be a key component in the farm-to-fork continuum. Consumers constitute the final step in the food chain and their behavior is critical in minimizing the risk of foodborne disease, including listeriosis (Yang and others 2006). The consumer model includes three parts: the transportation of the cold-smoked salmon from retail store to the consumer's refrigerator, the refrigeration of the product by the consumer,

and the preparation of the product prior to consumption. These steps were modeled within the consumer module worksheet depicted in figure 5.11.

5.1.7.1 Transportation by consumer

Growth of *L. monocytogenes* due to potential temperature and time abuse could be possible during the transportation of cold-smoked salmon by the consumer. A distribution was created within the control panel spreadsheet in the baseline model to represent this step of the product pathogen pathway.

5.1.7.2 Refrigerated storage by consumer

Growth due to potential temperature and time abuse was assumed during the refrigerated storage of cold-smoked salmon at the consumer level. A distribution was created within the control panel spreadsheet in the baseline model including the mean product temperature (38.2°F or 4.33°C) for home refrigerators published by Ecosure (2008).

5.1.7.3 Serving final concentration

After refrigerated storage at the consumer level, it is assumed that cold-smoked salmon is immediately served prior to consumption. No growth of *L. monocytogenes* was assumed at this level. This step was modeled as the output within the consumer module worksheet and represents the final concentration of *L. monocytogenes* immediately prior to consumption.

5.1.7.3.1 Serving size distribution

The serving size of cold-smoked salmon was described by the empirical distribution RiskCumul(57,142,{75,136,142},{0.75,0.95,0.99}) in grams of food eaten per serving. This distribution shows the 50th (median), 75th, 95th, and 99th percentiles of the weighted distributions of serving size. These percentiles for smoked seafood are 57, 75, 136, and 142 grams per serving, respectively. This distribution indicates that half of the servings were less than 57 grams and 95% of the servings were less than 136 grams. This distribution was based

on data used by the 2003 FDA/USDA Risk Assessment (see Table 5.3). These values were also used by other *Listeria* risk assessments (FDA/USDA 2001; FAO 2004) which used equivalent distributions. It should be noted that the original values used to generate the distribution of serving sizes of cold-smoked fish used in the simulation model were obtained from the Continuing Survey of Food Intakes by Individuals undertaken in the United States of America (CSFII) and the National Health and Nutrition Examination Survey (NHANES). It is important to recognize the limitations of existing data throughout the exposure assessment. The serving size was modeled within the dose-response module worksheet.

5.2 *Hazard Characterization*

Although hazard characterization could include a broader scope considering sequelae and severity assessment (Buchanan and Lindqvist 2000), it is often described as dose-response assessment within the food safety community. In this study, the hazard characterization was addressed as the dose-response module.

5.2.1 Dose-response Module

The probability of illness (listeriosis) was mathematically represented by the exponential dose-response model from FAO/WHO (2004) which was described in Chapter 3. The predictions generated by this dose-response model for the susceptible population, part of the FAO/WHO Risk Assessment of *L. monocytogenes* in Ready-to-Eat Foods, were compared by Hoelzer and others (2013) with other selected published models for *L. monocytogenes* developed using different data sets. The FAO/WHO (2004) model showed acceptable performance. This exponential model was based on the dose-response relationship between exposure estimates and infection rates (Buchanan and others 1997a). The dose-response model was combined with the serving size distribution (see section 5.1.7.3.1) and the modeled contamination level data to predict the probability of illness. The distributions

used for the dose-response module worksheet are included in Table 5.4. The probability of illness obtained from the exponential dose-response module using the different r-values and the serving size distribution are described in the next section, risk characterization.

5.3 *Risk Characterization*

The quantitative risk characterization summarizes all previous steps within a probabilistic risk assessment. In other words, it is the integration and interpretation of the previous steps by combining the dose-response model with the exposure assessment model. The hazard characterization of the cold-smoked salmon model aimed at describing the relationship between the level of *L. monocytogenes*, resulting from the exposure assessment, and the probability of subsequent development of listeriosis and resulting adverse health outcomes on susceptible populations. Chapter 5 estimates risk predicted by the cold-smoked salmon model using cases of listeriosis per million servings. While risk estimates of listeriosis caused by consumption of cold-smoked salmon could be calculated for different populations, this was deemed unnecessary since the susceptible population represents the vast majority of listeriosis cases. Furthermore, the primary goal of the risk assessment is to identify the product's critical control points using sensitivity analysis. Risk estimates for the susceptible population using the FAO/WHO (2004) exponential model were considered sufficient.

5.3.1 Risk Assessment Model Estimate

The baseline model predicts a median probability of listeriosis of 4.45×10^{-9}, which represents 0.0045 cases of listeriosis per million servings. In addition to the baseline model, 25 different scenarios were developed, each accounting for different conditions. The mean of medians of the probability of illness of the first twenty-four scenarios resulted in 9.1×10^{-9}, which represents 0.0067 cases of listeriosis per million servings. The FAO/WHO *L.*

monocytogenes ready-to-eat risk assessment model for cold-smoked fish estimated 0.053 cases of listeriosis per million servings. It is noteworthy that because of the combination and pooling of data from many diverse sources, the risk estimates may not accurately represent the situation for every cold-smoked salmon processing facility. Figure 5.14 shows the cases per million servings and the mean of medians estimated for the different scenario categories within the what-if scenarios worksheet. The results of these scenarios is discussed in Chapter 6.

5.3.2 Potential Impacts of the Process on Levels of *L. monocytogenes* in Cold-Smoked Salmon

There are several factors contributing to the partial control of *L. monocytogenes* in cold-smoked salmon. These factors range from the prerequisites of the HACCP system, to different steps inherent to the process of cold smoking, to mitigation strategies applied at different steps of the process. The potential impact of factors contributing to the partial control of *L. monocytogenes* in the cold-smoking process at the end of the food chain (consumer level) in the baseline model are depicted in Figure 5.15.

The impact of steps contributing to the partial control of *L. monocytogenes* in cold-smoked salmon was quantitatively estimated using sensitivity analysis, which identifies factors that are strongly positively or negatively associated with risk. Such factors are strong candidates for control to manage risk. The main step in the process negatively associated with risk (i.e., provided a partial reduction in the concentration of the pathogen) was cold smoking. Several studies have shown that the cold-smoking step produces a reduction in the concentration of *L. monocytogenes* (Fonnesbech Vogel and others 2001; Gram 2004; Porsby and others 2008; Montazeri and others 2013). Efforts to optimize the reductions produced in the concentration of *L. monocytogenes* at the cold-smoking level (e.g., phenolic concentrations, temperature/time combinations) could be considered to minimize the risk.

Figure 5.16 shows the impact of different log reductions in the concentration of *L. monocytogenes* caused by cold smoking. If the factors found in the sensitivity analysis are positively associated with risk, then risk mitigation strategies could be introduced to reduce the concentration of *L. monocytogenes* at those steps in the process. These mitigation strategies (e.g., application of antimicrobials at the thawing or rinse steps) can be used to partially control *L. monocytogenes*. Mitigation strategies are pivotal to achieving a performance objective at the end of the process. The total sum of the impacts (whether increases or reductions) of any combination of potential mitigation strategies in the cold-smoked salmon process is variable, and monitoring systems are crucial to control them.

Table 5.1 Chemical properties and efficacy of commercial liquid smokes as an antimicrobial against Listeria spp. in food systems

Liquid Smoke Tested	Manufacturer	Liquid smoke concentration	Titrable acidity acetic acid % (wt/wt)	pH	Phenol Content (mg/ml)	Carbonyl Content (g/100ml)	Processing parameters	Strain	Result	References
CharSol Supreme	Red Arrow Company (Manitowoc, WI)	60%	14-16	2.1-2.6	18-25	20-25	Salmon samples dipped in LS inoculated and smoked	*L. innocua*	3 log reduction (15s dip)	Vitt and others 2001
AM-3 AM-10	Mastertaste, Inc.	0.9%	2.2-2.3	4.2-4.3	Not listed	Not listed	Salmon strips treated with LS, inoculated vacuum sealed and stored at 4°C for up to 49 days	*L. innocua* ATCC 33090	<2 log CFU/g reduction (after 2 weeks)	Montazeri and others 2013; 2013b
CharSol-10	Red Arrow Company	100%	10.5-12	2.1-2.6	10-15	12-13	Frankfurters were inoculated, dipped in LS, vacuum packed and stored at 4°C for 72h	*L. monocytogenes* LCDC 4b	1 log initially and undetectable after 72 h	Messina and others 1988

Table 5.1 Chemical properties and efficacy of commercial liquid smokes as an antimicrobial against Listeria spp. *in food systems (Continued)*

Zesti Smoke	Mastertaste, Inc.	Formulated into product at 10,5, 2.5% (wt/wt)	Not listed	Not listed	Not listed	Not listed	Frankfurters were inoculated, vacuum packed and stored at 4°C for 12 weeks	*L. monocytogenes*	0.5-2 log CFU/ml reductions	Morey and others 2012
Zesti-B	Mastertaste, Inc. (Monterey, TN)	100%	3.5-5.6	2.5-3.3	1.7 max	19-22	Deli turkey dipped in LS, inoculated, vacuum packed, pasteurized, chilled and stored at 6.1°C for 10 weeks	*L. monocytogenes* Scott A	2 logs CFU/g (1s dip)	Gedela and others 2007

Table 5.2 Increase of contamination prevalence in cold-smoked salmon during storage due to growth of L. monocytogenes *to detectable levels*

Days of Storage	**Total Positive Samples (%)**	**% of *L. monocytogenes* positive samples with varying levels of contamination (MPN/g)**				**Mean MPN/g**	**Number of Samples**
		<10	**10-100**	**100-1000**	**>1000**		
0	34	28	5	1	0	220	190
20	40	10	20	9	2	3900	115
50	43	23	15	3	3	4300	75

Source: Data of Jørgensen and Huss, 1998.

Table 5.3 Values used to generate the distribution of serving sizes of cold-smoked salmon used in the quantitative risk assessment model for L. monocytogenes

Serving Size (g)	**Cumulative Probability**
0	0.00
57	0.50
75	0.75
136	0.95
142	0.99
284	1.00

Source: FDA/USDA 2003.

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Raw Salmon Reference Distribution	C10	Distribution based on Lm incidence using weighted mean and weighted standard deviation from Literature (Appendix 2)	RiskLognorm(20.27/100,20.83/100,RiskTruncate(0,100/100))
Raw Salmon Reference Distribution	C16	Binomial distribution used to direct sampling to describe Lm incidence in raw salmon	RiskBinomial(1,C10,RiskName("Raw Salmon Lm Incidence"))
Raw Salmon Reference Distribution	C17	Half the Lower Limit of Detection	0.02
Raw Salmon Reference Distribution	E26	Alt Lognormal distribution used to define the concentration values E23=5%, C23=0.04, E24=50%,	=RiskLognormAlt(E23,C23,E24,C24,E25,C25,RiskName("Initial Lm Reference Distribution")) C24=0.1, E25=95%, C25=0.4
Raw Salmon Reference Distribution	E28	Output concentration of Lm in raw salmon incoming or Primary production	=RiskOutput("Lognormal Result")+IF(C16=0,C17,E26)
Primary Production Module	B10	Frozen Raw Salmon at Supplier	CPanel!E3
CPanel	E3	Initial Concentration (Log cfu/g)	LOG('Raw Salmon Reference Distrib.'!E28)*E47
CPanel	E47	Initial concentration of bacteria under what-if scenarios	RiskSimtable('What-if Scenarios'!D5:D29)
Primary Production Module	D10	Frozen Transportation	=B10
Raw Product Processing Module	C12	Receiving Frozen Raw Salmon	='Primary Production Module'!$D$10
Raw Product Processing Module	C10	Receiving Frozen Raw Salmon	=10^C12

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Raw Product Processing Module	E10	Frozen Storage	=C10
Raw Product Processing Module	G10	Thawing	=E10*CPanel!E48
CPanel	E48	Thawing under What-if scenarios	=RiskSimtable('What-if Scenarios'!E5:E29)
Raw Product Processing Module	I10	Rinse 1	=G10
Raw Product Processing Module	L10	Filleting	=I10+RiskMakeInput(RiskUniform(0.01,0.1)*RiskPert(0.001,0.01,0.1),RiskName("Filleting"))
Raw Product Processing Module	L12	Filleting (Output in Log cfu/g)	=RiskOutput("Raw Prod Proc Module Log cfu Output")+LOG(L10,10)
Brining Module	C11	Brining	='Raw Product Proc. Module'!$L$12
Brining Module	C9	Brining	=10^C11
CPanel	E12	Brining Concentration	='Brining Module'!C9+RiskMakeInput(F12*I12,RiskName("Brining"))
CPanel	F12	Brining Concentration	=RiskLognorm(G12, H12), G12=Mean=0.005, H12=SD=0.1
CPanel	I12	Brining Concentration	=RiskPert(J12,K12,L12), J12=Min=0.001, K12=ML=0.01, L12=Max=0.1
Brining Module	D9	Brining	=CPanel!E12
Brining Module	D11	Brining	=LOG(D9,10)

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Brining Module	D15	Brining	=D11
Brining Module	E9	Brining	=10^D15
Brining Module	G9	Rinse 2	=E9-G13
Brining Module	G13	What-if Scenario Option	=CPanel!E21
CPanel	E21	Rinse#2 - Scenario	=0 (Assigned value of zero)
Brining Module	G11	Rinse 2	=LOG(G9,10)
Brining Module	F45	MPD (Log CFU/g)	=3.65+0.18*(G17)
Brining Module	G17	Temperature Brining Truncated	=IF(F17<4,4,F17)
Brining Module	F17	Temp Brining Step	=CPanel!E13
CPanel	E13	Temperature Brining	=RiskTriang(0.5,1.4,5.6,RiskName(D13))
Brining Module	J11	Hanging/Racking/ Equilibrating (Refrig. Storage 1)	=RiskOutput("Brining Module Log CFU Output")+IF(G11>F45,F45,G11)
Cold Smoking Module	C15	Cold Smoking	='Brining Module'!$J$11
Cold Smoking Module	D15	Cold Smoking	=C15-RiskTriang(CPanel!J26,CPanel!K26,CPanel!L26,RiskName("Cold Smoking"))

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
CPanel	J26	Cold-Smoking Reduction - Scenario	=RiskSimtable('What-if Scenarios'!F5:F29)
CPanel	K26	Cold-Smoking Reduction - Scenario	=RiskSimtable('What-if Scenarios'!G5:G29)
CPanel	L26	Cold-Smoking Reduction - Scenario	=RiskSimtable('What-if Scenarios'!H5:H29)
Cold-Smoking Module	F15	Equilibrating (Cooling)	=D15
Cold-Smoking Module	G54	MPD (log CFU/g)	=3.65+0.18*(Temp) Temp=CPanel!E23
CPanel	E23	Temperature After Cold-Smoking (Cooling)	=RiskTriang(1.4,4.2,32.2, RiskName(D23))
Cold-Smoking Module	I15	Equilibrating (Cooling)	=RiskOutput("Cold-Smoking Log CFU Output")+IF(F15>G54,G54,F15)
Post-cold Smoking Module	C11	Pinbone (vacuum) Skinning/triming/ cutting	='Cold Smoking Module'!$I$15
Post-cold Smoking Module	C9	Pinbone (vacuum) Skinning/triming/ cutting	=10^C11
CPanel	F29	Pinbone	=RiskLognorm(G29, H29) G29= 0.0005

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
			H29=0.1
CPanel	I29	Pinbone	=RiskPert(J29,K29,L29) J29=0.01 K29=0.1 L29=1
CPanel	E29	Pinbone	='Post-cold Smoking Module'!C9+RiskMakeInput(F29*I29,RiskName("Pinbone"))
CPanel	E49	Pinbone (vacuum) Skinning/triming/ cutting	=RiskSimtable('What-if Scenarios'!I5:I29)
Post-cold Smoking Module	H9	Pinbone (vacuum) Skinning/triming/ cutting	=CPanel!E29
Post-cold Smoking Module	H11	Pinbone (vacuum) Skinning/triming/ cutting	=LOG(H9,10)-CPanel!E49
CPanel	F28	Slicing	=RiskLognorm(G28, H28) G28= 0.00523250425549648 H28= 0.1
CPanel	I28	Slicing	=RiskPert(J28,K28,L28) J28= 3.16

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
			K28= 5.01 L28= 10
CPanel	E28	Slicing	=10^'Post-cold Smoking Module'!H11+RiskMakeInput(F28*I28,RiskName("Slicing"))
Post-cold Smoking Module	J9	Slicing	=CPanel!E28
Post-cold Smoking Module	M8	Portioning	=RiskLognorm(0.00505, 0.1)
Post-cold Smoking Module	M9	Portioning	=J9+RiskMakeInput(M8*RiskPert(0.001,0.01,0.1),RiskName("Portioning"))
CPanel	E27	Vacuum Packaging	='Post-cold Smoking Module'!M9+RiskMakeInput(F27*I27,RiskName("Vacuum Packaging"))
CPanel	F27	Vacuum Packaging	=RiskLognorm(G27, H27) G27= 0.0005 H27=0.1
CPanel	I27	Vacuum Packaging	=RiskPert(J27,K27,L27) J27=0.001

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
			K27=0.01 L27=0.1
Post-cold Smoking Module	P9	Vacuum Packaging	=CPanel!E27
Post-cold Smoking Module	P11	Vacuum Packaging	=LOG(P9,10)
Post-cold Smoking Module	T11	Labeling	=P11
Post-cold Smoking Module	V11	Boxing	=T11
Post-cold Smoking Module	X11	Freezing	=V11
Post-cold Smoking Module	Z11	Final Product Frozen Storage	=RiskOutput("Post_Cold_Smoking_Module_Output")+X11
Distribution and Marketing Module	C11	Frozen Transportation	='Post-cold Smoking Module'!$Z$11
Distribution and Marketing Module	D11	Frozen Transportation	=C11

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Distribution and Marketing Module	G11	Retail Frozen Storage	=D11
Distribution and Marketing Module	I11	Retail Display Refrigerated Storage	=G11+F41*(F23-0.9*F23)
Distribution and Marketing Module	F41	Retail Display Refrigerated Storage Sq. Root of GR (log CFU/h)	=0.0529+0.011*(Temp) Temp=CPanel!E32
CPanel	E32	Retail Display Refrigerated Storage	=RiskTriang(J32,K32,L32,RiskName(D32)) J32=-17 K32=5 L32=21 D32=Temp Distribution
Distribution and Marketing Module	F23	Retail Display Refrigerated Storage	=CPanel!E31
CPanel	E31	Retail Display Refrigerated Storage	=RiskPert(J31,K31,L31,RiskName(D31))*E50 J31=2.4 K31=36

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
			L31=168 D31= Time Distribution
CPanel	E50	Retail Display Refrigerated Storage	=RiskSimtable('What-if Scenarios'!J5:J29)
Distribution and Marketing Module	F43	MPD (log CFU/g)	=3.65+0.18*(Temp) Temp =CPanel!E32 =RiskTriang(J32,K32,L32,RiskName(D32))
Distribution and Marketing Module	I19	Retail Display Refrigerated Storage	=RiskOutput("Distribution_Module_Output")+IF(I11>F43,F43,I11)
Consumer Module	C11	Transportation by Consumer	='Distrib. and Marketing Module'!$I$19
Consumer Module	D11	Transportation by Consumer	=C11+F41*(F23-F23)
Consumer Module	F41	Sq. Root of GR (log CFU/h)	=0.0529+0.011*(Temp) Temp=CPanel!E34
CPanel	E33	Time Consumer	=RiskPert(J33,K33,L33,RiskName("Time Consumer"))*E51
CPanel	E51	Consumer Time (hrs)	=RiskSimtable('What-if Scenarios'!N5:N29)

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Consumer Module	F23	Refrigerated Storage by Consumer	=CPanel!E33
Consumer Module	G11	Refrigerated Storage by Consumer	=D11+F41*(F23-0.9*F23)
CPanel	E34	Temperature Consumer	=RiskTriang(J34,K34,L34,RiskName("Temp Consumer")) J34=-5 K34=3.44 L34=17
Consumer Module	I11	Serving Preparation Final Concentration	=G11
Consumer Module	F43	MPD (log CFU/g)	=3.65+0.18*(Temp) Temp= =CPanel!E34
Consumer Module	I38	Antimicrobial Scenario	=CPanel!E35 =0

Table 5.4 Distributions Used in the Cold-Smoked Salmon Quantitative Microbial Risk Assessment (QMRA) Model (Continued)

Module/Section **Worksheet (Tab)**	**Location (Cell)**	**Description or Step in the Process**	**Distribution** **Formula**
Consumer Module	I23	Serving Preparation Final Concentration	=RiskOutput("Consumer Log CFU Output")+IF(I11>F43,F43,I11)-I38
Dose-Response Module	B6	Serving Size (SS)	=RiskCumul(57,142,{75,136,142},{0.75,0.95,0.99})
Dose-Response Module	C6	Dose (output of consumer module)*SS	='Consumer Module'!$I$36*B6
Dose-Response Module	D6	Susceptible Population r-value (WHO/FAO 2004)	=0.00000000000106
Dose-Response Module	E6	$P=1-e^{-R*D}$	=RiskOutput(RiskConvergence())+1-EXP(-D6*C6)
Dose-Response Module	G6	Cases of Listeriosis per 1 million servings	=RiskOutput("Cases per Million Servings")+E6*1000000

Figure 5.1 Introductory worksheet for the quantitative risk assessment model for *L. monocytogenes* in cold-smoked salmon

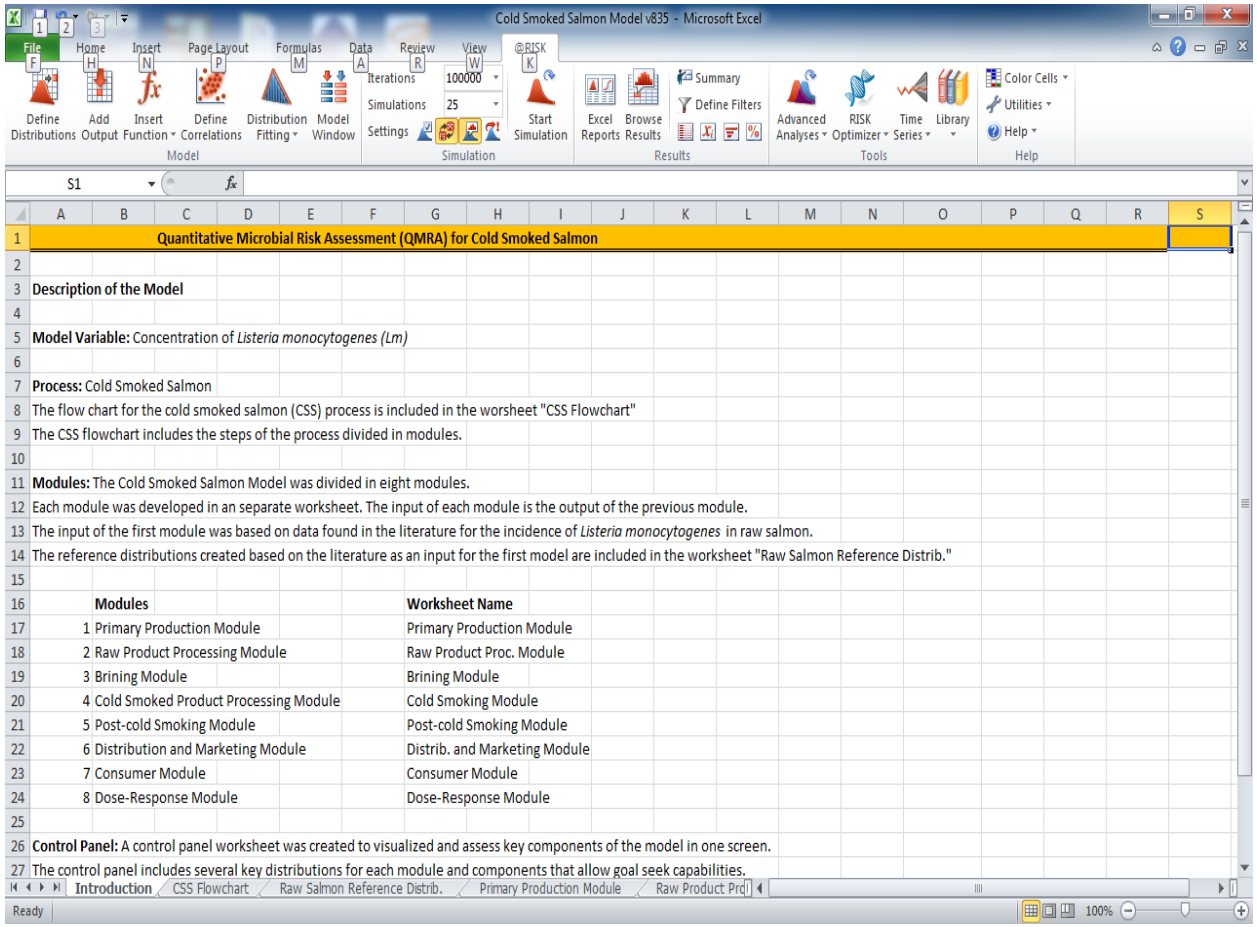


Figure 5.2 Baseline for the cold-smoked salmon MPPP model for *L. monocytogenes*

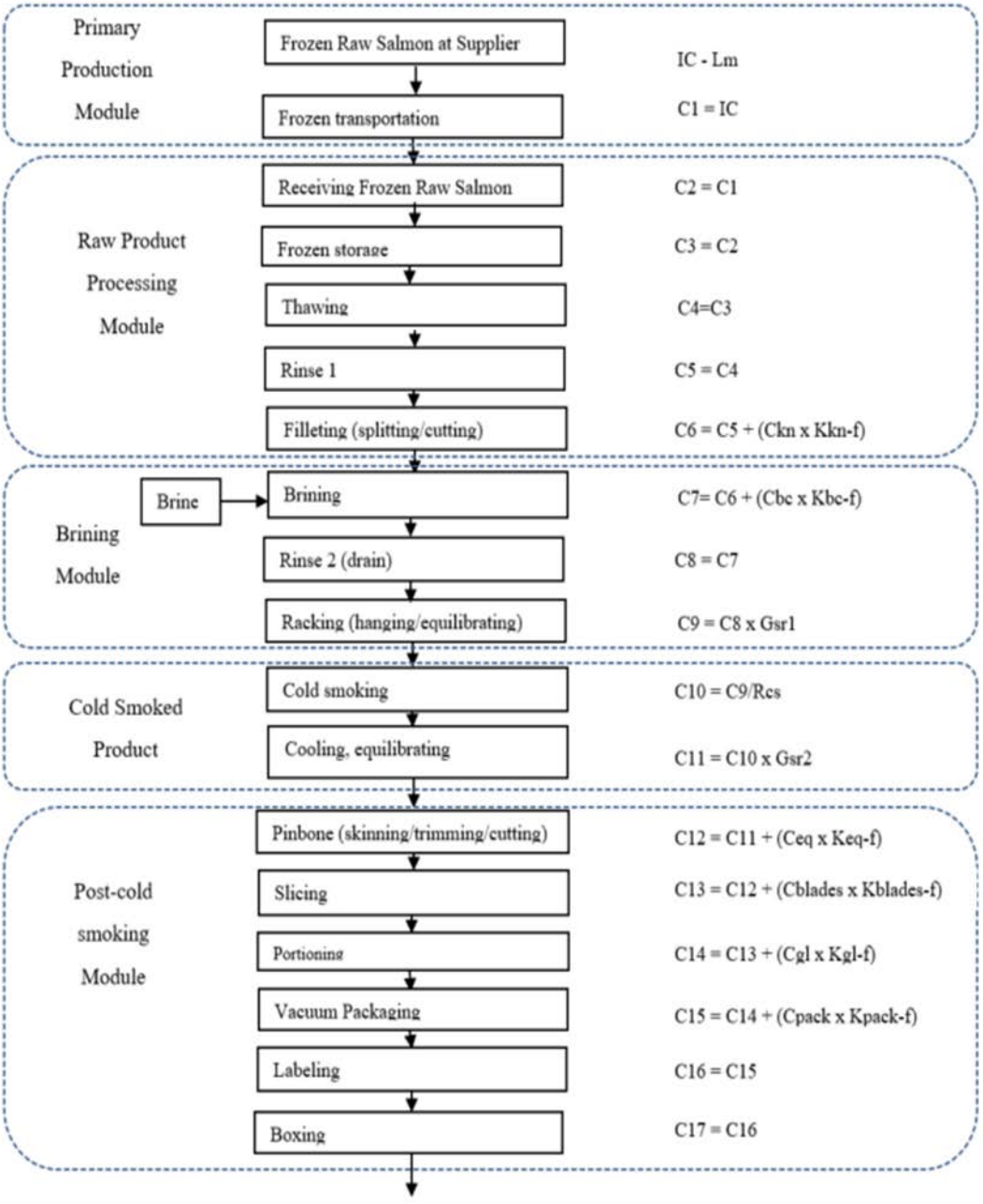


Figure 5.2 Continued

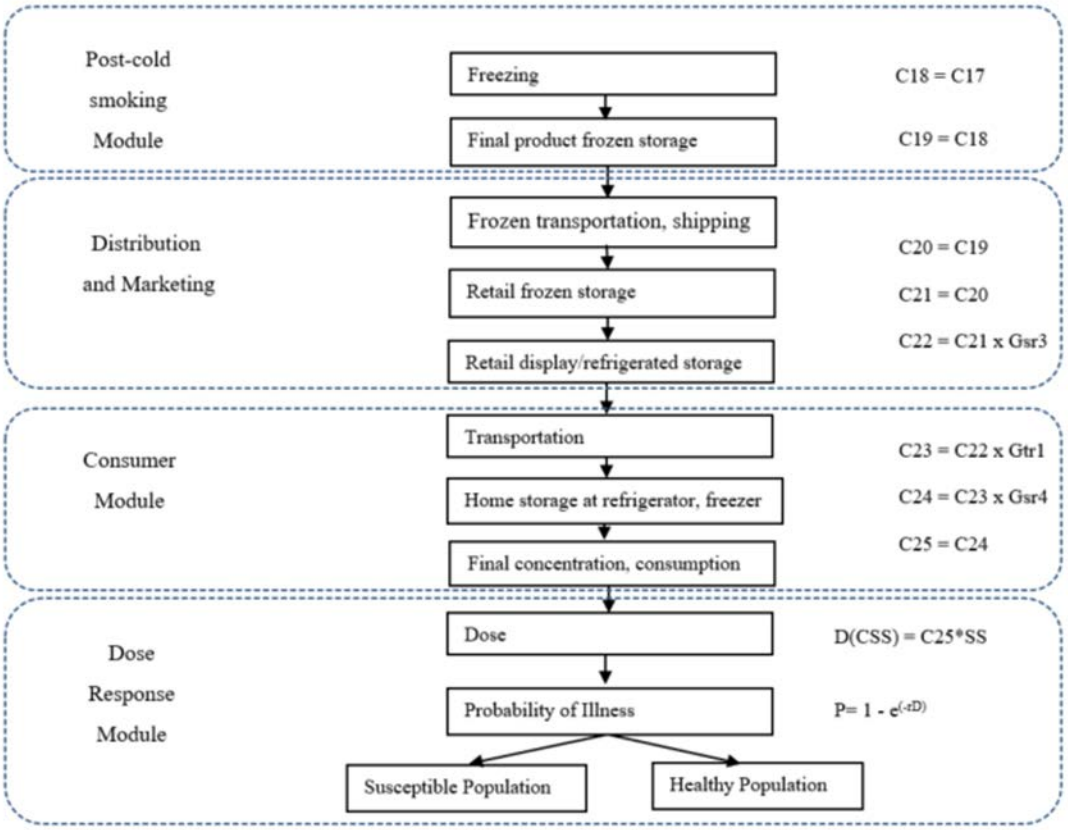


Figure 5.2 Continued

Where:

IC - *Lm*	Initial concentration of *Listeria monocytogenes* (*Lm*) in the product at primary production site
C1	Concentration of *Lm* in salmon during frozen transportation from primary production site to the processing facility
C2	Concentration of *Lm* in frozen salmon at reception step in the processing facility
C3	Concentration of *Lm* in frozen salmon after frozen storage step
C4	Concentration of *Lm* in frozen salmon after thawing step
C5	Concentration of *Lm* in thawed salmon after rinse 1 step
C6	Concentration of *Lm* in salmon after filleting step
C7	Concentration of *Lm* in salmon after brining step
C8	Concentration of *Lm* in salmon after rinse 2 step
C9	Concentration of *Lm* in salmon after racking/equilibrating step
C10	Concentration of *Lm* in salmon after cold-smoking step
C11	Concentration of *Lm* in salmon after cooling step
C12	Concentration of *Lm* in salmon after pinbone/trimming step
C13	Concentration of *Lm* in salmon after slicing step
C14	Concentration of *Lm* in salmon after portioning step
C15	Concentration of *Lm* in salmon after vacuum packaging step
C16	Concentration of *Lm* in salmon after labeling step
C17	Concentration of *Lm* in salmon after boxing step
C18	Concentration of *Lm* in salmon after freezing step
C19	Concentration of *Lm* in salmon after final product frozen storage step
C20	Concentration of *Lm* in salmon after frozen transportation from the facility to retail during distribution
C21	Concentration of *Lm* in salmon after retail frozen storage
C22	Concentration of *Lm* in salmon after retail display refrigeration
C23	Concentration of *Lm* in salmon after transportation by consumer
C24	Concentration of *Lm* in salmon after refrigeration by consumer
C25	Concentration of *Lm* in salmon immediately prior to consumption
C_{kn}	Concentration of *Lm* in knife at filleting step
C_{bc}	Concentration of *Lm* in the brine at brining step
C_{eq}	Concentration of *Lm* in the equipment at pinbone/trimming step
C_{blades}	Concentration of *Lm* in the blades at slicing step
C_{gl}	Concentration of *Lm* in gloves at portioning step
C_{pack}	Concentration of *Lm* in packaging at vacuum packaging step
K_{kn-f}	Transfer coefficient of *Lm* from knife to fish at filleting step
K_{bc-f}	Transfer coefficient of *Lm* from brine to fish at brining step
K_{eq-f}	Transfer coefficient of *Lm* from equipment to fish at pinbone/trimming step
$K_{blades-f}$	Transfer coefficient of *Lm* from blades to fish at slicing step
K_{gl-f}	Transfer coefficient of *Lm* from gloves to fish at portioning step
K_{pack-f}	Transfer coefficient of *Lm* from packaging to fish at vacuum packaging step
G_{sr1}	Potential growth of *Lm* at racking/equilibrating step
G_{sr2}	Potential growth of *Lm* at cooling step
G_{sr3}	Potential growth of *Lm* at retail display refrigeration
G_{sr4}	Potential growth of *Lm* during refrigeration by consumer
G_{tr1}	Potential growth of *Lm* during transportation by consumer
R_{cs}	Potential reduction of *Lm* concentration after cold-smoking step
D	Dose or number of ingested *Lm* (CFU/serving)
SS	Serving size
P	Probability of Listeriosis
r	Constant specific for *Lm* that helps define the shape of the dose-response curve

Figure 5.3 Cold-smoked salmon detailed flowchart worksheet including modules

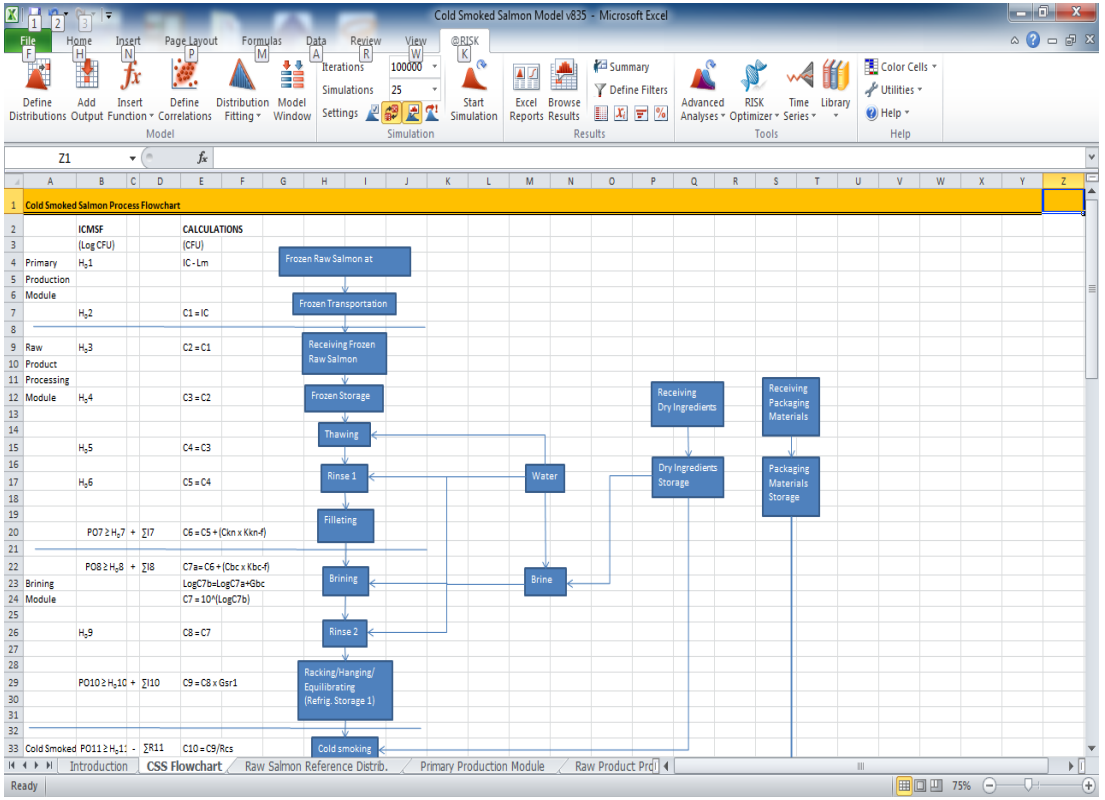


Figure 5.4 Primary production module worksheet

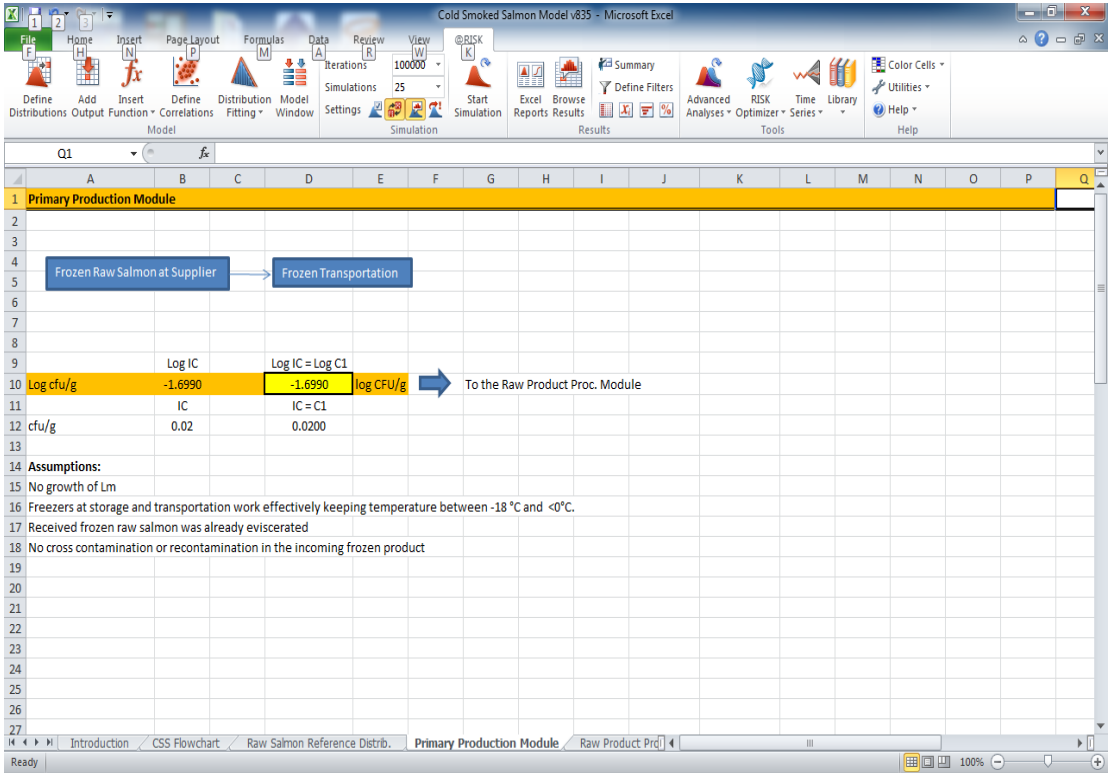


Figure 5.5 Raw salmon reference distribution worksheet for the concentration of *L. monocytogenes* at the primary production level

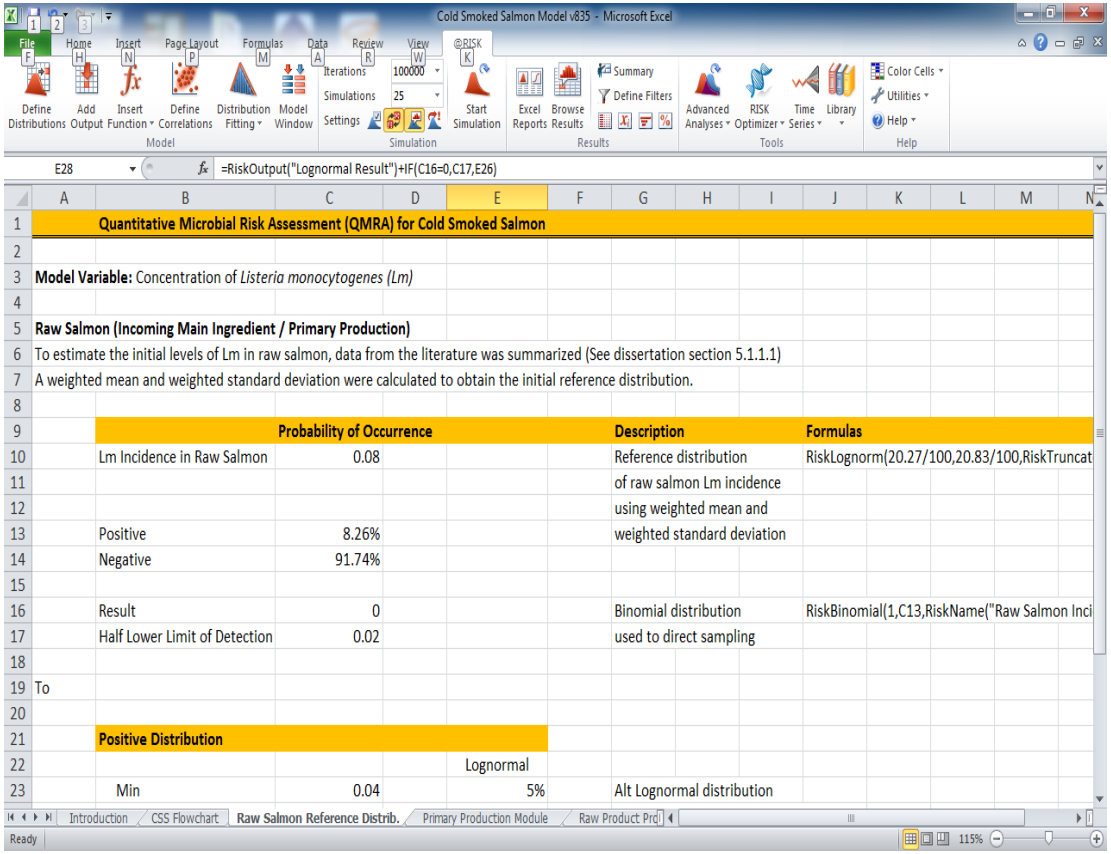


Figure 5.6 Concentration of *L. monocytogenes* in incoming raw salmon based on incidence and reference distributions

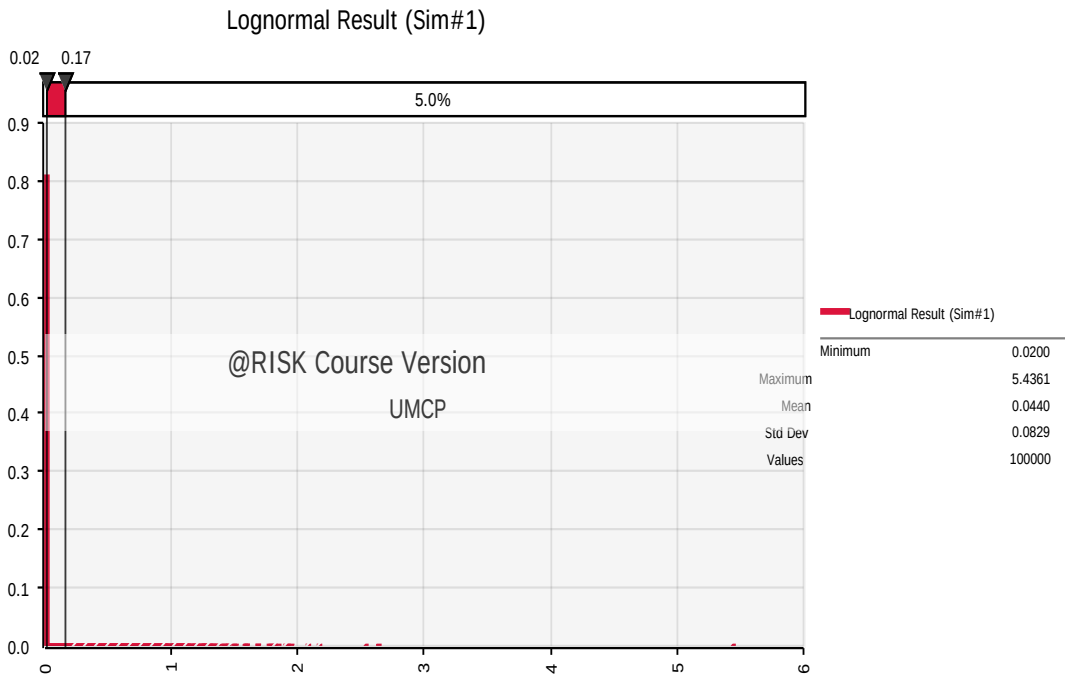


Figure 5.7 Raw product processing module worksheet

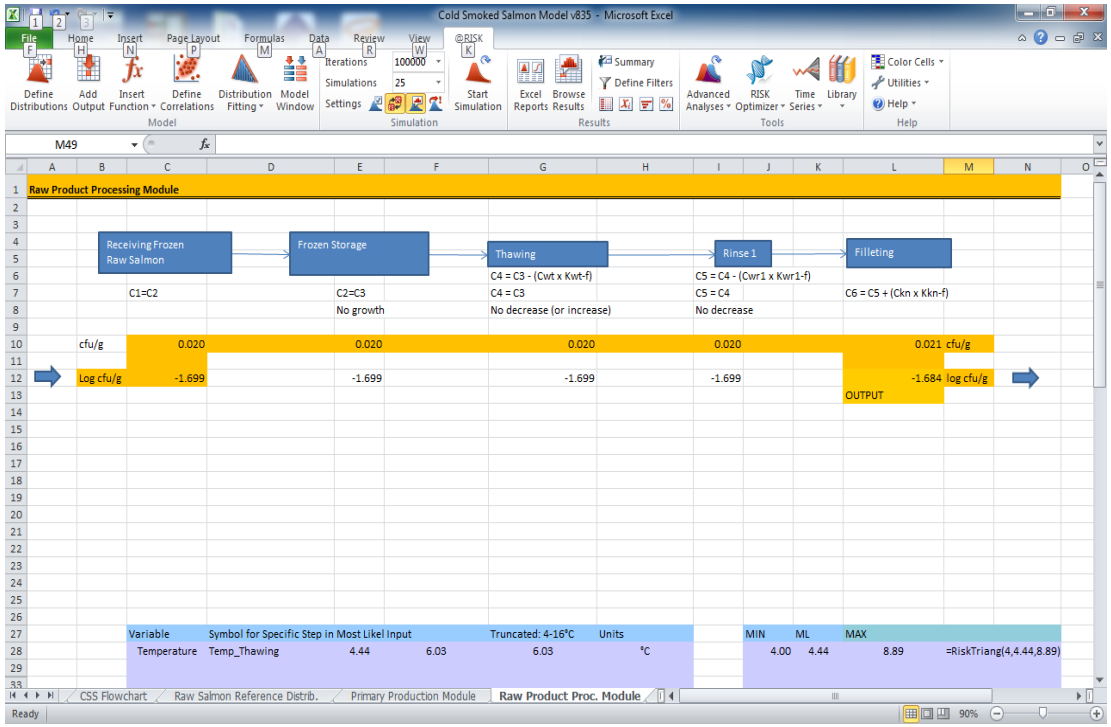


Figure 5.8 Brining module worksheet

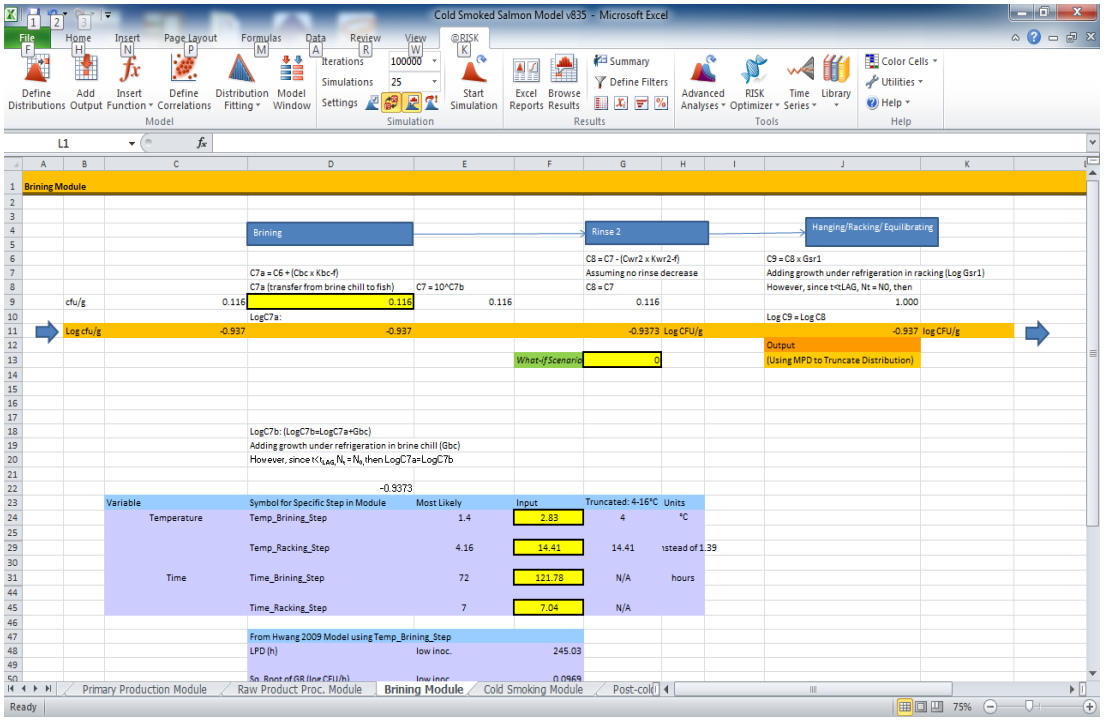


Figure 5.9 Cold-smoking processing module worksheet

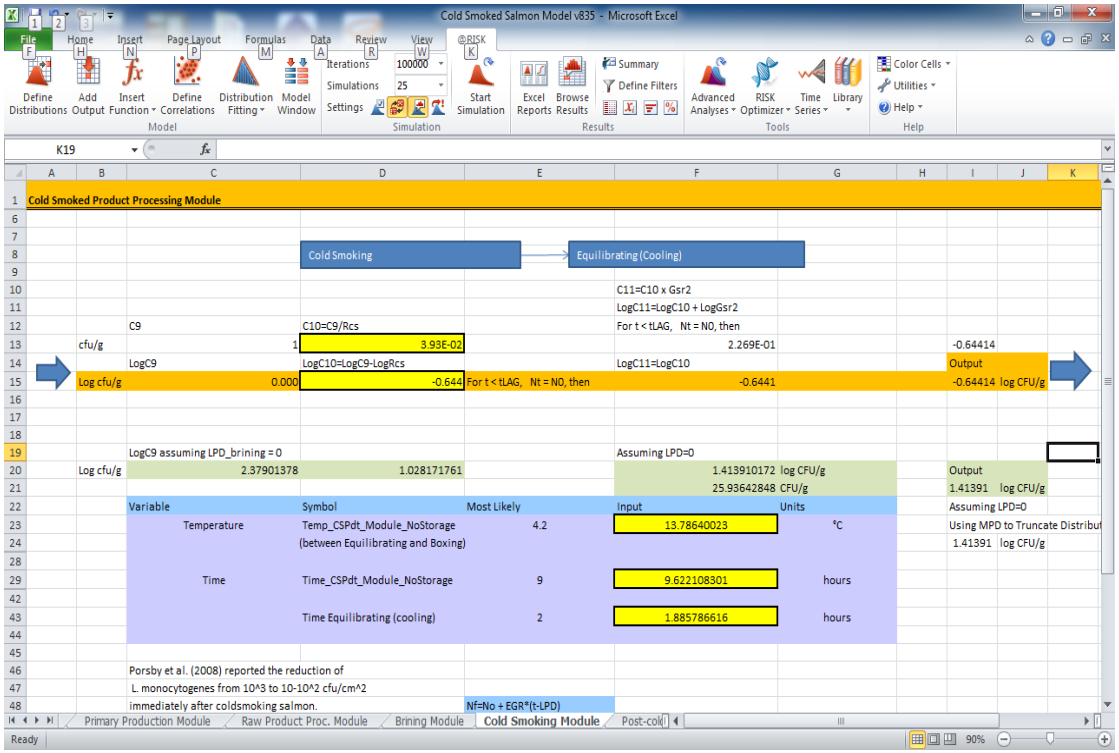


Figure 5.10 Post-cold smoking processing module worksheet

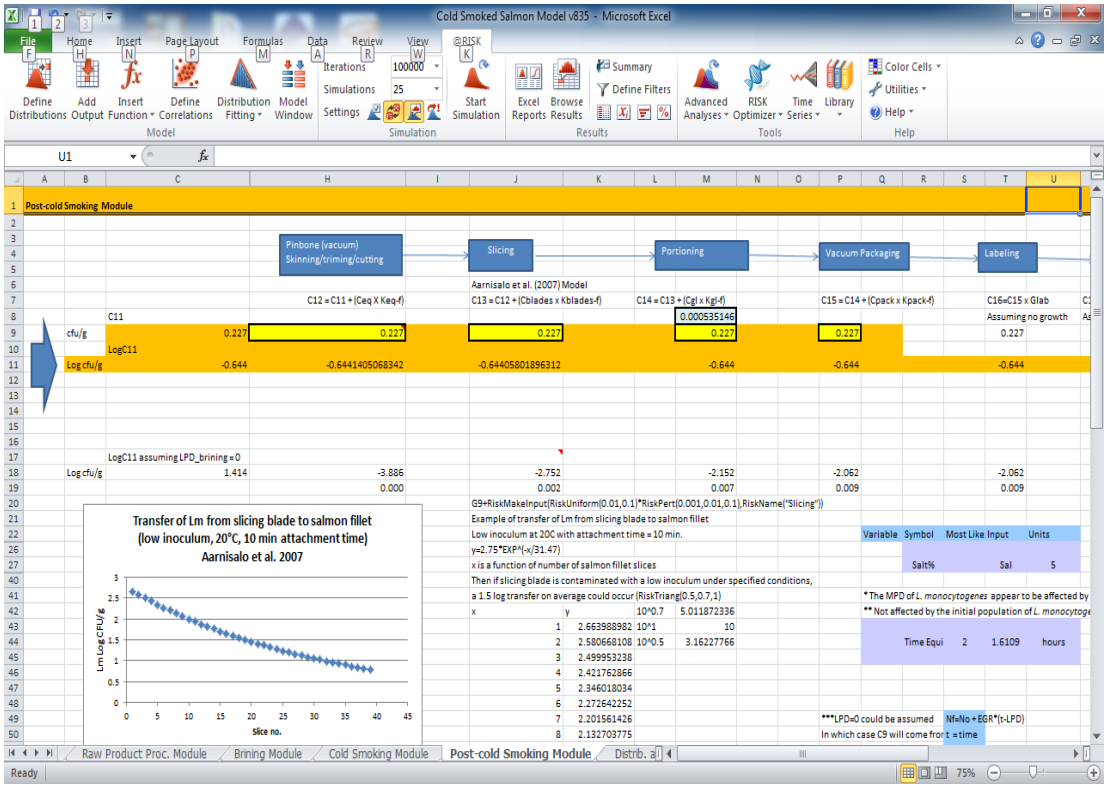


Figure 5.11 Distribution and marketing module worksheet

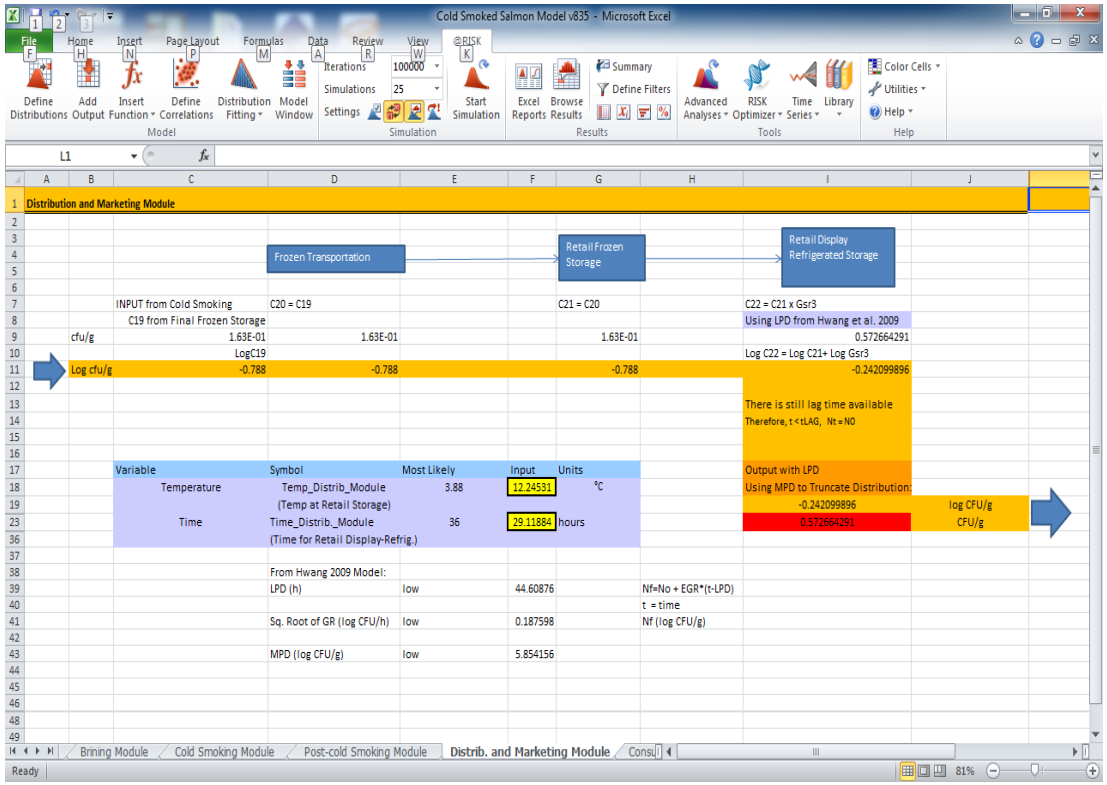


Figure 5.12 Consumer module worksheet

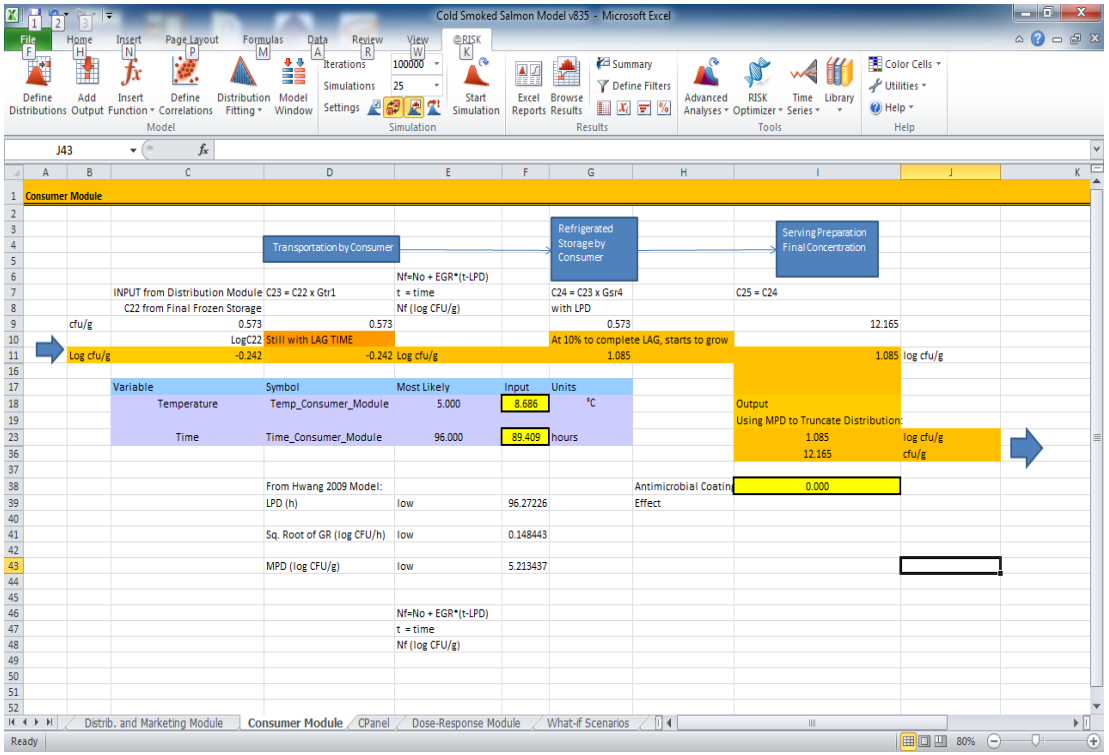


Figure 5.13 Dose-response module worksheet

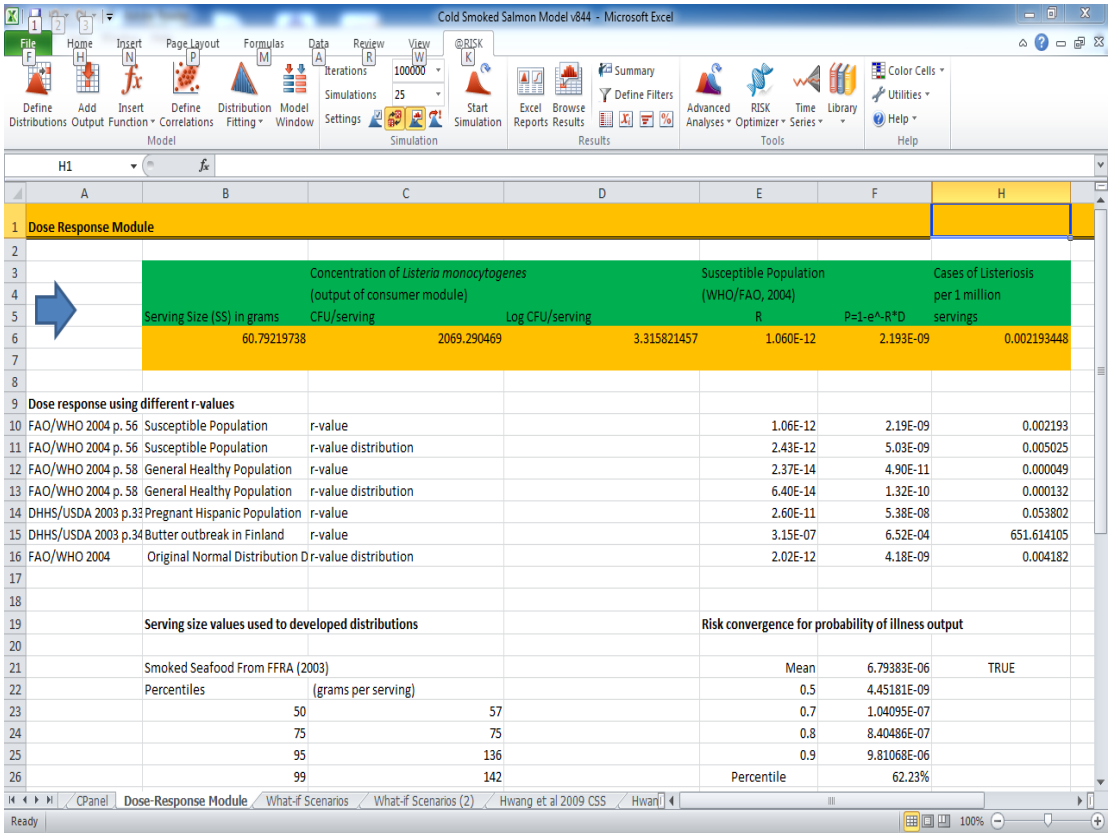
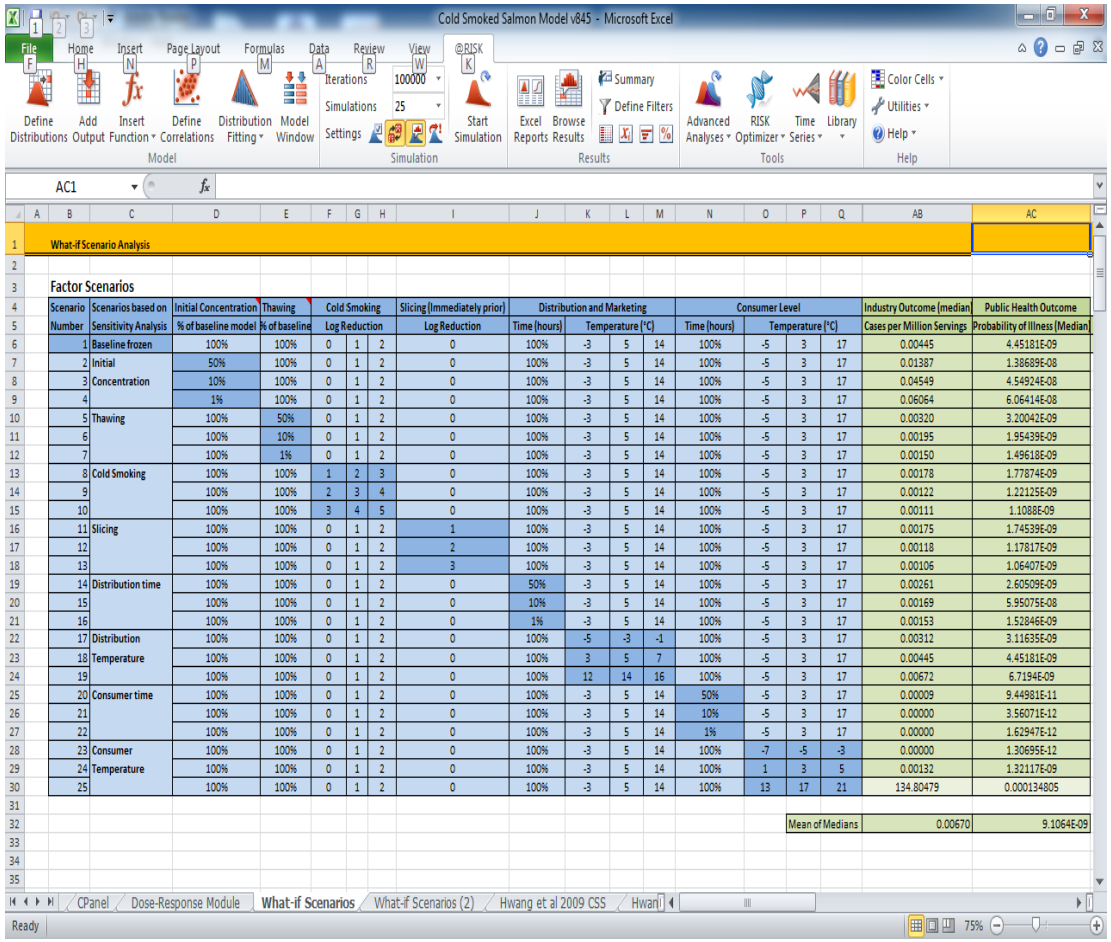


Figure 5.14 What-if scenario analysis worksheet



Scenario Number	Scenarios based on Sensitivity Analysis	Initial Concentration % of baseline model	Thawing % of baseline	Cold Smoking Log Reduction			Slicing (Immediately prior) Log Reduction	Distribution and Marketing Time (hours)				Consumer Level Time (hours)				Industry Outcome (median) Cases per Million Servings	Public Health Outcome Probability of Illness (Median)
									Temperature (°C)				Temperature (°C)				
1	Baseline frozen	100%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.00445	4.45181E-09
2	Initial	50%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.01387	1.38689E-08
3	Concentration	10%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.04549	4.54924E-08
4		1%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.06064	6.06414E-08
5	Thawing	100%	50%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.00320	3.20042E-09
6		100%	10%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.00195	1.95439E-09
7		100%	1%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	0.00150	1.49618E-09
8	Cold Smoking	100%	100%	1	2	3	0	100%	-3	5	14	100%	-5	3	17	0.00178	1.77874E-09
9		100%	100%	2	3	4	0	100%	-3	5	14	100%	-5	3	17	0.00122	1.22125E-09
10		100%	100%	3	4	5	0	100%	-3	5	14	100%	-5	3	17	0.00111	1.1088E-09
11	Slicing	100%	100%	0	1	2	1	100%	-3	5	14	100%	-5	3	17	0.00175	1.74539E-09
12		100%	100%	0	1	2	2	100%	-3	5	14	100%	-5	3	17	0.00118	1.17817E-09
13		100%	100%	0	1	2	3	100%	-3	5	14	100%	-5	3	17	0.00106	1.06407E-09
14	Distribution time	100%	100%	0	1	2	0	50%	-3	5	14	100%	-5	3	17	0.00261	2.60509E-09
15		100%	100%	0	1	2	0	10%	-3	5	14	100%	-5	3	17	0.00169	5.95075E-08
16		100%	100%	0	1	2	0	1%	-3	5	14	100%	-5	3	17	0.00153	1.52846E-09
17	Distribution	100%	100%	0	1	2	0	100%	-5	-3	-1	100%	-5	3	17	0.00312	3.11635E-09
18	Temperature	100%	100%	0	1	2	0	100%	3	5	7	100%	-5	3	17	0.00445	4.45181E-09
19		100%	100%	0	1	2	0	100%	12	14	16	100%	-5	3	17	0.00672	6.7194E-09
20	Consumer time	100%	100%	0	1	2	0	100%	-3	5	14	50%	-5	3	17	0.00009	9.44981E-11
21		100%	100%	0	1	2	0	100%	-3	5	14	10%	-5	3	17	0.00000	3.56071E-12
22		100%	100%	0	1	2	0	100%	-3	5	14	1%	-5	3	17	0.00000	1.62947E-12
23	Consumer	100%	100%	0	1	2	0	100%	-3	5	14	100%	-7	-5	-3	0.00000	1.30695E-12
24	Temperature	100%	100%	0	1	2	0	100%	-3	5	14	100%	1	3	5	0.00132	1.32117E-09
25		100%	100%	0	1	2	0	100%	-3	5	14	100%	13	17	21	134.80479	0.000134805

Figure 5.15 Factors with potential impact on the final level of *L. monocytogenes* at the consumer level

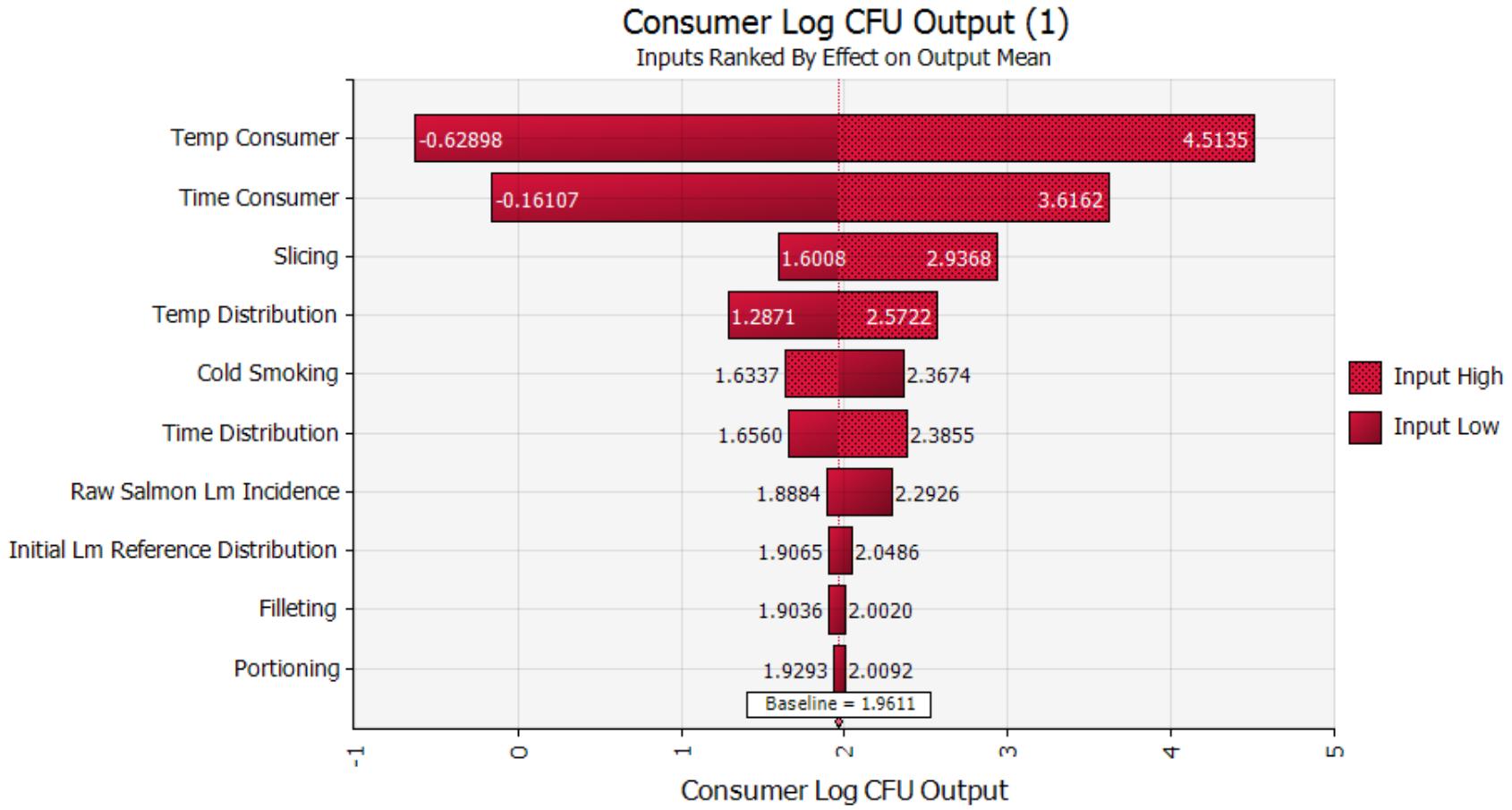


Figure 5.16 Impact of different cold-smoking what-if scenarios on the levels of *L. monocytogenes*

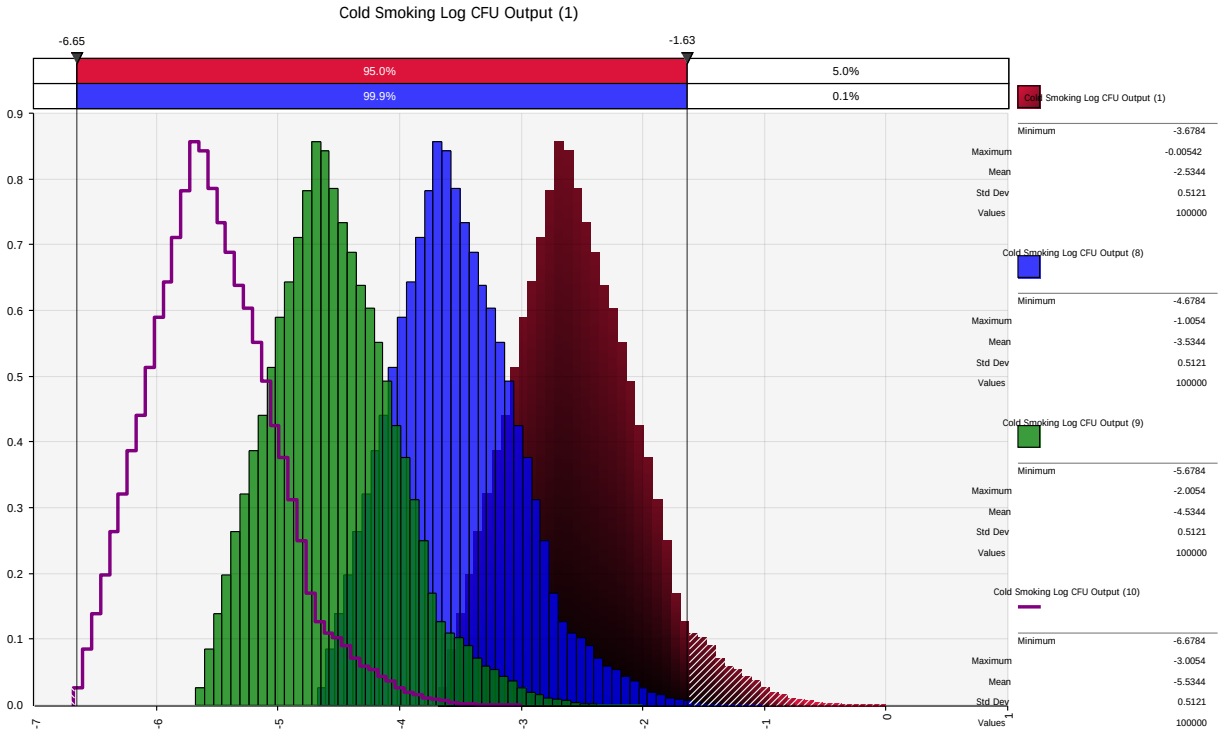
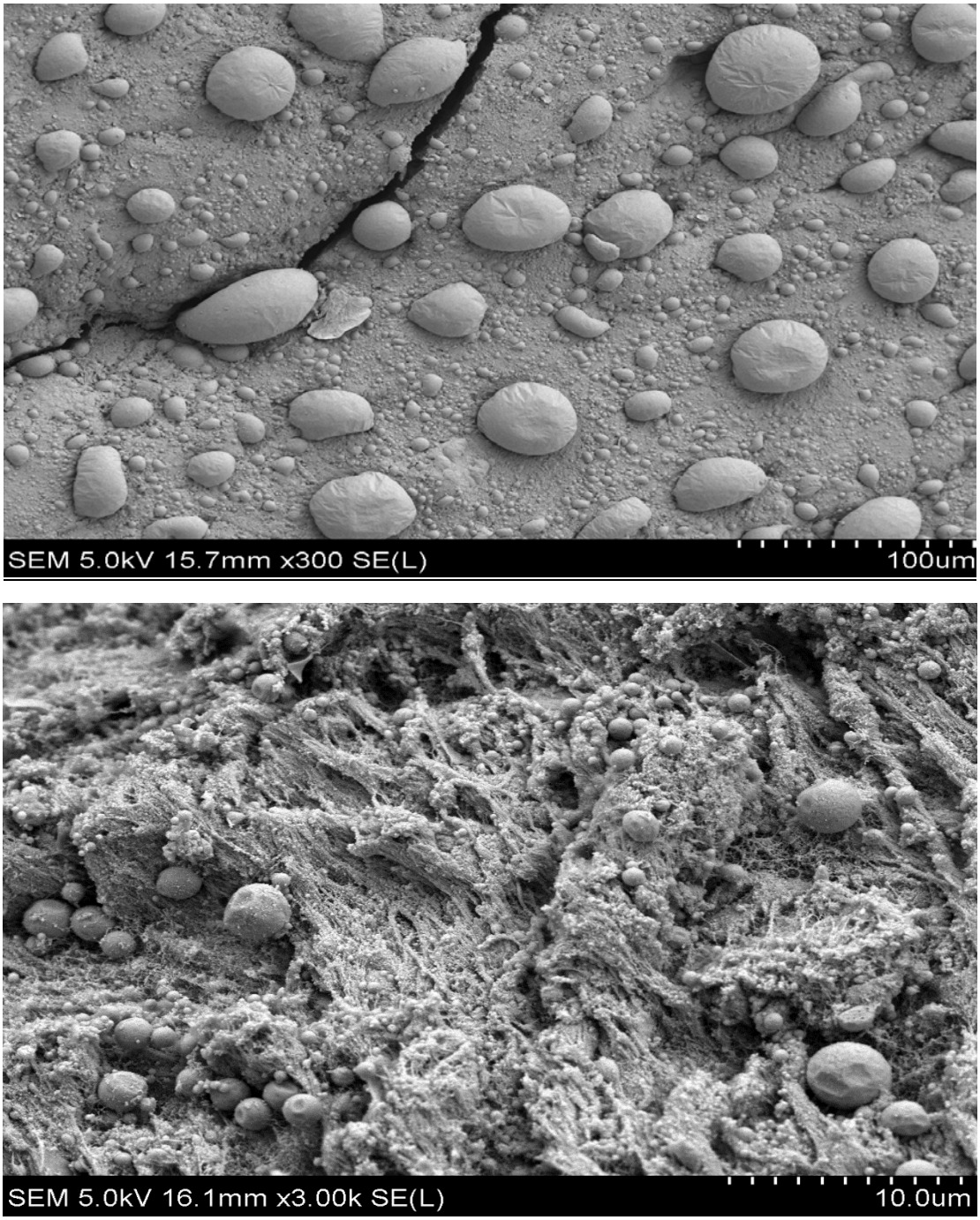


Figure 5.17 Scanning electron microscopy close-up images of surface regions of cold-smoked salmon showing fat droplets



Source: Williams 2011

Chapter 6: Risk-based Critical Control Points for *Listeria monocytogenes* on Frankfurters and in Cold-Smoked Salmon

Hazard Analysis Critical Control Point (HACCP) is a systematic approach to identify, evaluate, and control food safety hazards. Traditionally, HACCP plans are developed for one process line and a particular product, with plans entailing several hazards of various types that are identified and analyzed by conducting a hazard analysis (Wallace and others 2014). For the present research, however, we did not conduct a hazard analysis and deliberately focused on only one biological hazard, *Listeria monocytogenes,* in ready-to-eat (RTE) products. The two products selected were frankfurters and cold-smoked salmon (CSS), which encompassed the continuum of RTE products. Each step of the process and unit operation for the production of these RTE products was analyzed. Traditional HACCP plan forms were developed for comparison purposes, identifying the critical control points (CCPs) for both RTE products. CCPs are defined as steps in the process at which a food safety hazard can be eliminated, prevented, or reduced to acceptable levels. Although traditional CCPs were identified, the main focus of Chapter 6 was to present the risk-based CCPs for *L. monocytogenes* derived from the sensitivity and what-if scenario analyses results for both RTE products.

The graphic results of the sensitivity analysis (SA), as explained in Chapter 3, are tornado graphs, which in turn represent the most critical steps in the process. These steps are referred to as risk-based critical control points (RB-CCPs) throughout the present chapter and represent the steps in the process for which mitigation strategies could be most effective in regards to the level of *L. monocytogenes*, including controlling potential growth and thereby lowering the public health risk for listeriosis. The RB-CCPs were obtained from the

quantitative microbial risk assessment (QMRA) models for frankfurters and CSS previously presented in Chapters 4 and Chapter 5, respectively. The RB-CCPs for both products were presented on the tornado graphs in order of priority, from top to bottom. Thus, this chapter describes the most important factors affecting *L. monocytogenes* in the two selected RTE products. A detailed explanation of the resulting outputs of each module is offered, as the RB-CCPs were retrieved at the output level of each of the modules for both QMRA models.

Furthermore, what-if scenario analyses were developed and tornadoes retrieved at the consumer output level for both products. The description of the what-if scenario analyses developed from the baseline models and their results were included in the corresponding subsection of the present chapter for each of the RTE products. Although the approaches were different for each of the RTE products, they each contribute from different perspectives. In the case of frankfurters, the what-if scenarios considered reheated frankfurters in different subpopulations (e.g., perinatal, elderly). In the case of CSS, a total of 25 selected scenarios were developed changing one factor at the time throughout the process as it unfolded from primary production to consumption. Finally, a summary section compared key aspects of the case studies, noting scenarios that accomplished a low relative risk to public health for listeriosis in both RTE products and their corresponding RB-CCPs.

In brief, Chapter 6 was divided into four sections. Section 6.1 and 6.2 covered the SA and what-if scenario results for frankfurters and CSS, respectively. Section 6.3 consisted of a summary that included the RB-CCPs for frankfurters and CSS. The RB-CCPs were obtained after running all of the what-if scenarios in the QMRA models using @Risk 7.5. This version of @Risk includes “inputs ranked by effect of output mean” and “contribution to variance” tornadoes, among others. These two types of tornadoes were selected with the research hypothesis in mind. The results concerning the hypotheses were addressed in the concluding remarks of this chapter (section 6.4).

6.1 *Case Study One—Determination of Risk-based Critical Control Points for* Listeria monocytogenes *on Frankfurters*

Traditionally, HACCP plans for meat products and the critical control points (CCPs) derived from them are developed based on HACCP plan forms. For purposes of comparison, the present research developed a traditional HACCP plan form for frankfurters (see Table 6.1) based on the process line at the visited frankfurter plant, as well as on the literature, including generic HACCP plans (USDA 1999). The risk-based CCPs (RB-CCPs) for the frankfurter manufacturing process were identified using sensitivity analysis (SA) and are presented in section 6.1.1. In addition, the what-if scenario analyses results for frankfurters reheated at the consumer level are presented in section 6.1.2. This section includes the predicted median cases of listeriosis for the total United States population on a per-serving basis and the relative risk rankings of each scenario based on criteria by Carrington and others (2004).

6.1.1 Frankfurters Sensitivity Analysis Results

The risk-based critical control points (RB-CCPs) found at the end of each module for the frankfurters baseline model are presented in this section, including the tornadoes corresponding to the inputs ranked by effect on output mean and contribution to variance tornadoes (see Figures 6.1). It is noteworthy that the contribution to variance tornado graphs help show how much of the variance in the output variable is attributable to each individual input. These were the results of the sensitivity analysis (SA) at the output level of each module. As described in Chapter 4, the frankfurter process was divided into the following six modules: (1) ingredients, (2) raw product processing, (3) cooked product processing, (4) distribution and marketing, (5) consumer module, and (6) dose-response. The result sections for each of these modules at the baseline model level follow.

6.1.1.1 Frankfurters sensitivity analysis results—Ingredients module

The tornado graph results for the ingredients module at the output level, after the refrigerated transportation by the supplier and immediately before the raw beef pre-blend arrives at the processing facility, showed the initial level of *L. monocytogenes* ("cfu/g IC") in the incoming raw beef pre-blend as the main critical control point at this level. This refers to the incidence of *L. monocytogenes* resulting from any practices and conditions at which the supplier stored the raw beef pre-blend under refrigeration before receiving it at the facility. This was the only factor shown in the "contribution to variance" tornado. In addition to the initial level of *L. monocytogenes* ("cfu/g IC") in the incoming raw beef pre-blend, the "inputs ranked by effect on change in output mean" tornado also includes, as a minor factor, the duration of the transportation of the raw beef pre-blend ("Log cfu/g ttr1(h)") from the supplier to the facility. Thus, similar results were obtained for both types of tornadoes: the "inputs ranked by effect on output mean" and the "contribution to variance" tornadoes showing the initial level of *L. monocytogenes* ("cfu/g IC") in the incoming raw beef pre-blend as the main critical control point (see Figure 6.1.1). Therefore, at this level, it could be inferred that the refrigerated conditions prior to receiving the raw beef pre-blend were critical to minimizing potential levels of *L. monocytogenes*.

6.1.1.2 Frankfurters sensitivity analysis results—Raw product processing module

The tornado graph results for the raw product processing module at the output level, after cooking and immediately before chilling, indicated that the main critical control point was the temperature of cooking ("Tck1 (°C)") the frankfurters at the facility level. The second most important factor shown in the tornado graphs was the duration of cooking ("tck1") at the facility level. This remained true for both tornadoes, the "inputs ranked by effect on output mean" and the "contribution to variance" (see Figure 6.1.2). However, the

"inputs ranked by effect on output mean" tornado also showed the following minor factors: initial level of *L. monocytogenes* ("cfu/g IC"), duration of the refrigerated storage ("ts1"), and duration of the transportation ("Log cfu/g / ttr1 (h)") from the supplier to the facility.

6.1.1.3 Frankfurters sensitivity analysis results—Cooked product processing module

The sensitivity analysis results for the cooked product processing module at the output level, after final product storage and immediately before the transportation in the distribution module, showed the "handling at packaging" step of the process as the main critical control point at this level in both the "inputs ranked by effect on output mean" and the "contribution to variance" tornadoes (see Figure 6.1.3).

6.1.1.4 Frankfurters sensitivity analysis results—Distribution and marketing module

The output tornado graphs' results for the retail distribution and marketing module show the same main three factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.1.4). These three factors were "handling at packaging," "duration of transportation to retail," and "duration of refrigerated storage during retail." Furthermore, the "inputs ranked by effect on output mean" tornado included additional factors in the following order: "pH of unopened package at retail distribution, temperature of unopened package at retail distribution, nitrite concentration of unopened package at retail distribution, and salt concentration of unopened package at retail distribution."

6.1.1.5 Frankfurters sensitivity analysis results—Consumer module

The output tornado graph results for the consumer module of the baseline model show the same six factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.1.5). These six factors were "handling at packaging," "duration of refrigerated consumer storage," "duration of transportation to retail," "duration of transportation from retail to consumer," "duration of refrigerated storage during retail," and

"temperature of unopened package at consumer refrigerator." In addition, the "inputs ranked by effect on output mean" tornado included the following factors: "pH of unopened package at consumer refrigerator, pH of unopened package at retail distribution, temperature of unopened package at retail distribution, nitrite concentration of unopened package at retail distribution." It is noteworthy that the main six factors or RB-CCPs were the same for both types of tornadoes in the same ranking order for the frankfurters, not reheated (FNR) or baseline model.

6.1.1.6 Frankfurters sensitivity analysis results—Dose-response module

The results of the output tornado graphs for the dose-response module of the baseline model show the same main seven factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.1.6). These seven factors from top to bottom were "handling at packaging," "duration of refrigerated consumer storage," "serving size," "duration of transportation to retail," "duration of transportation from retail to consumer," "duration of refrigerated storage during retail," and "temperature of unopened package at consumer refrigerator." In addition, the "inputs ranked by effect on output mean" tornado included the following factors: "pH of unopened package at consumer refrigerator, pH of unopened package at retail distribution, salt concentration of unopened package at consumer refrigerator." It is noteworthy that the main seven factors or RB-CCPs were the same for both types of tornadoes in the same ranking order for the frankfurters not reheated (FNR) or baseline model.

6.1.2 Frankfurters What-if Scenario Analysis Results

In the case of frankfurters, a ready-to-eat product, the main scenario, in addition to the baseline model, included the mitigation strategy of reheating frankfurters at the consumer level to reduce the risk of *L. monocytogenes* immediately prior to consumption. In addition, the results for what-if scenarios considering other specific susceptible subpopulations (i.e., elderly, perinatal) were included and summarized in Table 6.2. Another mitigation strategy

found in the literature was the reformulation of frankfurters (Carrington and others 2004). Although reformulation of frankfurters was not considered in the present study, it should be considered in future work.

The results for the frankfurters, not reheated (FNR) baseline model for the susceptible population based on r-values from FAO/WHO (2004) were $4.18x10^{-9}$ median cases of listeriosis per serving. These results differ by nearly a log cycle compared to the "Frankfurters, not reheated" category obtained by Carrington and others (2004) of $6.5x10^{-8}$ median cases per serving (see Table 6.2). Since Carrington and others considered >5 cases per billion servings as the cut-off for relative high risk, the predicted median cases of listeriosis for the predicted values in the present study corresponded to a moderate risk compared to the "Frankfurters, not reheated" category developed by Carrington and others, which falls under the relative high-risk category for listeriosis on a per-serving basis. The predicted median cases of listeriosis for the elderly and perinatal subpopulations fall under the high-risk category as described by Carrington and others with $3.31x10^{-8}$ and $1.77x10^{-7}$ cases per serving.

The results for the frankfurter, reheated (FR) what-if scenario accomplished a "low risk" category based on the criteria by Carrington and others (2004) for all the selected subpopulations. Thus, the reheating frankfurter scenario represents a mitigation strategy at the consumer level thereby accomplishing effectively lower public health risk for listeriosis. This mitigation strategy at the consumer level consists of reheating the frankfurter, achieving a median of 4.49 log reduction prior to consumption, as described in Table 6.3.

The sensitivity analysis results for the baseline model, frankfurters non-reheated (FNR), and the frankfurter reheated (FR) what-if scenarios at the consumer output level were illustrated in Figures 6.1.5 and 6.1.7, respectively. The risk-based CCPs obtained for the

baseline model and the frankfurter reheated (FR) scenario at the consumer output level were reported and summarized in Table 6.4.

6.1.2.1 Frankfurters reheated—Consumer module

The tornado graph results for the what-if scenario of reheating frankfurters (FR) at the output level of the consumer module showed the same seven factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.1.7). These seven factors were "reheating log reduction," "handling at packaging," "duration of refrigerated consumer storage," "duration of transportation to retail," "duration of transportation from retail to consumer," "duration of refrigerated storage during retail," and "temperature of opened package at consumer refrigerator." In addition, the "inputs ranked by effect on output mean" tornado included the following factors: "temperature of unopened package at retail distribution," "nitrite concentration of opened package at consumer refrigerator," and "pH of unopened package at retail distribution." It is noteworthy that the main seven factors were the same for both tornadoes. The extent of the frankfurter reheating log reduction was represented by a cumulative distribution (see Table 6.3) created for this module based on data by FFRA (2003). A summary including all of the RB-CCPs found at the consumer level for the two scenarios was included in Table 6.4. These RB-CCPs were ranked to create a heat chart where the red color represented the "priority" RB-CCPs found at the top of the tornadoes. Although this is not the classification of CCP1 and CCP2 proposed by ICMSF Book 4 (1988), differentiating or ranking CCPs in order of priority could be beneficial to better understand many HACCP plans and future adaptations of them.

6.1.2.2 Frankfurters reheated—Dose-response module

The tornado graph results for the what-if scenario of reheating frankfurters (FR) at the output level of the dose-response module showed the same main eight factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure

6.1.8). These eight factors were “reheating log reduction,” “handling at packaging,” “duration of refrigerated consumer storage,” “serving size,” “duration of transportation to retail,” “duration of transportation from retail to consumer,” “duration of refrigerated storage during retail,” and “temperature of opened package at consumer refrigerator.” In addition, the “inputs ranked by effect on output mean” tornado included the “temperature of unopened package at retail distribution,” and the “pH of unopened package at retail distribution.” Figure 6.1.9 depicts the what-if scenario analysis results comparing the probability of listeriosis for the baseline scenario of frankfurters not reheated (FNR) versus the frankfurters reheated (FR) and showed the log reduction accomplished by this mitigation strategy.

6.2 *Case Study Two—Determination of Risk-based Critical Control Points for* Listeria monocytogenes *in Cold-Smoked Salmon*

Traditionally, Critical Control Points (CCPs) in HACCP plans for seafood products in the United States are developed based on the “Fish and Fishery Products Hazards and Controls Guidance” (FDA 2011) and the “Hazard Analysis and Critical Control Point Training Curriculum” (National Seafood HACCP Alliance 2011). A traditional HACCP plan form for cold-smoked salmon (CSS) was developed (see Table 6.5) based on these documents covered during the Segment 2 session of the Seafood HACCP course from the Seafood HACCP Alliance (SHA) training protocol. In like manner, a hazard analysis form was developed for this product, yet it was not included because the only pathogen of concern for the present risk-based HACCP research was *L. monocytogenes*. The risk-based CCPs (RB-CCPs) for the CSS process were identified using sensitivity analysis (SA) and presented in section 6.2.1. In addition, the what-if scenario analyses results for the 25 scenarios were presented in section 6.2.2 and included the predicted median cases of listeriosis for the total United States population on a per-serving basis and relative risk rankings of each scenario based on criteria by Carrington and others (2004).

6.2.1 Cold-Smoked Salmon Sensitivity Analysis Results

The risk-based critical control points (RB-CCPs) found at the end of each module for the cold-smoked salmon (CSS) baseline model were presented in this section including the tornadoes corresponding to the inputs ranked by effect on output mean and the contribution to variance (see Figures 6.2). These were the results of the sensitivity analysis (SA) at the output level of each module. As described in Chapter 5, the CSS process was divided into the following eight modules: (1) primary production, (2) raw product processing, (3) brining, (4) cold smoking, (5) post-cold smoking, (6) distribution and marketing, (7) consumer module, and (8) dose-response. The result sections for each of these modules at the baseline model level follow.

6.2.1.1 CSS Sensitivity analysis results—Primary production module

The tornado graph results for the primary production module at the output level after the frozen transportation and immediately before the product arrives at the processing facility showed the "raw salmon *L. monocytogenes* incidence" as the main critical control point at this level. This refers to the final incidence of *L. monocytogenes* resulting not only from the primary production practices (e.g., fishing) but also from the conditions at which the supplier stored the raw salmon (if storage occurred) prior to freezing, the conditions during the actual freezing of the raw salmon, and the storage and transportation conditions after freezing the raw salmon. The only other factor shown in the tornado graphs at this level was the "initial *L. monocytogenes* reference distribution," which refers to the levels of *L. monocytogenes* in the incoming product. Similar results were obtained for both the "inputs ranked by effect on output mean" and the "contribution to variance" tornadoes (see Figure 6.2.2). Although the raw salmon arrives frozen in the baseline model, the conditions of the supply chain at each step stated above could be considered key to receive salmon without *L. monocytogenes* or with the

lowest possible levels of this pathogen.

6.2.1.2 CSS sensitivity analysis results—Raw product processing module

The tornado graph results for the raw product processing module at the output level after filleting and immediately before brining showed the "raw salmon *L. monocytogenes* incidence" as the main critical control point. The other two factors shown in the tornado graphs were the "initial *L. monocytogenes* reference distribution" and "filleting." This holds true for both the "inputs ranked by effect on output mean" and the "contribution to variance" tornadoes (see Figure 6.2.2). Following the description of the cold-smoked salmon process from Chapter 5, no recontamination of the raw salmon was assumed in the first part of this module until the filleting step of the process, which consists of splitting and cutting raw salmon with a knife. Thus, the critical points in the previous module were carried over to this module with the addition of the filleting step.

6.2.1.3 CSS sensitivity analysis results—Brining module

The tornado graph results for the brining module at the output level after racking and immediately before cold smoking showed the "raw salmon *L. monocytogenes* incidence" as the main critical control point at this level. Factors also included in both tornado graphs were the "initial *L. monocytogenes* reference distribution" and "filleting," which were carried over to this module from the previous one. In addition, the "brining" step of the process appeared in both tornadoes. Finally, "temperature brining" was the last factor showing in the "inputs ranked by effect on output mean" tornado, although it was not present in the "contribution to variance" tornado (see Figure 6.2.3).

6.2.1.4 CSS sensitivity analysis results—Cold smoking processing module

The sensitivity analysis results for the cold-smoked product processing module at the output level after cooling/equilibrating and immediately before pinbone showed the "cold smoking" step of the process as the main critical control point at this level in both tornadoes.

The other factors shown in the tornado "inputs ranked by effect on output mean" were, in the following order: "raw salmon *L. monocytogenes* incidence, initial *L. monocytogenes* reference distribution, filleting, brining, temperature brining, and temperature after cold smoking." The "contribution to variance" tornado for the brining module showed similar results (see Figure 6.2.4) but without the last three factors mentioned above for the "inputs ranked by effect on output mean" tornado.

6.2.1.5 CSS sensitivity analysis results—Post-cold smoking processing module

The tornado graph results for the post-cold smoking processing module at the output level after final product frozen storage and immediately before the frozen transportation or shipping at the distribution and marketing module level show, for the most part, the same main factors for both tornadoes, but in different order of priority. In the case of the "inputs ranked by effect on output mean" tornado, the "slicing" step of the process was the main critical control point, followed by "cold smoking, raw salmon *L. monocytogenes* incidence, initial *L. monocytogenes* reference distribution, portioning, pinbone, brining, temperature brining, vacuum packaging, and filleting." On the other hand, in the case of the "contribution to variance" tornado, "cold smoking" was the main critical control point, followed by "raw salmon *L. monocytogenes* incidence, slicing, initial *L. monocytogenes* reference distribution, pinbone, and portioning." It is noteworthy that the main three factors were the same for both tornadoes (see Figure 6.2.5).

6.2.1.6 CSS sensitivity analysis results—Distribution and marketing module

The tornado graph results for the distribution and marketing module at the output level after retail display/refrigerated storage and immediately before transportation to the consumer showed the same main three factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.2.6). These three factors were "slicing, temperature distribution, and time distribution." Furthermore, the "inputs ranked by effect on

output mean" tornado included additional factors in the following order: "brining," "portioning," "initial *L. monocytogenes* reference distribution," "temperature brining," "vacuum packaging," "cold smoking," and "filleting."

6.2.1.7 CSS sensitivity analysis results—Consumer module

The tornado graph results for the consumer module at the output level after consumption of the cold-smoked salmon showed the same main two factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes (see Figure 6.2.7). These two factors were "temperature consumer" followed by "time consumer." These factors refer to the temperature and time of the storage under freezing or refrigerating conditions by the consumer. In addition, the "inputs ranked by effect on output mean" tornado included the following factors: "slicing, temperature distribution, cold smoking, time distribution, raw salmon *L. monocytogenes* incidence, initial *L. monocytogenes* reference distribution, filleting, and portioning." The "contribution to variance" tornado included the additional following factors: "temperature distribution, cold smoking, time distribution, raw salmon *L. monocytogenes* incidence, and slicing." It is noteworthy that the main two factors were the same for both tornadoes in the same order.

6.2.1.8 CSS sensitivity analysis results—Dose-response module

The tornado graph results for the dose-response module at the output level in this module showed the same main four factors for both the "inputs ranked by effect on output mean" and "contribution to variance" tornadoes. These four factors were "temperature consumer," followed by "time consumer, serving size, and temperature distribution." The first two factors refer to the temperature and time of the storage under freezing or refrigerating conditions by the consumer, whereas the "temperature distribution" refers to the temperature at which the product was stored at the distribution and marketing level. In addition, the "inputs ranked by effect on output mean" tornado included the following factors: "slicing, time

distribution, cold smoking, filleting, initial *L. monocytogenes* reference distribution, and temperature brining." It is noteworthy that the main four factors were the same for both tornadoes in the same exact order (see Figure 6.2.8).

6.2.2 Cold-Smoked Salmon What-if Scenario Analyses Results

The cold-smoked salmon (CSS) scenarios were selected based on the sensitivity analysis results from the baseline model and expert recommendations. The specific changes to create the scenarios were based on expert elicitation recommendations. These changes were summarized within the model in the "what-if scenarios" worksheet. Furthermore, Chapter 6 summarized all the CSS what-if scenarios in Table 6.6. This table showed a total of 25 different scenarios for CSS. The baseline model corresponds to the first scenario and the other 24 were developed by modifying one factor at a time based on expert recommendations. The predicted results for the CSS baseline model show 4.46×10^{-9} median cases of listeriosis per serving. These results were similar to the "smoked seafood" category obtained by Carrington and others (2004) of 6.2×10^{-9} median cases of listeriosis per serving. At the same time, Carrington and others considered >5 cases per billion servings as the cut-off for high-risk classification. Thus, the predicted median cases of listeriosis for CSS in the present research corresponded to a moderate risk compared to the "smoked seafood" category developed by Carrington and others (2004), which falls under the relative high-risk category for listeriosis. In addition, Table 6.6 showed the results for the predicted median cases of listeriosis per serving for each scenario and their relative risk ranking classification based on criteria by Carrington and others (2004). Four of the CSS scenarios accomplished a "low risk" category based on these criteria. These specific scenarios represent mitigation strategies at the consumer level by accomplishing lower public health risk for listeriosis. These mitigation strategies at the consumer level consist of reducing the storage time or temperature of the CSS at certain levels prior to consumption. For example, scenario 20 represents cutting

in half the storage duration of CSS in the consumer refrigerator, scenario 21 represents reducing by ten times the storage duration (to 10%) in the consumer refrigerator, scenario 22 represents lowering the storage duration to 1% or, in other words, consuming the CSS almost upon arrival in the consumer refrigerator, minimizing the time for *L. monocytogenes* growth. Scenario 23 represents freezing the product upon arrival with a most likely temperature of -5°C. The sensitivity analysis results for the baseline model at the consumer output level (Figure 6.2.7) was compared with equivalent results for what-if scenarios 20, 21, 22, 23 (Figures 6.2.9, 6.2.10, 6.2.11, and 6.2.12, respectively). The CSS case study results summary, including the RB-CCPs found at the consumer level for the baseline model and the selected four "low risk" what-if scenarios, were presented in Table 6.7. These and other results, not only for CSS but also for frankfurters, are compared and discussed in section 6.3.

6.3 *Summary—Determination of Risk-based Critical Control Points*

Although the modeling approaches and the selection of what-if scenarios were different for the frankfurters and cold-smoked salmon processes, these two QMRA models contributed data from different perspectives. In the case of frankfurters, the what-if scenarios considered not only reheating the frankfurters at the consumer level but also the public health outcome for three different subpopulations (i.e., susceptible, perinatal, and elderly). Whereas, in the case of CSS, a total of 25 selected scenarios were developed, changing one factor at the time throughout the process from primary production to consumption (e.g., consumer storage duration and temperature, retail distribution duration and temperature, cold smoking). This summary section aims at pointing out the scenarios that accomplished a low relative risk to public health for listeriosis for both RTE products and the RB-CCP associated with them.

In the case of frankfurters, reheating the product (FR) at the consumer level was found to be an effective mitigation strategy that achieved low risk for public health for listeriosis in all three categories—susceptible, elderly and perinatal subpopulations. The top-

ranked RB-CCPs for this scenario was the “reheating log reduction.” This RB-CCP was followed by “handling at packaging, duration of refrigerated consumer storage, and duration of transportation to retail distribution,” which obtained similar rankings for the baseline model scenario of frankfurters not reheated (FNR). A complete picture of the RB-CCPs was depicted in the frankfurters SA summary (Table 6.4) where the red color represents the top-ranked or “priority” RB-CCPs.

In the case of cold-smoked salmon (CSS), the results of the what-if scenarios showed that four of the what-if scenarios (i.e., scenario 20, scenario 21, scenario 22, and scenario 23) accomplished a “low risk” classification for listeriosis based on the public health outcome of predicted median cases of listeriosis on a per-serving basis as proposed by Carrington and others (2004). Scenarios 20, 21, and 22 refer to reducing the duration of the consumer storage to 50%, 10%, and 1%, whereas scenario 23 refers to freezing the CSS upon arrival at the consumer level. Although each of these scenarios accomplished a low risk for listeriosis, the outcome of the tornadoes and thus the RB-CCPs for them were slightly different depending on which factors were modified. For example, reducing the duration of the storage time to 50% (Scenario 20) was not enough to eliminate the temperature and duration of the storage RB-CCPs at the consumer level. However, reducing the storage duration to 10% or 1% (Scenarios 21 and 22, respectively), eliminated the duration and temperature of the storage at the consumer level as top-ranked RB-CCPs. Thus, even in the same process, different scenarios could have a slightly different ranking or “priority” of RB-CCPs depending on the conditions. The RB-CCPs were obtained from the SA for each scenario which demonstrated the potential to lower the risk of listeriosis. A summary of the RB-CCPs for CSS were consolidated in Table 6.7. For example, the baseline scenario and scenario 20 shared the following RB-CCPs: temperature of consumer storage, duration of consumer storage, slicing, temperature of distribution, cold smoking. Scenarios 21 and 22 shared the following top RB-

CCPs: temperature and time of retail distribution, slicing, and cold smoking. Scenario 23 refers to freezing the CSS, and although this results in eliminating the duration of the storage at the consumer level, the temperature at which the consumer stores the CSS was still an important top-ranked RB-CCP. Other CCPs for this scenario were temperature and time of distribution, slicing, and cold smoking. The red color in the SA summary for CSS (Table 6.7) represents the top-ranked or "priority" RB-CCPs, the yellow color represents a medium priority level RB-CCPs and the green color represents a low-priority level RB-CCP depending on each scenario and type of tornado.

6.4 *Concluding Remarks*

Although different conditions in the consumer module were key to lowering the risk of listeriosis in these two RTE products, it is important to emphasize that RB-CCPs were unique for each processing line. Furthermore, in some cases RB-CCPs for specific scenarios had variations in their ranking order. Thus, the RB-CCPs for RTE product categories cannot be generalized as each processing line and RTE product has specific unit operations and variability conditions. In this regard, the working hypothesis for the present research stated that CCPs are steps in the process that significantly reduce the mean and/or variance of a hazard (see Chapter 2 section 2.3). Following the logic of the present research, the ability to find the most critical or top-ranked RB-CCPs would depend on the criteria used to determine low relative risk for listeriosis. In this research, the criteria used were drawn from Carrington and others (2004). Thus, the low risk category was defined as <1 case per billion servings. Therefore, the scenarios that accomplished this criterion were the ones selected as "priority" RB-CCPs. However, scenarios that accomplished lower levels could also be defined as CCPs.

The consumer level was repeatedly seen to be an area that needs special attention for both of the RTE products, as it is the module with greatest potential to effectively reduce the median cases of listeriosis per serving. At the same time, the mean of the hazard could be

effectively reduced by a unit operation in the process (e.g., cold smoking) or mitigation strategy; nevertheless, that does not necessarily mean that this particular step (i.e., cold smoking) in the process would be considered a RB-CCP under the Carrington (2004) criteria. For example, although cold smoking scenarios 8, 9, and 10 effectively reduced the mean of the hazard (see Figure 6.2.13) by -2, -3, and -4 log reductions, respectively, they did not achieve a low relative risk for listeriosis (see Table 6.6) based on Carrington and others (2004) criteria. The consumer module is also the one with the greatest potential to reduce the variability of key parameters (e.g., storage temperature and storage duration of CSS, reheating of frankfurters). It is also noteworthy that the "contribution to variance" tornado graphs help explain how much of the variance in the output variable is attributable to each individual input.

In conclusion, this study found that by using sensitivity analysis and what-if scenario analyses it is possible to identify RB-CCPs, which are the steps in the process that effectively reduce the mean and/or variance of a hazard and its associated public health risks. In addition, some of the what-if scenarios described, including mitigation strategies such as food storage conditions that slow or prevent growth, could reduce the rate of listeriosis. These scenarios could help public health officials and food industry stakeholders make better-informed decisions as they quantitatively evaluate public health risks. Finally, Chapter 7 compares not only the risk assessment derived HACCP plans to traditional HACCP plans for frankfurters and cold-smoked salmon, but also the risk assessment derived HACCP plans for these two ready-to-eat products. Chapter 7 also includes the application of risk management metrics such as microbiological criteria.

Table 6.1 Frankfurters what-if scenario analyses summary

Relative Risk Ranking and Predicted Median Cases of Listeriosis for Frankfurters (Per-Serving Basis) Comparison of baseline model (FNR) and frankfurters reheated (FR) what-if scenario risk of Listeriosis per serving for three subpopulations							
	Scenarios	**Reheating of Frankfurters Median Log Reduction**	**Predicted Median Cases per Serving Using different r-values from the literature**			**Median Cases per Servings FDA/USDA 2003**	**Relative Risk Ranking Based on Carrington and others 2004**
			Susceptible Population FAO/WHO 2004	**Elderly Population FAO/WHO 2001**	**Perinatal Population FAO/WHO 2001**		
	Baseline						Moderate Risk
1	**Frankfurter non-reheated (FNR)**	N/A	4.18×10^{-9}	3.31×10^{-8}	1.77×10^{-7}	6.5×10^{-8}	High Risk
2	**Frankfurter Reheated (FR)**	4.49 (see Table 6.3)	3.16×10^{-14}	2.51×10^{-13}	1.34×10^{-12}	6.3×10^{-11}	Low Risk

Table 6.2 Frankfurters reheating log reduction data and distribution

Reheating Median Log Reduction		Distribution Created for Reheating Log Reduction
Percentiles	**Median Reduction**	
50	4.49	=RiskCumul(4.49,6.68,{5.3,6.18,6.68},{0.75,0.95,0.99},RiskName("Reheating L Reduction"))
75	5.3	
95	6.18	
99	6.68	

Data obtained from FDA/USDA (2003)

Table 6.3 Frankfurters sensitivity analyses summary

Risk-based Critical Control Points (RB-CCPs) summary for FR and FNR baseline scenarios at the consumer output level				
Priority Level Results from the Sensitivity Analysis (SA)	**Baseline Scenario (FNR)**		**Frankfurter Reheated (FR)**	
Risk-based Critical Control Points (RB-CCPs)	SA Output Mean[a]	SA Contribution to Variance[b]	SA Output Mean[a]	SA Contribution to Variance[b]
Reheating Log Reduction at Consumer Level			1	2
Handling at packaging	1	1	2	1
Duration of Refrigerated Consumer Storage	2	2	3	3
Duration of Transportation to Retail Distribution	3	3	4	4
Duration of Transportation from retail to consumer	4	4	5	5
Duration of refrigerated storage during retail	5	5	6	6
Temperature of unopened package at consumer refrigerator	6	6		
Temperature of opened package at consumer refrigerator			7	7
pH of unopened package at consumer refrigerator	7			
pH of unopened package at retail distribution	8		10	
Temperature of unopened package at retail distribution	9		8	
Nitrite concentration of unopened package at retail distribution	10			
Nitrite concentration of opened package at consumer			9	

[a] Inputs ranked by effect on output mean tornado (ranking based on these results)
[b] Contribution to variance tornado (ranking based on these results)

Table 6.4 Cold-smoked salmon what-if scenario analyses summary

Relative Risk Ranking and Predicted Median Cases of Listeriosis for Cold Smoked Salmon (Per Serving Basis)

Scenario Number	Scenarios based on Sensitivity Analysis	Initial Lm Level	Thawing % of baseline	Cold Smoking Log Reduction			Slicing (Immediately prior) Log Reduction	Distribution and Marketing				Consumer Level				Median Cases per Servings	Relative Risk Ranking (based on Carrington et al. 2004)
								Time (hours)	Temperature (°C)			Time (hours)	Temperature (°C)				
1	**Baseline (frozen)**	100%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	4.46×10^{-9}	Moderate Risk
2	**Initial Lm**	50%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	1.37×10^{-8}	High Risk
3	**Level**	10%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	4.49×10^{-8}	High Risk
4		1%	100%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	5.97×10^{-8}	High Risk
5	**Thawing**	100%	50%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	3.20×10^{-9}	Moderate Risk
6		100%	10%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	1.95×10^{-9}	Moderate Risk
7		100%	1%	0	1	2	0	100%	-3	5	14	100%	-5	3	17	1.5×10^{-9}	Moderate Risk
8	**Cold**	100%	100%	1	2	3	0	100%	-3	5	14	100%	-5	3	17	1.74×10^{-9}	Moderate Risk
9	**Smoking**	100%	100%	2	3	4	0	100%	-3	5	14	100%	-5	3	17	1.20×10^{-9}	Moderate Risk
10		100%	100%	3	4	5	0	100%	-3	5	14	100%	-5	3	17	1.09×10^{-9}	Moderate Risk
11		100%	100%	0	1	2	1	100%	-3	5	14	100%	-5	3	17	1.71×10^{-9}	Moderate Risk
12	**Slicing**	100%	100%	0	1	2	2	100%	-3	5	14	100%	-5	3	17	1.16×10^{-9}	Moderate Risk
13		100%	100%	0	1	2	3	100%	-3	5	14	100%	-5	3	17	1.05×10^{-9}	Moderate Risk
14	**Distribution**	100%	100%	0	1	2	0	50%	-3	5	14	100%	-5	3	17	2.55×10^{-9}	Moderate Risk
15	**time**	100%	100%	0	1	2	0	10%	-3	5	14	100%	-5	3	17	1.64×10^{-9}	Moderate Risk
16		100%	100%	0	1	2	0	1%	-3	5	14	100%	-5	3	17	1.49×10^{-9}	Moderate Risk
17	**Distribution**	100%	100%	0	1	2	0	100%	-5	-3	-1	100%	-5	3	17	3.13×10^{-9}	Moderate Risk
18	**Temperature**	100%	100%	0	1	2	0	100%	3	5	7	100%	-5	3	17	4.46×10^{-9}	Moderate Risk
19		100%	100%	0	1	2	0	100%	12	14	16	100%	-5	3	17	6.74×10^{-9}	High Risk
20	**Consumer**	100%	100%	0	1	2	0	100%	-3	5	14	50%	-5	3	17	9.58×10^{-11}	Low Risk
21	**time**	100%	100%	0	1	2	0	100%	-3	5	14	10%	-5	3	17	3.56×10^{-12}	Low Risk
22		100%	100%	0	1	2	0	100%	-3	5	14	1%	-5	3	17	1.62×10^{-12}	Low Risk
23	**Consumer**	100%	100%	0	1	2	0	100%	-3	5	14	100%	-7	-5	-3	1.30×10^{-12}	Low Risk
24	**Temperature**	100%	100%	0	1	2	0	100%	-3	5	14	100%	1	3	5	1.33×10^{-9}	Moderate Risk
25		100%	100%	0	1	2	0	100%	-3	5	14	100%	13	17	21	1.35×10^{-4}	High Risk

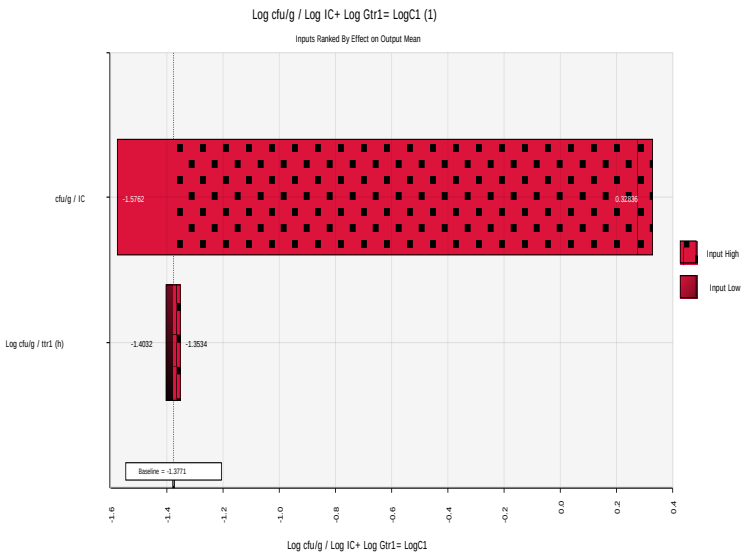
Table 6.5 Cold-smoked salmon sensitivity analyses summary

Risk-based Critical Control Points (RB-CCPs) summary for selected scenarios and baseline model at the consumer output level										
Risk-based Critical Control Points (RB-CCPs)	**Baseline Scenario**		**Scenario 20**		**Scenario 21**		**Scenario 22**		**Scenario 23**	
Priority Level Results from the Sensitivity Analyisis (SA)	SA Mean[a]	SA Variance[b]	SA Mean[a]	SA Variance[b]	SA Mean[a]	SA Variance[b]	SA Mean[a]	SA Variance[b]	SA Mean[a]	SA Variance[b]
Temperature Consumer	1	1	1	1	5	5	9		3	2
Time Consumer	2	2	2	2	6	7	10		8	7
Slicing	3	7	3	7	1	6	1	5	1	6
Temperature Distribution	4	3	4	3	2	1	2	1	2	1
Time (Retail) Distribution	6	5	5	4	3	2	3	2	4	3
Cold Smoking	5	4	6	5	4	3	4	3	5	4
Raw Salmon Lm Incidence	7	6	7	6	7	4	5	4	6	5
Initial Lm reference distribution			8	8	8	8	6	6	7	
Portioning	9		9		9		7		9	
Filleting	8									
Pinbone			10		10	9	8	7	10	

[a] Inputs ranked by effect on output mean tornado
[b] Contribution to variance tornado

Figure 6.1 Frankfurter baseline model - Ingredients module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

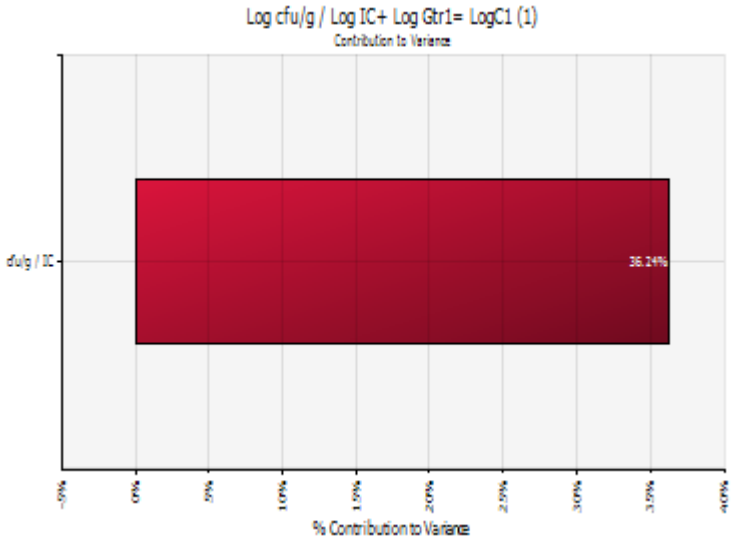
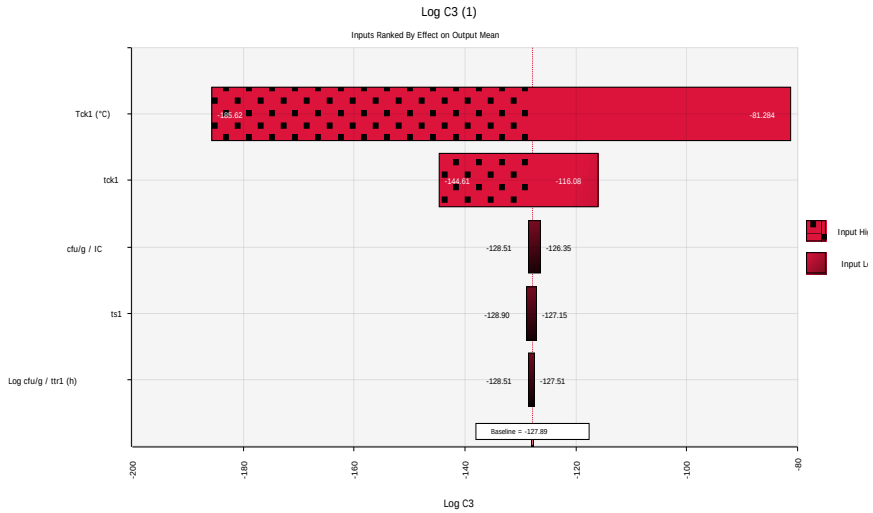


Figure 6.2 Frankfurters baseline model - Raw product processing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

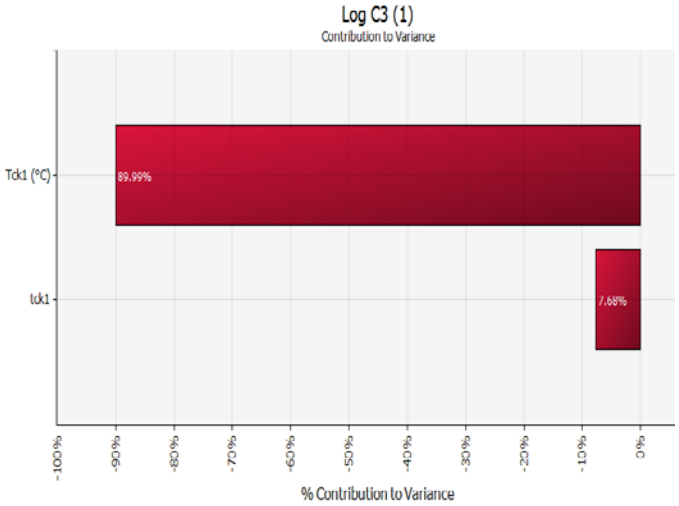
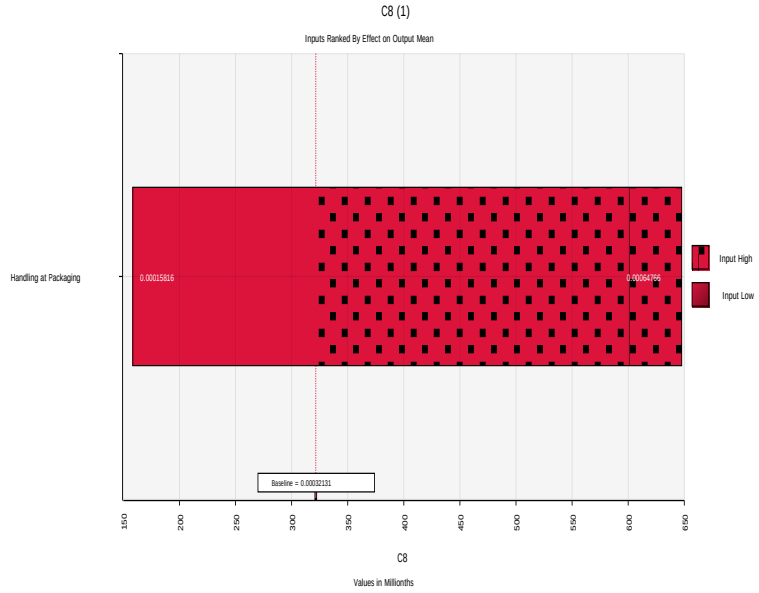


Figure 6.3 Frankfurters baseline model - Cooked product processing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

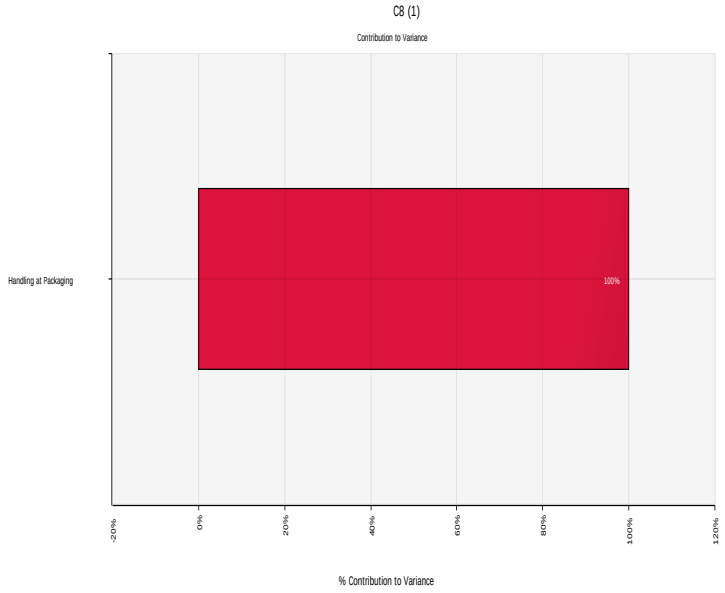
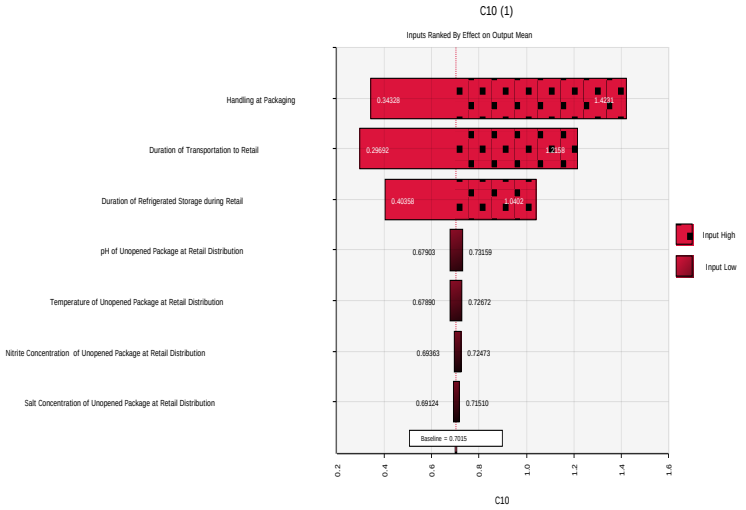


Figure 6.4 Frankfurter baseline model - Distribution and marketing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

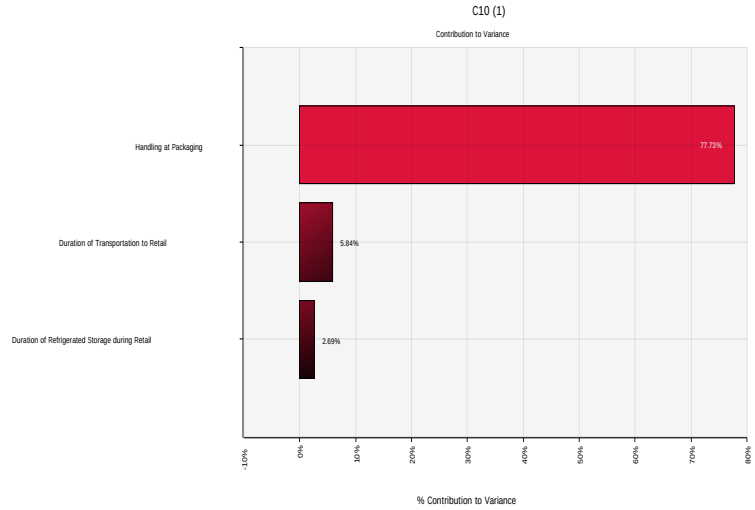
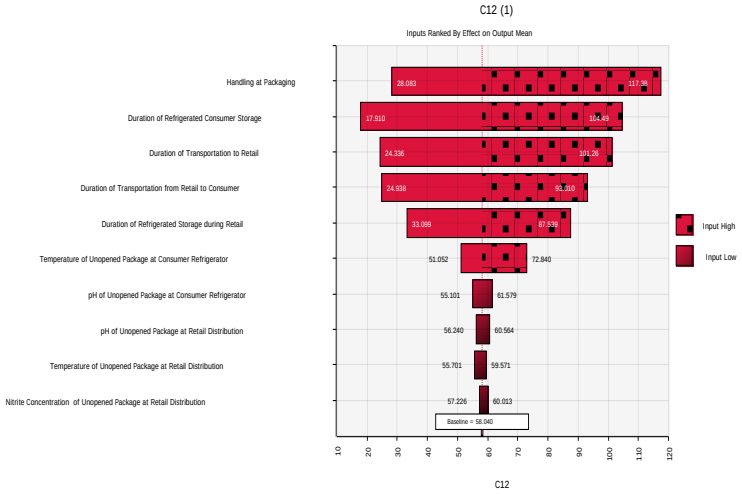


Figure 6.5 Frankfurter baseline model - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado (FNR)*



B. *Contribution to variance tornado (FNR)*

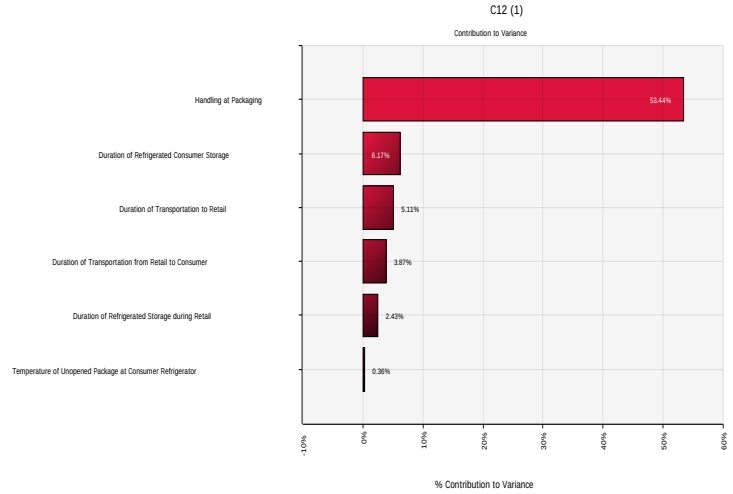
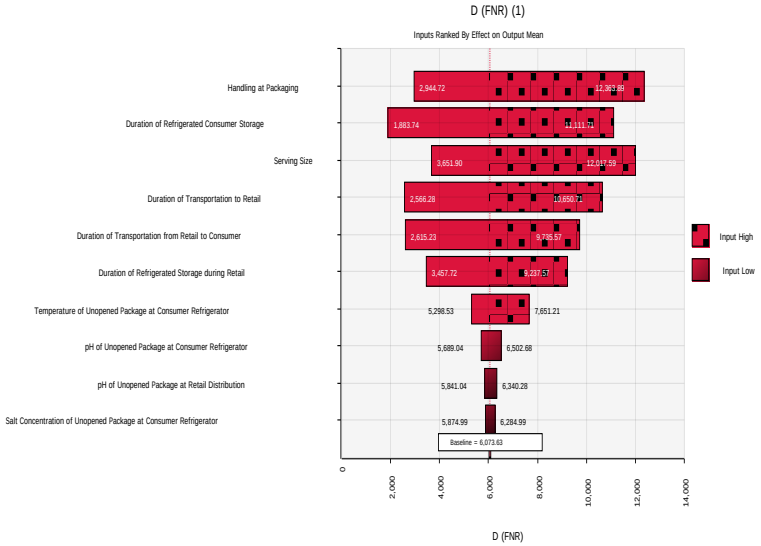


Figure 6.6 Frankfurter baseline model - Dose-response module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado (FNR)*



B. *Contribution to variance tornado (FNR)*

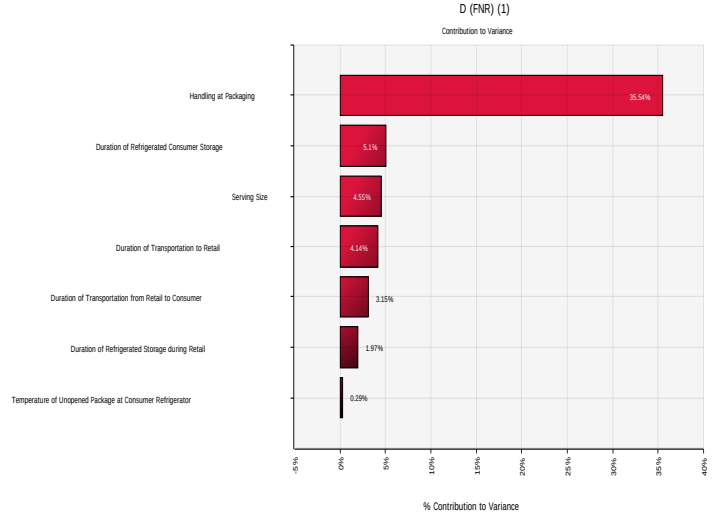
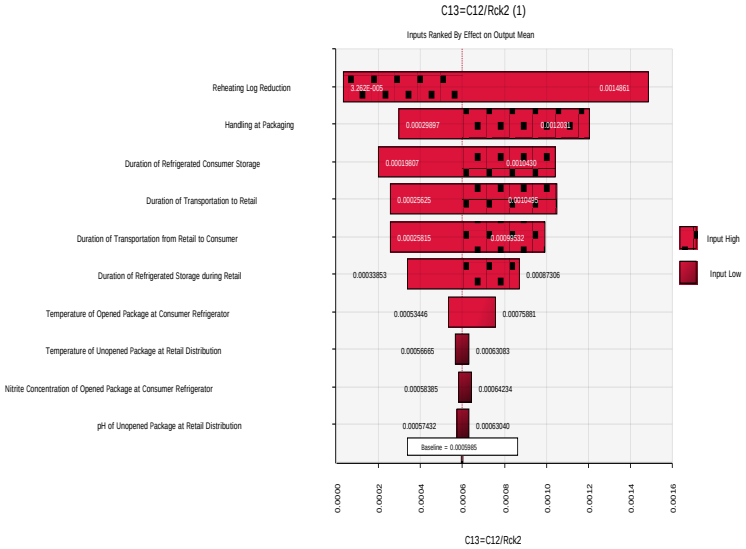


Figure 6.7 Frankfurter reheated what-if scenario results - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado (FR)*



B. *Contribution to variance tornado (FR)*

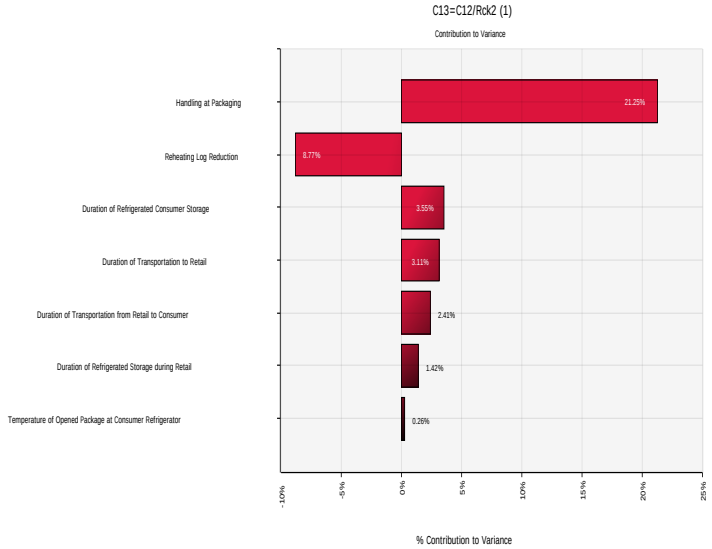
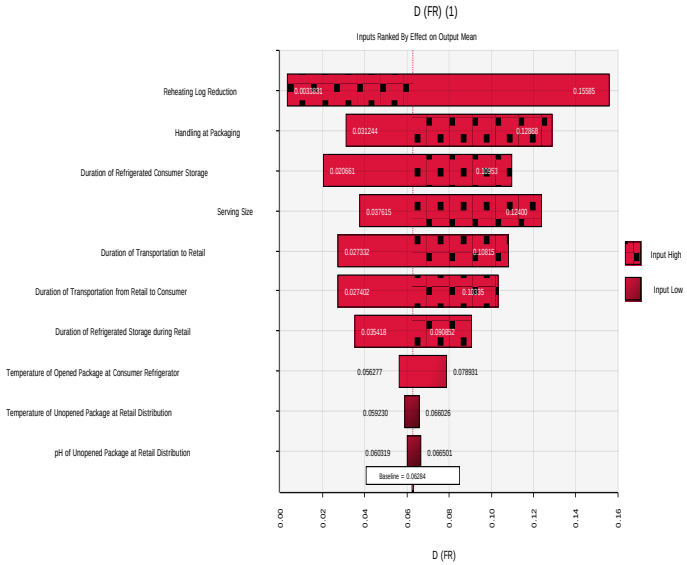


Figure 6.8 Frankfurter reheated what-if scenario results - Dose-response module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado (FR)*



B. *Contribution to variance tornado (FR)*

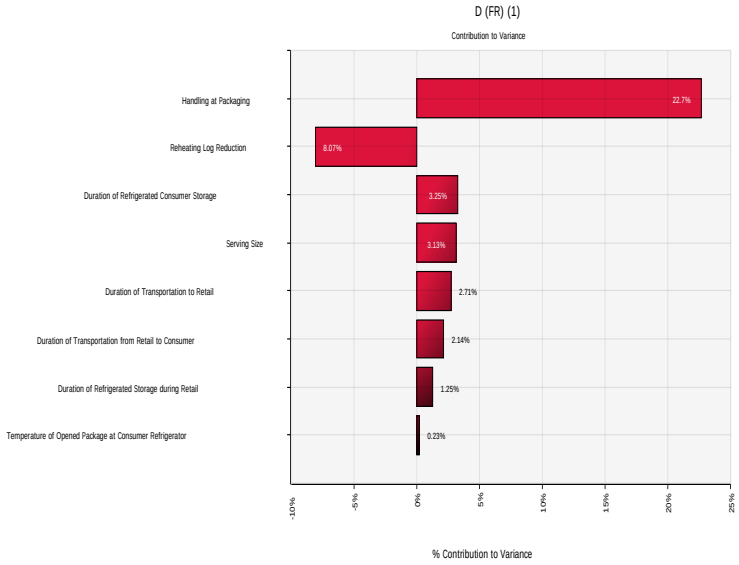


Figure 6.9 Frankfurter not reheated (FNR) baseline model and frankfurter reheated (FR) - What-if scenario results

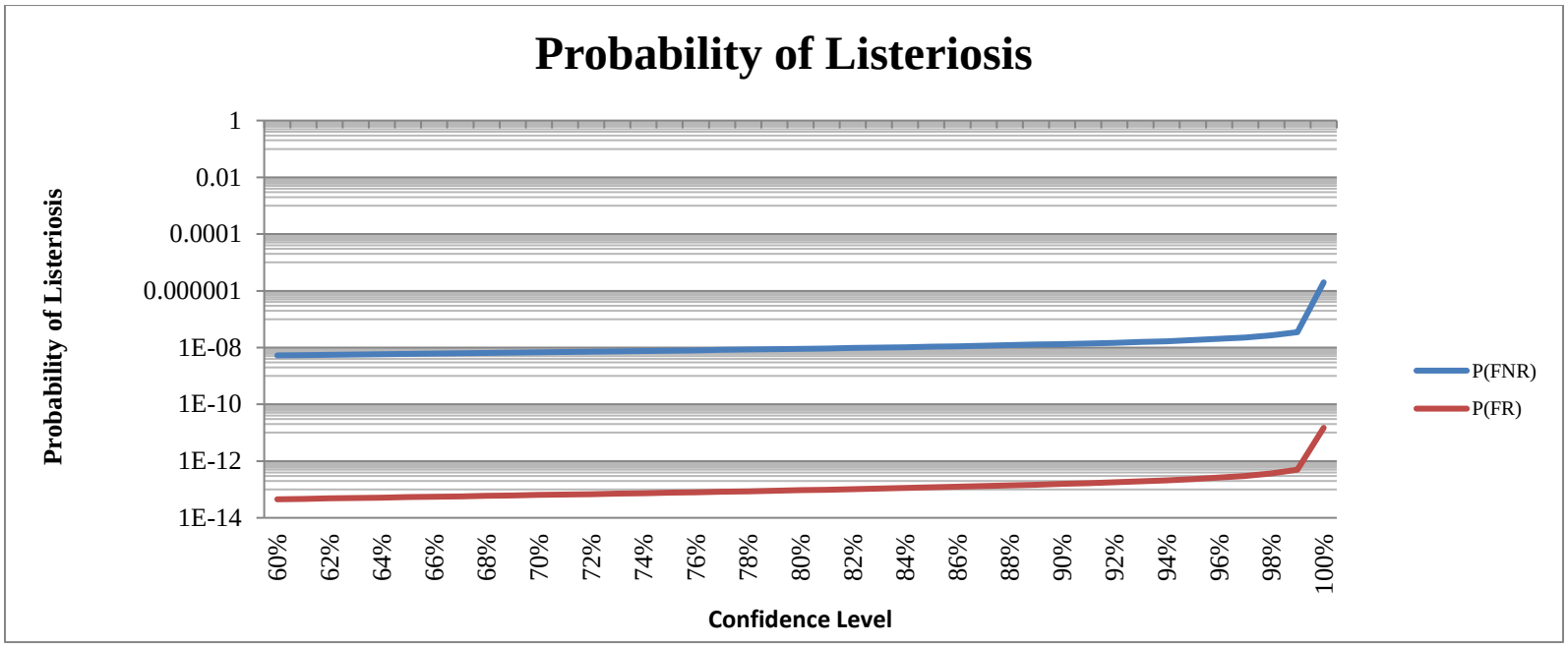
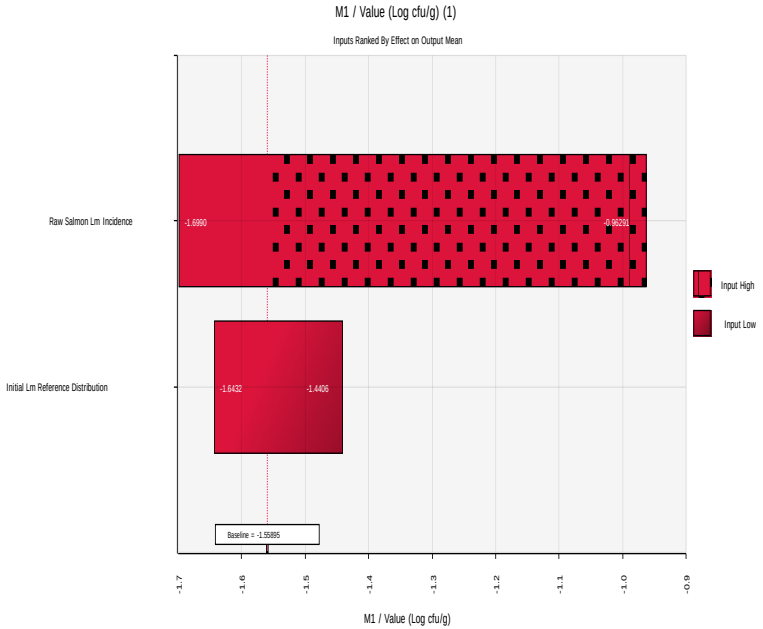


Figure 6.10 Cold-smoked salmon baseline model - Primary production module - Sensitivity analysis results in @Risk

C. Inputs ranked by effect on output mean tornado



D. Contribution to variance tornado

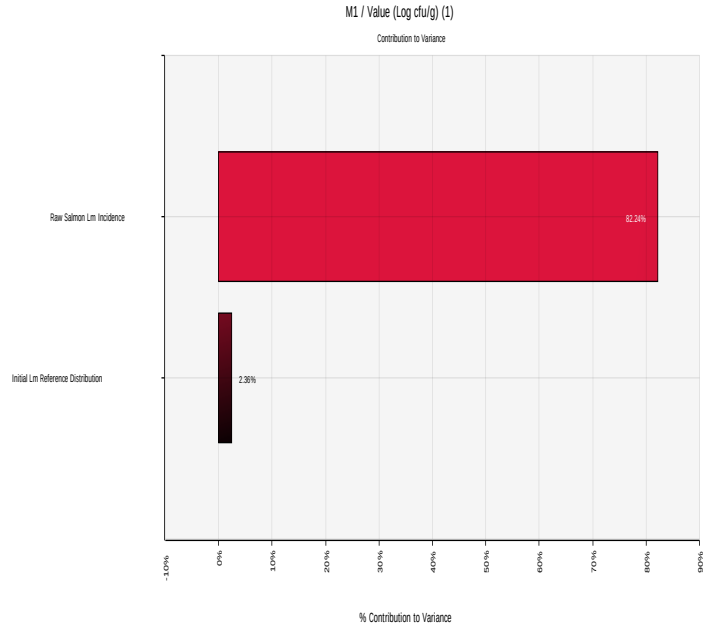
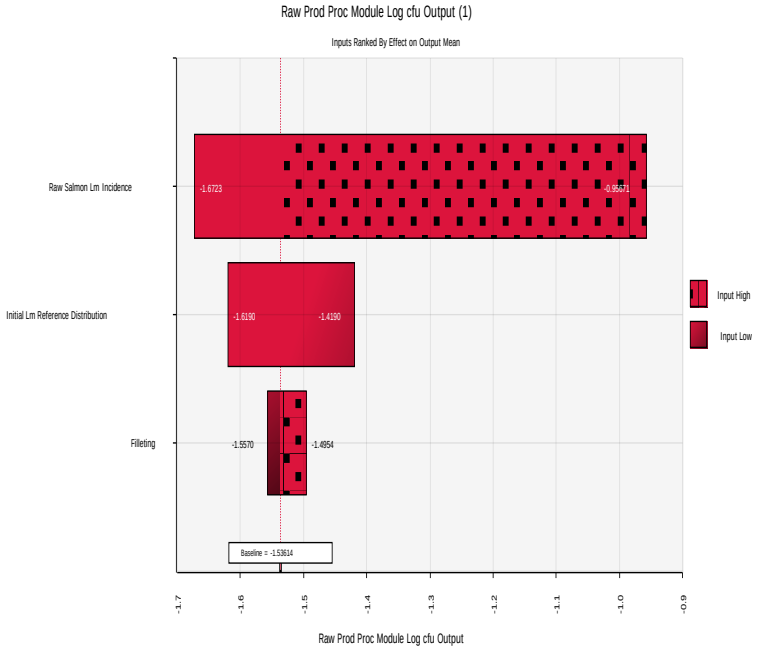


Figure 6.11 Cold-smoked salmon baseline model - Raw product processing module - Sensitivity analysis results in @Risk

C. *Inputs ranked by effect on output mean tornado*



D. *Contribution to variance tornado*

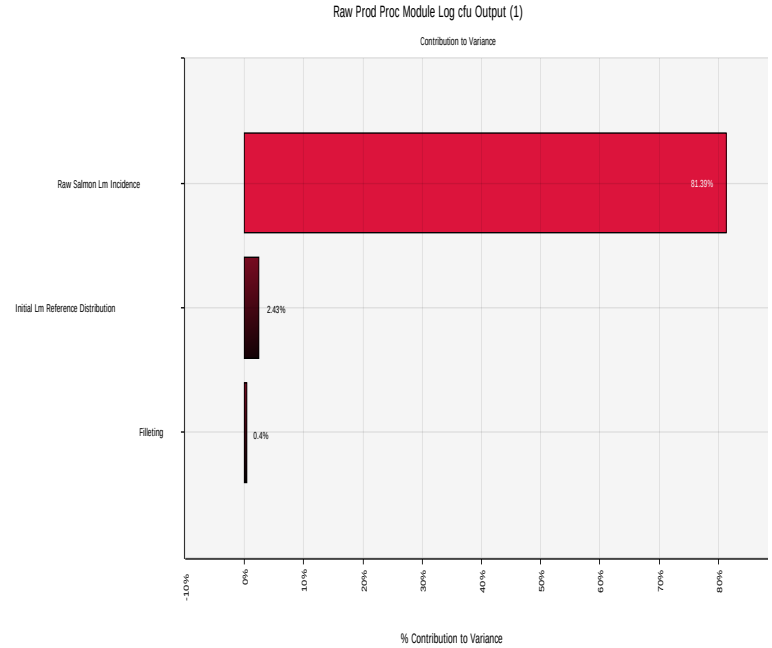
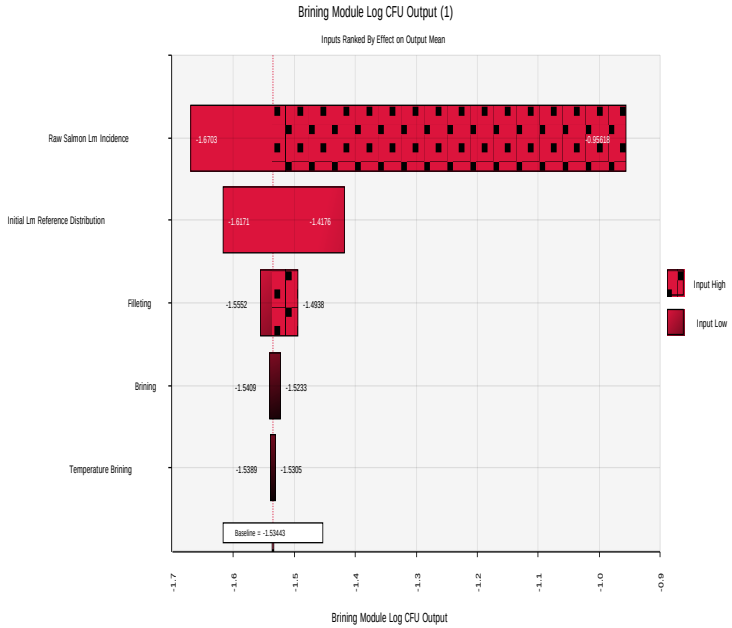


Figure 6.12 Cold-smoked salmon baseline model - Brining module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

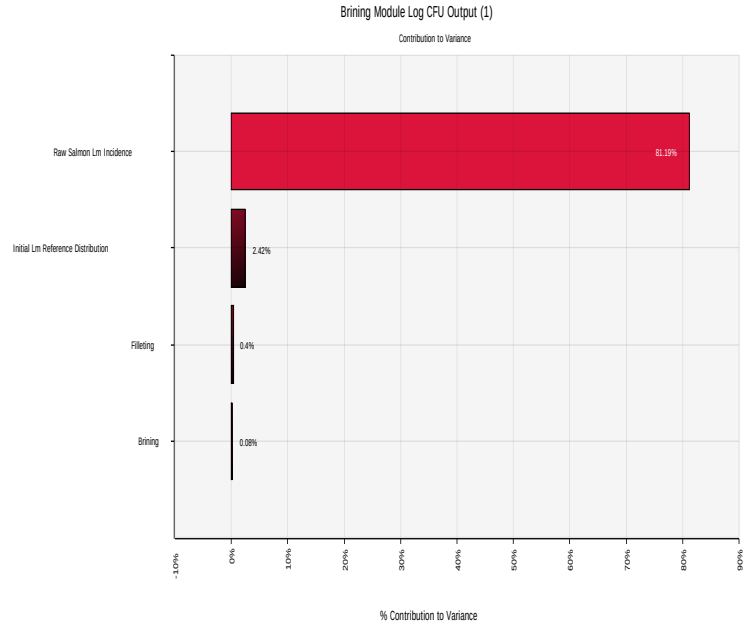
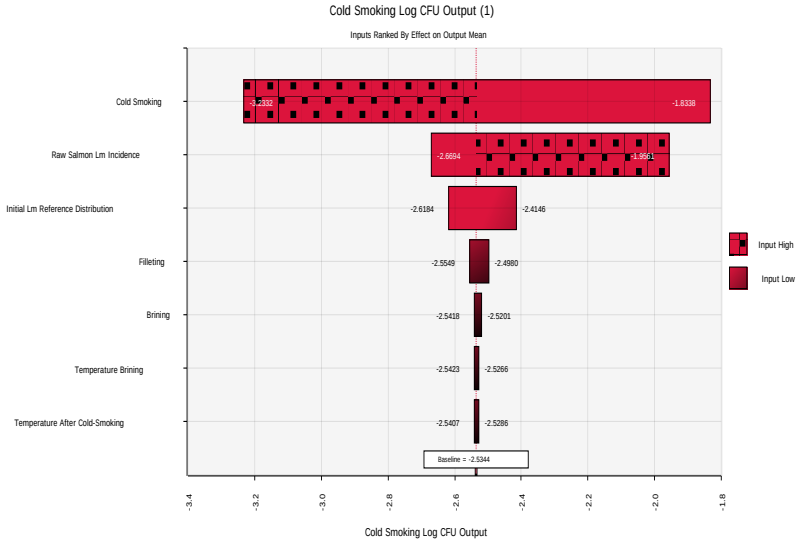


Figure 6.13 Cold-smoked salmon baseline model - Cold-smoking processing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

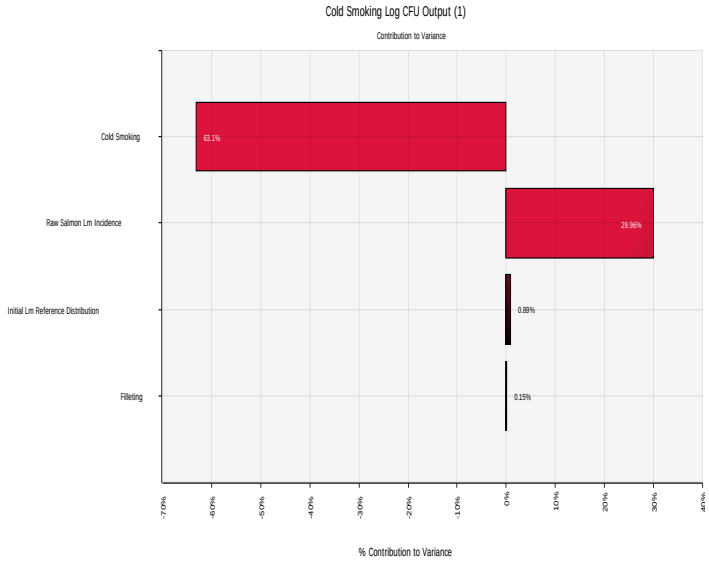
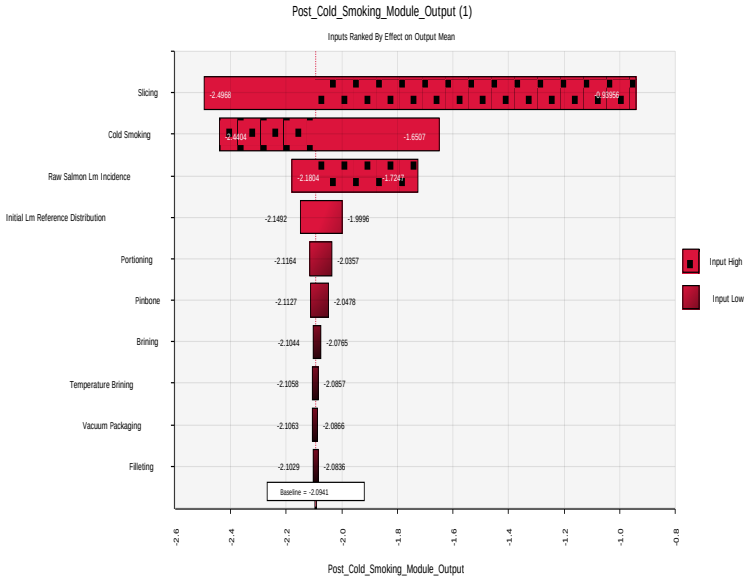


Figure 6.14 Cold-smoked salmon baseline model - Post-cold smoking processing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

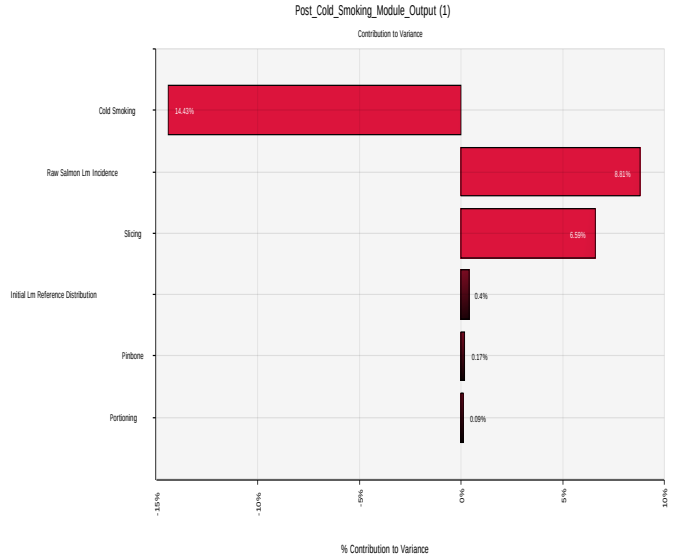
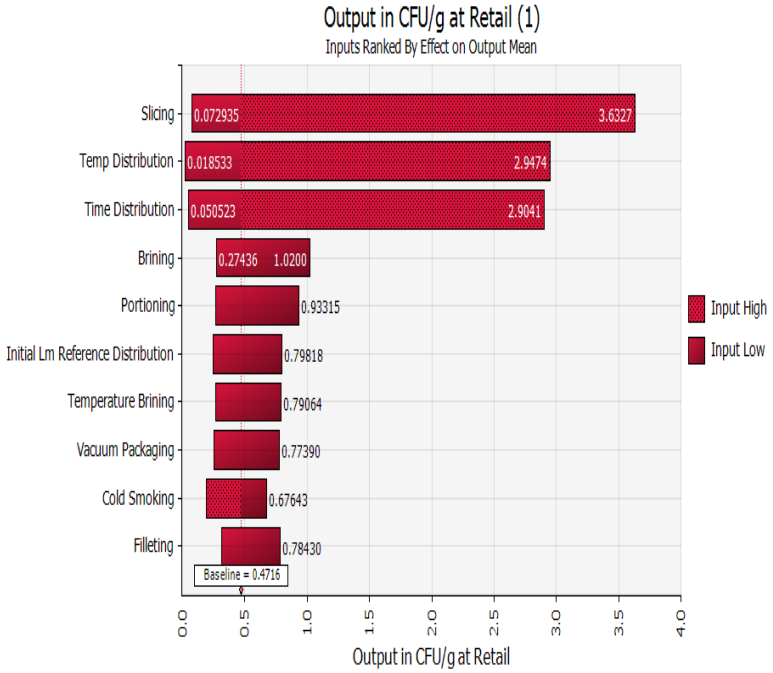


Figure 6.15 Cold-smoked salmon baseline model - Distribution and marketing module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

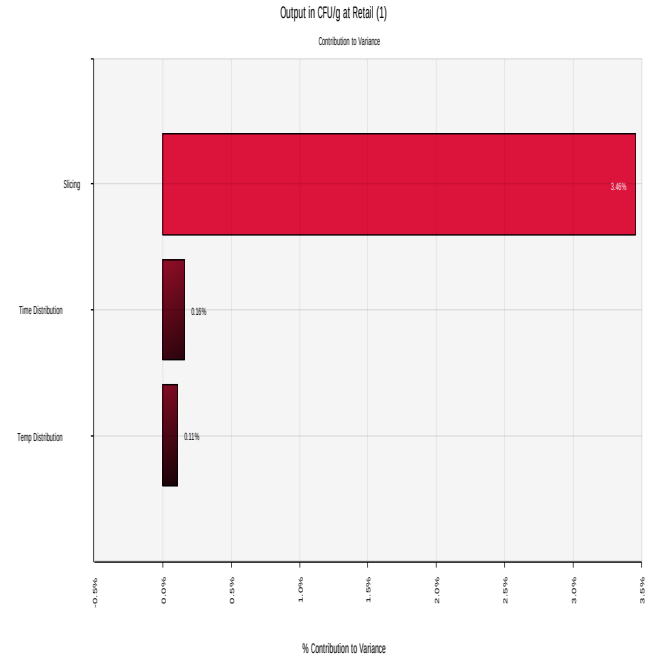
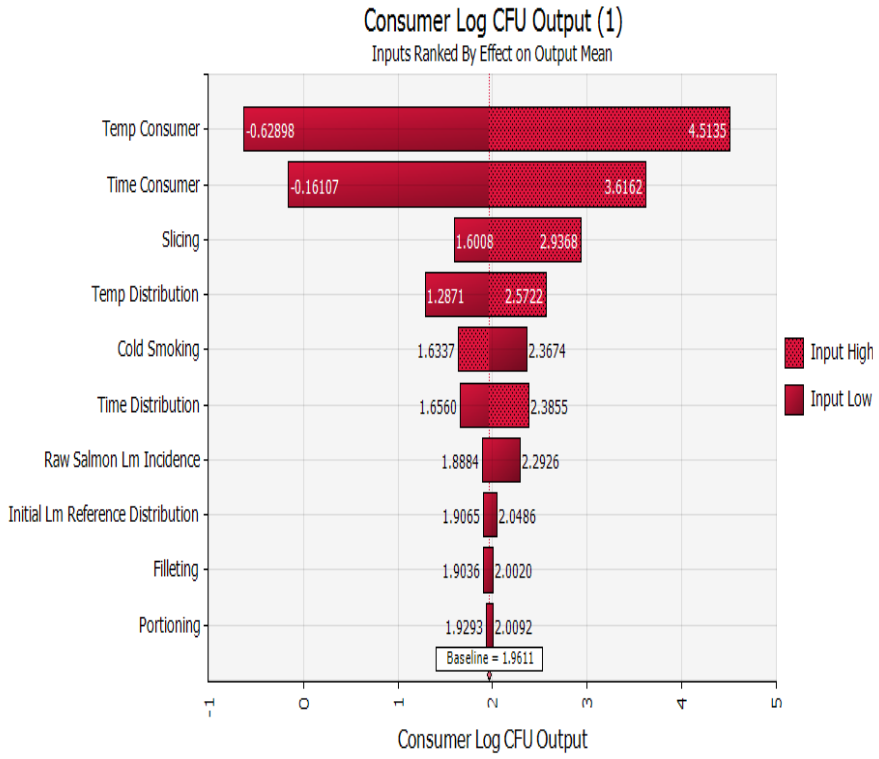


Figure 6.16 Cold-smoked salmon baseline model - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

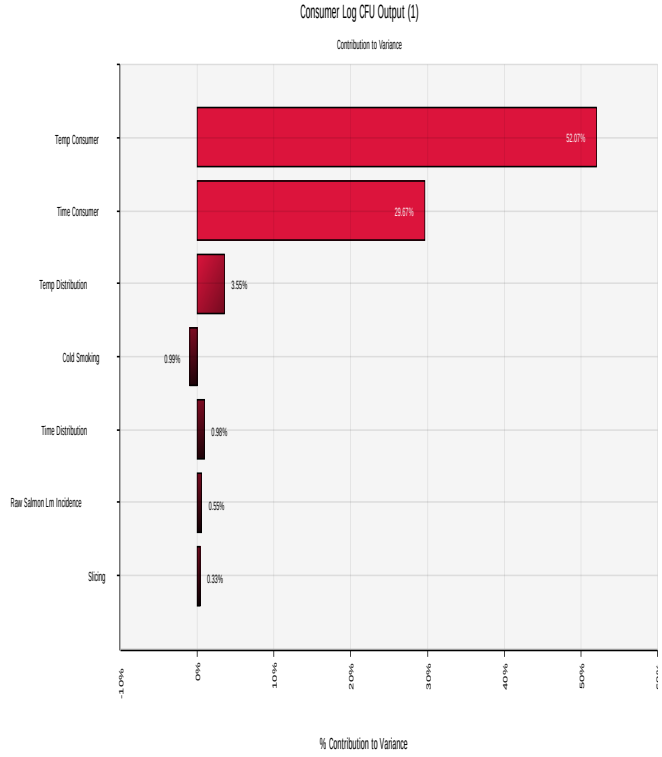
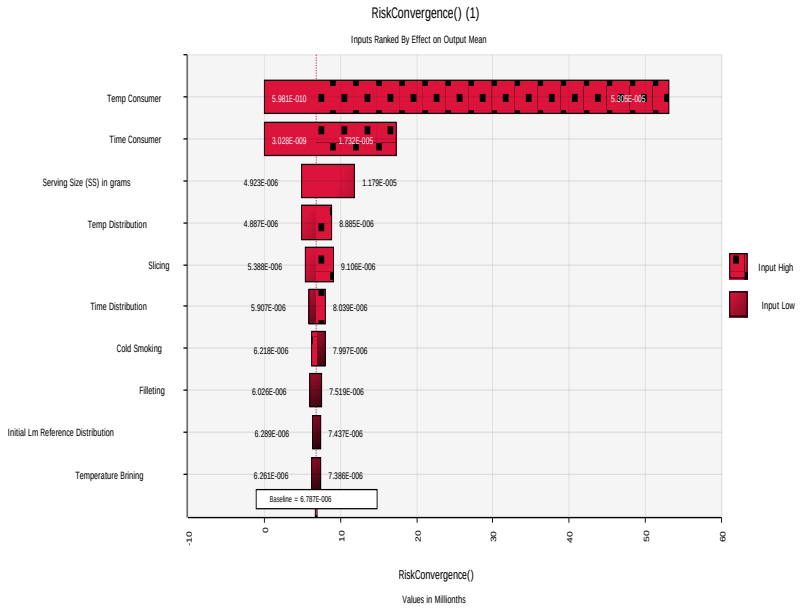


Figure 6.17 Cold-smoked salmon baseline model - Dose-response module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

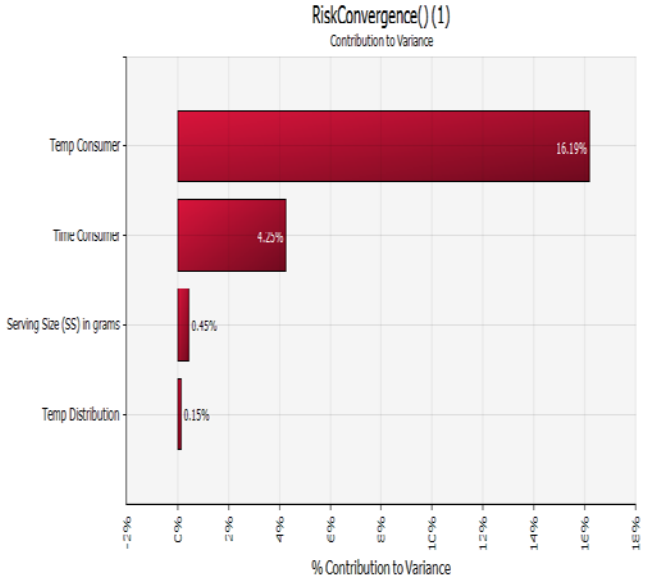
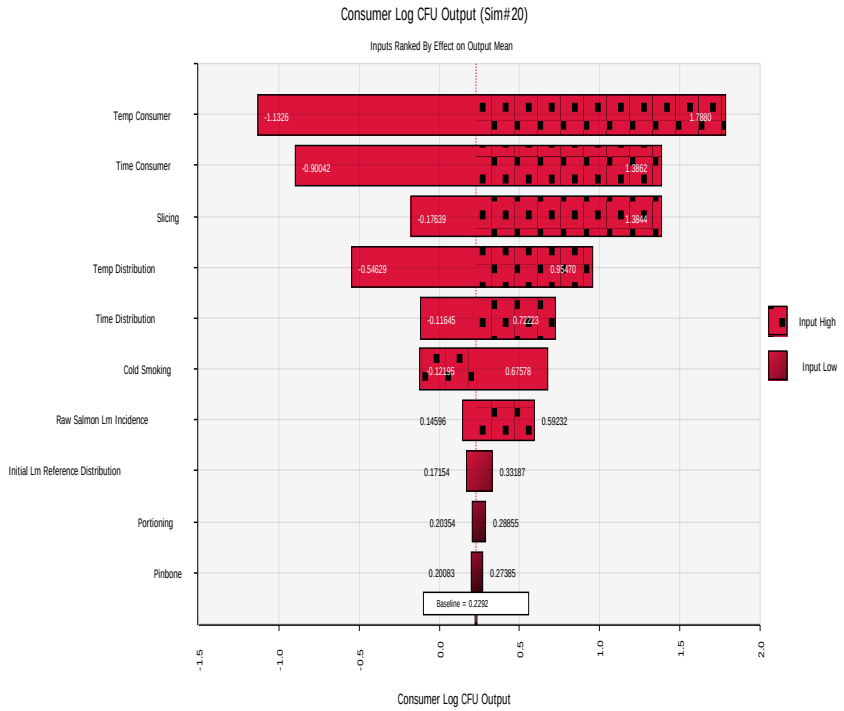


Figure 6.18 Cold-smoked salmon Scenario 20 - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

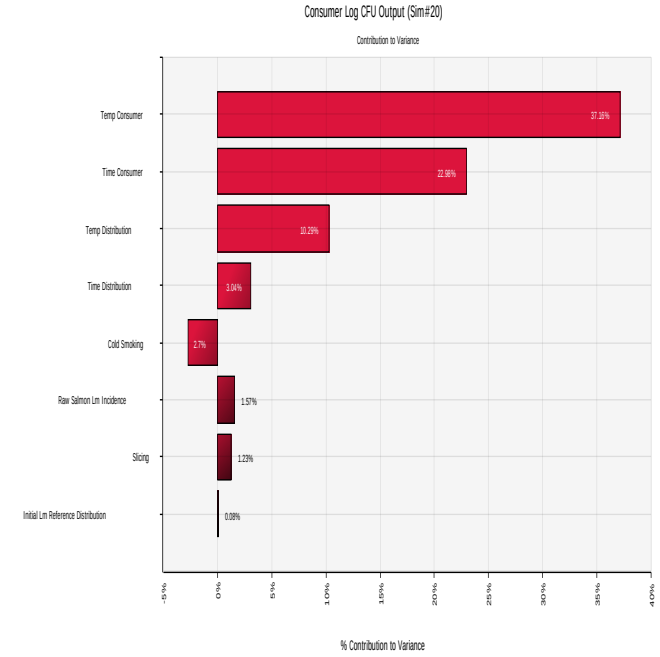
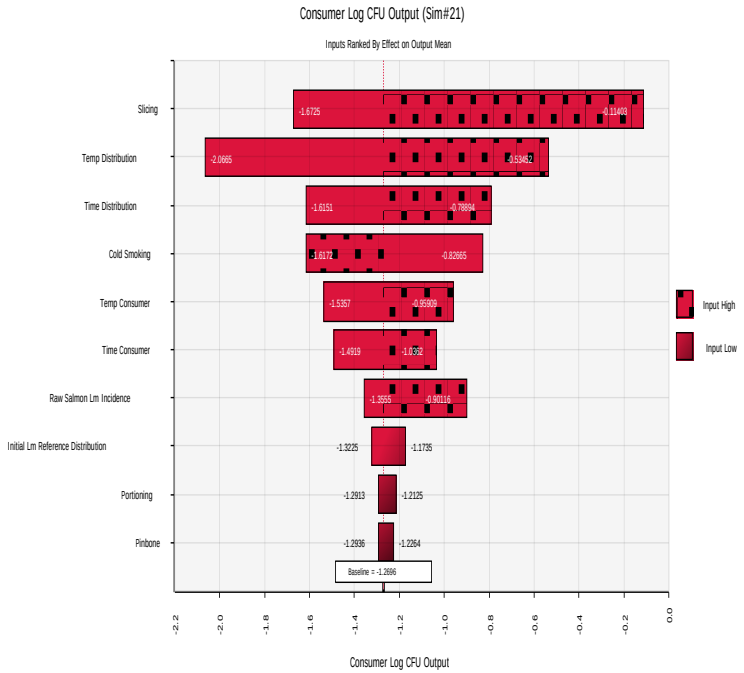


Figure 6.19 Cold-smoked salmon Scenario 21 - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

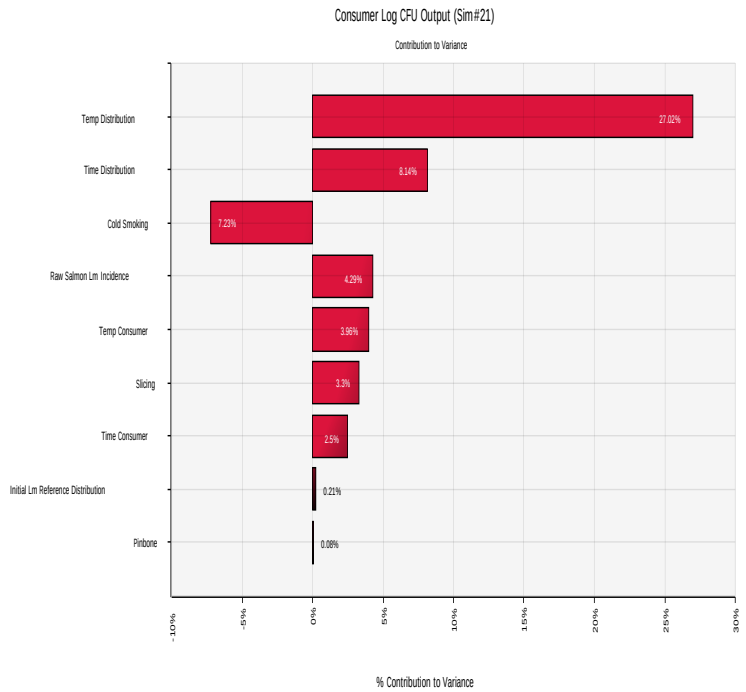
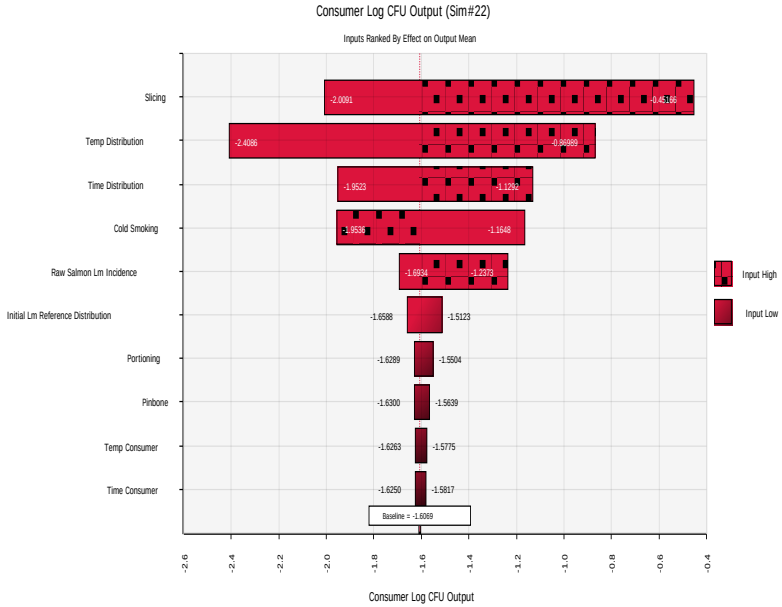


Figure 6.20 Cold-smoked salmon Scenario 22 - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

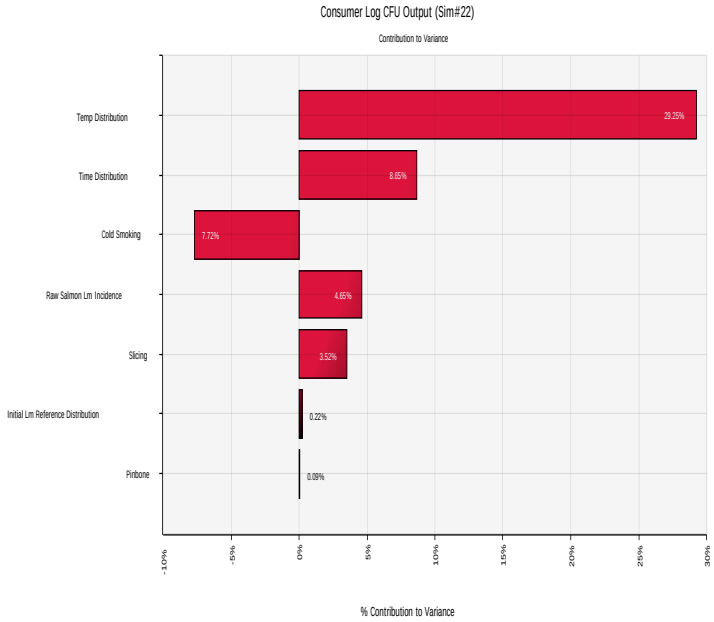
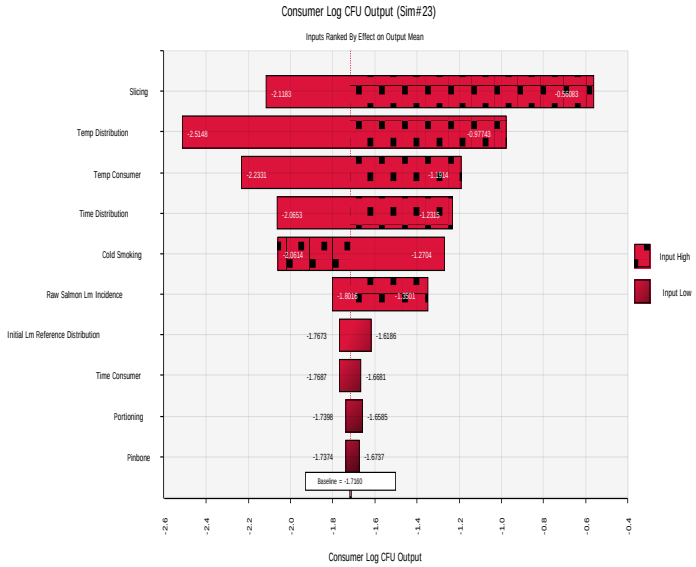


Figure 6.21 Cold-smoked salmon Scenario 23 - Consumer module - Sensitivity analysis results in @Risk

A. *Inputs ranked by effect on output mean tornado*



B. *Contribution to variance tornado*

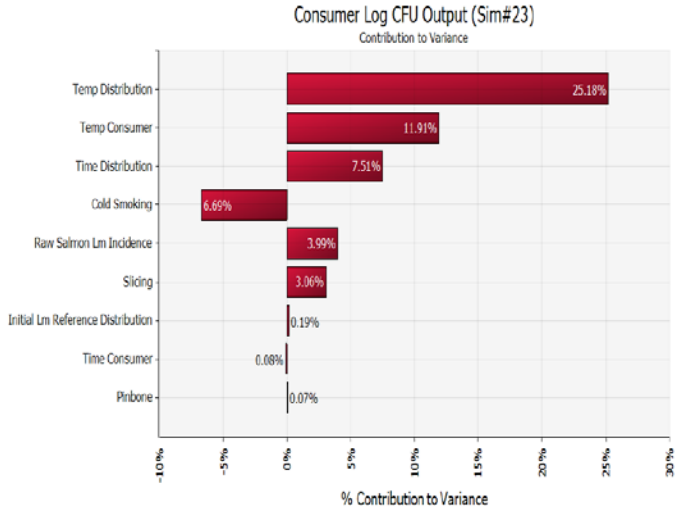
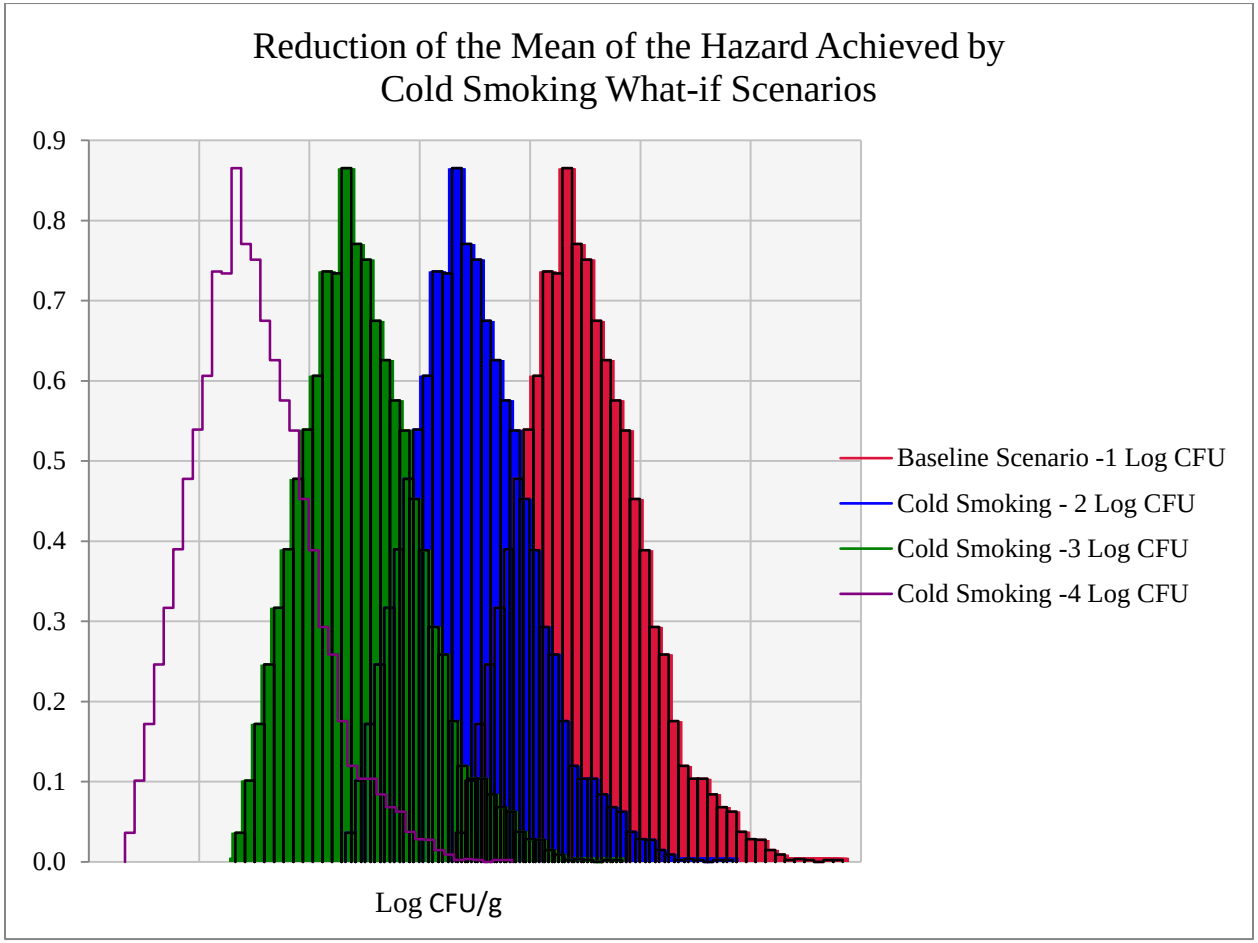


Figure 6.22 CSS Scenarios 8, 9, and 10 - What-if scenario results



Chapter 7: General Discussions and Conclusions on the Risk Assessment Derived HACCP Plans for Selected Ready-to-Eat Food Products

Chapter 6 provided the risk-based critical control points (RB-CCPs) obtained through the sensitivity analysis (SA) and what-if scenario analyses for the selected ready-to-eat (RTE) products (i.e., frankfurters and cold-smoked salmon). Chapter 7 uses these results to create the risk-based Hazard Analysis Critical Control Point (RB-HACCP) plans for each of the products with regards to *Listeria monocytogenes*, the main pathogen of interest in these RTE foods. As explained in Chapter 2, invasive *L. monocytogenes* infection (listeriosis) is a rare but serious foodborne illness, causing an estimated 19% of deaths associated with foodborne diseases in the United States (U.S.) and costing an estimated $2.8 billion annually (Scallan and others 2011; USDA/ERS 2015). During the 1980s and 1990s, food safety measures targeting ready-to-eat meat and poultry products helped reduce the incidence of listeriosis by >50% (Cartwright and others 2013; Jackson and others 2016). Specifically, improved control measures starting in the 1990s have greatly reduced the prevalence of *L. monocytogenes* in many food categories, particularly in meats and meat products (Buchanan and others 2017). Indeed, the incidence of listeriosis in the U.S. decreased by 24% from 1996 through 2001 (Lorber 2010). Since 2001, listeriosis incidence has remained constant in the U.S. despite intensive efforts, staying above the Healthy People 2020 target of 0.2 cases per 100,000 (Cartwright and others 2013; CDC 2015; Jackson and others 2016; Buchanan and others 2017). The more severe, systemic form of listeriosis is now recognized as

occurring more frequently in small outbreaks than previously known (Buchanan and others 2017). Furthermore, Europe has had an upward trend of human listeriosis cases over the period 2009-2013 (EFSA 2017). Danish researchers in particular have identified cold-smoked fish as possibly being responsible for more listeriosis cases than previously recognized (Gillesberg Lassen and others 2016). Thus, cold-smoked salmon (CSS) provides an exemplary case study to address the public health challenges posed by *L. monocytogenes*, explicitly within quantitative RB-HACCP plans. Robust RB-HACCP plans represent a proactive solution addressing the root cause of potential foodborne outbreaks, especially if those food safety risk management systems are linked to public health outcomes.

This chapter presents the discussions and conclusions for the RB-HACCP plans derived from the quantitative microbial risk assessment (QMRA) models developed for frankfurters and CSS. It is noteworthy that these two products represent the continuum of RTE food products: frankfurters are given an overwhelming heat treatment, while CSS is a minimally processed RTE food product. The present chapter is divided into four main sections. The first section (section 7.1) focuses on a general discussion of the evolution of the HACCP system and describes how RB-HACCP has the potential to do a better job than current food safety management systems. The second section (section 7.2) discusses the development of RB-HACCP plans for frankfurters and CSS, and includes a comparison between risk-based and traditional HACCP plans for each of these RTE food products, as well as a comparison between the two RB-HACCP plans. This section then relates these RB-HACCP plans to food safety risk management metrics such as the food safety objective/performance

objective (FSO/PO) paradigm, thereby directly linking RB-HACCP plans to public health goals. Section 7.2 is followed by concluding remarks on the risk assessment-derived HACCP plans (section 7.3), and summarizes the main findings of this dissertation. The final section (section 7.4) addresses future research needs and offers encouragement to others to continue this important work.

7.1 *General Discussions on the Current Evolution of the HACCP System*

As mentioned in Chapter 1, the combination of the Hazard Analysis and Critical Control Point (HACCP) system and current Good Manufacturing Practices (cGMPs) is the foundation and gold standard worldwide for the management of food safety risks (NACMCF 1998; CAC 2009b; Buchanan and Williams 2013). Although HACCP has evolved over more than five decades (Sperber and Stier 2009), the current evolution of HACCP-based programs at the national level with the incumbent Preventive Controls for Human Food (PCHF) rule, as well as at the international level with the forthcoming revision (CAC 2017a, 2017b, 2017c, 2017d) to the Codex Alimentarius General Principles of Food Hygiene (GPFH) (CAC/RCP 1-1969) and current Annex: HACCP System and Guidelines for its Application (CAC 2003), is remarkable in the history of HACCP and clearly demonstrates its continuous evolution. This section provides an explanation of how a risk-based HACCP (RB-HACCP) approach could contribute to the evolution of HACCP-based systems. In the U.S., food safety management systems are evolving to include more holistic, risk-based approaches such as the Preventive Controls for Human Food (PCHF). Although the acronym HARPC (Hazard Analysis and Risk-Based Preventive Controls) is not used within the “Current Good Manufacturing Practice, Hazard Analysis, and Risk Preventive Controls; Final Rule”

(FDA 2015), its requirements are found in the Food Safety Modernization Act (FSMA) Preventive Controls Rule for Human Food. In particular, 21CFR 117.135(a)(2) of the Preventive Controls final rule states that preventive controls include: "(i) Controls at critical control points (CCPs), if there are any CCPs; and (ii) Controls, other than those at CCPs, that are also appropriate for food safety." Thus, the PCHF combines an HACCP-based approach with current Good Manufacturing Practices (cGMP), including not only traditional critical control points (CCPs) but also prerequisite programs (PRPs), encouraging a broader "risk-based" approach. Since the majority of recalls are due to failures to adhere to PRPs rather than failures in the HACCP plans (ICMSF 2002), the evolution to preventive controls and treating PRPs like traditional CCPs (e.g., verification, recordkeeping) seems logical. In fact, 21CFR 117.135 (b) of the Preventive Controls final rule states that "preventive controls must be written." Thus, the final rule requires a written Food Safety Plan (FSP) to be developed using the PCHF approach. The FSP framework identifies not only process preventive controls (e.g., CCPs) but also sanitation preventive controls (e.g., PRPs) in a processing line. For example, the risk of *L. monocytogenes* from the environment would result in different approaches depending on whether a HACCP/GMP plan or a PCHF/FSP is developed and implemented. Applying HACCP/GMP implies that the risk of *L. monocytogenes* from the environment is considered a PRP and thus will not have any specific program requirements for keeping PRPs as records. For example, although recontamination with *L. monocytogenes* is reasonably likely to occur during the slicing step of the process (Chaitiemwong and others 2014), according to the seafood HACCP procedures from the "Segment Two" course, slicing is considered part of PRP and does

not require record keeping because it is not considered a traditional CCP. On the other hand, applying the PCHF/FSP approach would result in a control measure of a sanitation preventive control which would require monitoring, verification, corrective action, and record keeping. In other words, a sanitation preventive control is handled similarly to a CCP requirement.

It is noteworthy that HACCP was created as a food safety assurance system focusing on prevention at a time when most food safety systems relied mainly on end-product testing. Thus, while a preventive approach to food safety is nothing new, PCHF incorporates important elements (e.g., supply-chain programs, recall plan) for a more comprehensive protection of foods under the jurisdiction of the U.S. Department of Health and Human Services (HHS), Food and Drug Administration (FDA). Exceptions to PCHF under this jurisdiction include but are not limited to the Juice and Seafood HACCP regulations (e.g., CSS). In addition, manufacturing facilities under the jurisdiction of the U.S. Department of Agriculture (USDA) are also exempt from PCHF (e.g., frankfurters). Although the RTE food products in the two case studies in this research used to develop the risk assessment-derived HACCP plans are exempt from the PCHF rule, there is still room for further evolution of the PCHF/HACCP system. For example, changes in the new approach to cGMPs are starting to affect the Seafood HACCP Training Curriculum (National Seafood HACCP Alliance 2017a): “A few additional requirements have been added to or modified in the Seafood HACCP Regulation in response to passage of the Food Safety Modernization Act (FSMA).” Specifically, the FDA updated the cGMPs or GMPs regulations as part of a broader modernization of food safety regulations: “By 2018, 21 CFR Part 117 – Subpart B-

Good Manufacturing Practices will replace 21 CFR Part 110 Good Manufacturing Practices" (National Seafood HACCP Alliance 2017b). Two notable changes to GMPs have been incorporated into the new edition of the Seafood HACCP Training Curriculum (National Seafood HACCP Alliance 2017a): "1) The general provisions of the GMPs call for employee training in food safety and food hygiene, and 2) FDA's longstanding position that GMPs address the prevention of allergen 'cross contact' is now explicit in the revised regulatory text" (National Seafood HACCP Alliance 2017a, 2017b). Thus, the exempt categories will most likely evolve in the future to have more PCHF harmonized food protection programs.

At the international level, equivalent food safety standards with preventive controls are starting to evolve in countries such as Canada, New Zealand, and Australia. While there are several ongoing initiatives to update, in particular the concept of HACCP, the Codex Alimentarius Commission (CAC or the Commission) has been a key part of the latest HACCP awakening around the world. The Commission was established by the United Nations (UN) as a joint activity of the Food and Agriculture Organization (FAO) and the World Health Organization (WHO) to protect consumer health and promote fair practices in food trade. FAO and WHO are the main specialized UN agencies with a mandate to address a range of issues to support global food safety and protect consumer's health, typically with WHO representing issues related to public health and the FAO representing issues related to food production along the food chain. In particular, the Codex Committee for Food Hygiene (CCFH) is currently undertaking a revision (CAC 2017a, 2007b, 2017c, 2017d) of the GPFH (CAC/RCP 1-1969 Rev. 2003) and its current Annex: HACCP System and Guidelines for its

Application (CAC 2003). While the "revision of the General Principles of Food Hygiene, [is] at an early stage" (USDA/FSIS 2016), this upcoming revision will integrate the current HACCP annex into the main body of this standard, combining the GPFH and its annex into one document (CAC 2017b). In addition, the committee plans to redraft specific sections of the GPFH (CAC 2017b, 2017d) for revision at the Forty-Ninth Session of the Codex Committee on Food Hygiene (CCFH49) scheduled for November 13-17, 2017 (CAC 2017c). The provisional agenda for this meeting explains that the working document CX/FH 17/49/5 and comment document CX/FH 17/49/5-Add.1 will be discussed at the physical working group (PWG) to be held in-session in November. While the report (CX/FH 17/49/5) of the electronic working group (EWG) was posted on the Codex website (CAC 2017a) on July 26, 2017, other related documents (e.g., comment document CX/FH 17/49/5-Add.1) are not currently available to the general public. The provisional agenda also explains that the "report of this working group will be made available as a CRD at the Session" (CAC 2017c). Although all the latest Codex Alimentarius HACCP working documents are not available, the CCFH "aims for a revised [HACCP] standard addressing all recent developments in the field of food safety risk management" (CAC 2015) and encourages users to "draw on guidance in existing Codex documents e.g. CAC/GL 63 2007" (CAC 2017b). These documents refer to the Principles and Guidelines for the Conduct of Microbiological Risk Management (CAC 2007b, CAC 2008a). In a similar manner, the Preventive Controls for Human Food (PCHF) system aims to incorporate a broader risk-based approach. Since both standards (i.e., Codex Alimentarius HACCP and PCHF) aim to be risk-based, they would benefit from the RB-HACCP approach

developed in this dissertation. An explanation of how RB-HACCP could contribute to the evolution of HACCP-based systems is presented below. In particular, since the latest Codex HACCP working documents are not yet available, this section focuses on how RB-HACCP could promote a greater likelihood of arriving at PCHF-type FSPs. Although including Preventive Controls within a PCHF plan is a step forward in the evolution of HACCP, without a direct link to public health goals through the food safety objective (FSO) concept and the use of the FSO and other risk management metrics, the goal to achieve true risk-based preventive controls would not be realized.

My review of current Food Safety Preventive Controls Alliance (FSPCA) Preventive Controls for Human Food Curriculum (FSPCA 2016) and formal training as Preventive Controls Qualified Individual (PCQI) suggest that a RB-HACCP system approach could enhance the current PCHF approach to FSPs in several ways. Specifically, a RB-HACCP could be a less subjective way to develop a FSMA-type FSP in the following respects:

1. Using the RB-HACCP approach could help in objectively addressing the identification of essential areas in a process with regards to a specific pathogen, by quantitatively identifying risk-based critical control points (RB-CCPs) and associated control measures as they relate specifically to public health goals using the food safety objectives/performance objectives (FSO/PO) paradigm. In fact, the RB-HACCP approach not only identifies RB-CCPs but also prioritizes them using sensitivity analysis and related analyses. Thus, RB-HACCP represents a more objective rationale of what should be included in the

preventive control programs to achieve a transparent, science-based, and truly risk-based food safety system.

2. The RB-HACCP approach presents the advantage of a real-time tool that could be used to monitor potential deviations of the process in regards to specific parameters (e.g., temperature and duration/time).

3. RB-CCPs combine CCPs, operational Prerequisite Programs (oPRPs), and Prerequisite Programs (PRPs) into a single integrated food control system. It would be possible, however, to classify the RB-CCPs into these three categories by using, for example, the oPRP decision tree by Michigan State University (2009). It is noteworthy that Codex has identified that their current decision tree applied to identify CCPs should be reviewed (CAC 2017b). There are various decision trees that can be used depending on the specific needs (e.g., ILSI Europe and van Schothorst [2004] decision tree for raw materials); however, that discussion lies outside of the scope of this dissertation.

4. The RB-HACCP through the use of the FSO/PO paradigm promotes a broader risk-based systems approach of the process by considering the effects of the food chain prior to (e.g., ingredients module) and subsequent to (e.g., consumer module) what is typically considered the food processing operations (i.e., manufacturing facilities) when developing RB-HACCP plans as food safety management tools.

To summarize, the HACCP system is currently evolving at the national (i.e., PCHF) and international (i.e., Codex HACCP) levels. The use of risk assessment modeling

techniques to develop quantitative RB-HACCP plans could enhance the evolution of this standard by, among other advances, relating HACCP-based systems to risk management metrics and ultimately to public health goals.

7.2 *Discussions on the Development of Risk Assessment Derived HACCP Plans for Selected Ready-to-Eat Food Products*

This section on the development of risk assessment-derived HACCP plans for selected RTE food products presents, as its main feature, the RB-HACCP plans for frankfurters and cold-smoked salmon (CSS), specifically developed for *L. monocytogenes*. This study pre-established *L. monocytogenes* as the hazard of interest for RTE food products since developing a quantitative risk assessment for each of the potential hazards for these two RTE foods was outside the scope of the project. Thus, the RB-HACCP plans did not consider other biological (e.g., other pathogenic bacteria, parasites), chemical (e.g., allergens, environmental chemicals), or physical (e.g., metal inclusion) hazards. Traditional HACCP plans are typically developed around an individual processing line at the facility level. However, such plans should be informed by knowledge of the steps earlier in the supply chain and subsequent to the product being released into the marketplace. Thus, risk assessment-derived HACCP plans that include supply chain and consumer modules are helping address one of the long-term HACCP evolutionary goals of devising better-informed food safety management systems. The results clearly establish the impact of consumer storage and use practices to assure the safety of the final product and achieve the public health goals of lowering the risk of listeriosis.

Section 7.2 is divided into four main sections: the discussions on case study one (section 7.2.1) which compares traditional and risk-based frankfurter HACCP plans; the discussions on case study two (section 7.2.2) which compares traditional and risk-based CSS HACCP plans; the discussions on the RB-HACCP plans for both case studies (section 7.2.3) which compares the RB-HACCP plans for frankfurters and CSS; and the discussions on food safety risk management metrics (section 7.2.4) which relates the risk assessment derived HACCP plans to food safety objectives and other risk management metrics.

7.2.1 Case Study One—Risk-based HACCP plan for frankfurters compared to a traditional HACCP plan

This section presents the traditional HACCP plan for frankfurters (section 7.2.1.1), the risk assessment derived HACCP plan for frankfurters (section 7.2.1.2), and a comparison between traditional and risk-based HACCP plans for frankfurters (section 7.2.1.3). As an HACCP-based approach, the RB-HACCP plan forms maintained a layout similar to the traditional HACCP plan forms adopted in 1997 by the National Advisory Committee on Microbiological Criteria for Foods (NACMCF 1998). However, the column names were adapted to account for the risk-based evolution of HACCP including risk metrics and other risk assessment derived terminology. Section 7.2.1.2 describes in detail the specific columns in the RB-HACCP plan.

7.2.1.1 Traditional HACCP plans for frankfurters

The traditional HACCP plan for frankfurters was based on the HACCP plan developed by the undisclosed visited frankfurter facility and not the “Generic HACCP

Model for Fully Cooked, Not Shelf Stable Meat and Poultry Products" (USDA 1999), which is not specific for frankfurters but instead was prepared for ham and roast beef. Prerequisite programs (PRPs) are the foundation upon which traditional HACCP plans are typically developed and are essential to the reliable functioning of the HACCP plan. It is noteworthy that the PRPs included (PRP documents not shown), but were not limited to, the following: a diagram of the plant layout indicating product flow, employee traffic patterns, and separation of raw and cooked product; a potable water supply; cleaning and sanitizing standard operating procedures (SOPs); SOPs for receiving and storing ingredients; and a recall program including traceability of raw materials to suppliers, coding of finished product, and traceability through distribution. A risk assessment-derived flow chart for the frankfurters process is depicted in Figure 7.1. For a detailed explanation of each step of this process refer to Chapter 4. Table 7.1 shows the traditional HACCP plan form developed by the visited frankfurter facility. This HACCP plan includes the temperature during thermal processing as the only critical control point (CCP) for this process. It is noteworthy that although the visited facility's critical limit was specified in the HACCP plan as 158°F achieved instantaneously, the actual cooking temperature in the center of the frankfurters found at the continuous thermal process in the processing facility was on average 164.49°F (data not shown). Thus, the operating limit for the cooking temperature at this facility was above the CCP specified in this traditional HACCP plan.

It should be mentioned that although different time and temperature combinations could achieve the same performance criteria of a six-logarithm reduction (6D) of *L. monocytogenes* in ready-to-eat chilled foods (Lund and others 1989), an

internal temperature of 158°F (70°C) in the center of product for a length of time of 2 minutes (Gaze and others 1989) achieves a 6D reduction in the number of *L. monocytogenes* (FDA 2011). D-values are dependent upon the specific food being heated; however, the above-stated values are conservative and generally apply to all foods (FDA 2011, p. 419). Although this is a sufficient log reduction for this pathogen as indicated in validated time/temperature tables (lethal rate 1.000) (FDA 2011, p. 422), one should take into account the variability of the system under normal operating conditions (ICMSF 2002, p. 64). For example, if the thermal process must exceed 158°F 99.9% of the time, the above-mentioned temperature (158°F) and pertinent standard deviation (SD) should be considered. Assuming SD=0.8, then the internal temperature of 160.4°F in the center of product should be reached to assure that the thermal process exceeds 158°F 99.9% of the time to achieve an acceptable level of safety. If process variation can be reduced to 0.5°F, then the CL could be set at 159.5°F to achieve the same degree of risk. Similar calculations could apply to the duration of the thermal treatment. In the case of the frankfurter facility visited, the average temperature of 164.49°F reflects four weeks of data (data not shown) from the selected oven tunnel (N) operating with significantly cooler temperature ($P < 0.0001$) than the other tunnels (E and W). However, data from three different ovens (N, E, W) and four different seasons (one week of data per season) was collected and processed using SAS (data not shown). Significant differences in seasonality were not found. As explained in Chapter 4, the selected oven (N) that was studied in more detail had extended treatment duration in different sections of the continuous equipment, resulting in an

overwhelming thermal treatment. Further details are provided in section 7.2.1.3 in the comparison between traditional and RB-HACCP plans for frankfurters.

7.2.1.2 Risk-based HACCP plan for frankfurters

The risk assessment-derived HACCP plan for frankfurters was developed based on the results of the sensitivity analysis (i.e., risk-based critical control points) for the frankfurters model. Table 7.2 shows the RB-HACCP plan form for frankfurters specifically in regards to *L. monocytogenes*. The layout of the RB-HACCP plan form is similar to the traditional HACCP plan form (NACMCF 1998). However, the first column includes the risk assessment-derived critical control points (RB-CCPs) instead of traditional CCPs. Similarly to USDA generic HACCP plans (USDA 1999, 2004) that assign a "location" to each traditional CCP with a number and letter (e.g., 3B represents CCP#3 and a biological hazard) in the first column of the traditional HACCP plan, the RB-HACCP plans include a priority level under the RB-CCP column based on the results of the sensitivity analysis for *L. monocytogenes*. Thus, the RB-CCPs for frankfurters were grouped into five categories and numbered 1RB through 5RB in accordance with their priority level based on the sensitivity analysis. For example, the RB-CCP with the highest priority was the "consumer module" (1RB), which included, in the case of frankfurters, three subcategories: duration of refrigerated storage, temperature of refrigerated storage, and reheating log reduction (FR). It is noteworthy that the priority level described above is not a classification of CCPs, as previous revisions of the HACCP system in 1990 by NACMCF rejected the use of a two-class CCP system (Buchanan 1990).

The second column of the RB-HACCP plan form was called "specific significant hazard(s)", which in the case of this study referred to *L. monocytogenes.*

The third column, risk-based critical limits (RB-CLs), shows the possibility of having risk-based management metrics (i.e., performance objectives [POs], performance criteria [PCs], food safety objective [FSO]), as well as process criteria (PcC), parameters, values, and/or distributions depending on the RB-CCP. For example, it is expected that the reheating log reduction at the consumer level fulfils the temperatures and times that would result in a log reduction following the distribution: RiskCumul(4.49,6.68,{5.3,6.18,6.68},{0.75,0.95,0.99},RiskName("Reheating Log Reduction")).

This cumulative distribution was created based on data from the "Quantitative Assessment of Relative Risk to Public Health from Foodborne *Listeria monocytogenes* among Selected Categories of Ready-to-Eat Foods" (FDA/USDA 2003).

All of the other columns of the RB-HACCP plans were, in principle, very similar to the traditional HACCP plan form. The fourth, fifth, sixth, and seventh columns correspond to monitoring to determine what, how, frequency, and who should monitor each particular RB-CCP. An example of a monitoring activity would be "hold and test" of the final product. This is considered a monitoring activity and not a verification activity (Buchanan and Schaffner 2015). The eighth column corresponds to corrective actions which in the case of the RB-HACCP would likely be lower in number because changes in the input variables of the process could be accessed in real time to determine the effects on the outputs, resulting in a proactive hands-on system to address potential deviations of the process in a timely manner. The ninth column "verification" in regards to the risk-based system as it relates to *L. monocytogenes* could also be referred to as process control verification testing of finished product

(sometimes referred to as “cross-lot” or “between-lot” testing), which has been used to detect changing patterns or trends of contamination. This type of testing allows differentiation between occasional “in control” positive samples and an emerging loss of control. The presence of *L. monocytogenes* in the finished product can also indicate the lack of control of *L. monocytogenes* in the processing environment, which needs to be addressed with a *Listeria* environmental monitoring plan. The tenth and final column corresponds to record keeping. Although this column is the same as in the traditional HACCP plan, in a RB-HACCP plan some records could be managed more efficiently from an Excel spreadsheet connected to the QMRA model. It is noteworthy that using spreadsheet software for predictive microbiology applications has been proposed in the past (Buchanan 1991, 1993a, 1993b, 1993c) including practical applications to HACCP plans (Fujikawa and Kokubo 2001).

7.2.1.3 Comparison between traditional and risk-based HACCP plans for frankfurters

A comparison between the critical control points (CCPs) from two traditional HACCP plans and the risk-based CCPs (RB-CCPs) for the frankfurters not reheated (FNR) baseline scenario and the frankfurters reheated (FR) scenario is depicted in Table 7.3. The two traditional HACCP plans included the one developed by the visited facility specifically for their frankfurter production and the USDA generic HACCP plan for fully cooked, not shelf stable meat and poultry products (USDA 1999) which was developed for ham and roast beef and was not specific for frankfurters. The traditional HACCP plan developed by the facility visited only had the thermal treatment of frankfurters as a CCP (see section 7.2.1.1), whereas the USDA generic HACCP plan for fully cooked, not shelf stable meat and poultry products (USDA 1999) had receiving

raw meat, storage of raw meat, cooking, chilling, portioning, packaging/labeling, and finished product storage as CCPs. The RB-CCPs obtained from the sensitivity analysis were the following: initial level of *L. monocytogenes* in meat pre-blend, temperature of the thermal treatment at processing, duration of thermal treatment at processing, handling at packaging/portioning, duration of consumer-refrigerated storage, temperature of consumer-refrigerated storage (open/unopened packages), duration and temperature during retail distribution, duration of transportation to retail distribution, duration of transportation from retail to consumer, and reheating log reduction at consumer level (FR). Table 7.3 compares the CCPs from the traditional HACCP plans and the RB-CCPs from the RB-HACCP plan.

Although the risk assessment derived HACCP plan (RB-HACCP) included equivalent RB-CCPs for all the CCPs from the USDA generic HACCP plan for fully cooked non-shelf stable meat and poultry products (USDA 1999) and additional RB-CCPs, the chilling step was not found to be an RB-CCP, based on the modeled conditions. The chilling step was modeled based on the specific conditions at the visited facility. As explained in Chapter 4, the frankfurters were chilled together with their artificial casings by submersion in acidified brine immediately before being peeled. The steamed peeling step implied the removal of the artificial casings using a heated blade, which is the only part of the peeler that could be considered a contact surface. For simplification purposes, it was assumed that the artificial casings were the only part of the product in contact with the pasteurized brine. In addition, this particular facility used pasteurized brine with citric acid at >0.5M which has listeriostatic activity (Young and Foegeding 1993; Buchanan and Golden 1994). Furthermore, the pasteurized brine

was maintained at low temperatures (24°F) resulting in an exit core temperature of 30°F (-1.1°C). This exit core temperature was achieved through the automated system in 14.5 minutes. Although it is unlikely under the above-mentioned conditions to experience a growth or recontamination problem with *L. monocytogenes* in this particular step under the specified conditions, the rate of chilling could be a potential food safety concern that should be addressed in an HACCP plan, especially for fully cooked products with larger diameters. For example, although frankfurters chill quickly because of their small diameter, the examples for the USDA generic HACCP plans for heat-treated not shelf stable meat and poultry products (USDA 1999) require chilling ham and roast beef from "120°F to 55°F within 6 hours, and chilling to continue to 40°F."

Similar to brine chilling, peeling could represent an essential step if control measures, adequate sanitary designed equipment, and/or the necessary precautions are not in place. In the case of the facility visited after which the frankfurter process was modeled, peeling was performed using an automated peeler with steam and the only food contact surface at this step was a heated blade. In facilities with peelers that do not include steam and a heated blade, inoculation of each frankfurter could occur as they are being peeled (Wenger and others 1990). Although the peeling step did not show as RB-CCP in the case of the modeled process based on the above-explained conditions at the frankfurter facility visited, the peeling step could become essential (e.g., sanitation preventive control) in facilities with different types of peelers and at least one without steam, sanitary designed, and most importantly, a heated blade. Thus, the RB-HACCP plan developed for frankfurters is particular for a specific processing

line within a process. It would be possible, however, to develop risk assessment modules for certain modules including unit operations or steps in the process that could be later assembled to follow the sequence of a different processing line for a specific pathogen. Well-trained personnel, consultants, inspectors, and/or regulators are indispensable to assure that a RB-HACCP plan includes all pertinent risk-based CCPs and preventive controls, even if they are not shown in the sensitivity analysis or model because they are not likely to cause considerable increases in the mean or variations in the levels of the pathogen of interest for a particular facility at the specified conditions. RB-HACCP plans could also help with the design or validation of PCHF-type Food Safety Plans.

7.2.2 Case Study Two—Risk-based HACCP plan for cold-smoked salmon compared to a traditional HACCP plan

This section presents a traditional HACCP plan for cold-smoked salmon (CSS) (section 7.2.2.1), a risk assessment-derived HACCP plan for CSS (section 7.2.2.2), and a comparison between traditional and risk-based HACCP plans for CSS (section 7.2.2.3).

7.2.2.1 Traditional HACCP plans for cold-smoked salmon

A traditional HACCP plan for CSS was developed during the group exercise at the end of segment two of the Seafood HACCP Alliance course from the Association of Food and Drug Officials (AFDO) in accordance with the Seafood HACCP Training Curriculum (National Seafood HACCP Alliance 2011) and the Fish and Fishery Products Hazards and Controls Guidance (FDA 2011). This course included a review of Title 21 CFR 123 (a Fish and Fishery Product), hazard analysis procedures, and

practical exercises for the completion and development of a HACCP plan, among other topics. The course satisfies the mandatory training requirement contained in 21CFR, part 123.10.

The traditional HACCP plan was originally developed for hot-smoked salmon together with a randomly selected group composed mainly of seafood industry personnel during the previously described course in September 2016. This HACCP plan was then compared with the latest version (December 2016) of the "hot-smoked salmon reduced oxygen packed HACCP model" (National Seafood HACCP Alliance 2016) and adapted for a CSS processing line. In addition, the impact of the GMP revisions (117/21 CFR 117, Subpart B) on the Seafood HACCP Training Curriculum (National Seafood HACCP Alliance 2017a, 2017b) was considered as part of the additional requirements added to or modified in the Seafood HACCP Regulation in response to passage of the Food Safety Modernization Act (FSMA). Table 7.4 shows this traditional HACCP plan form for CSS developed during the Seafood HACCP Alliance/AFDO course as described above. The mention of "pathogenic bacteria growth temperature abuse" (as advised by the Seafood HACCP Training Curriculum, and the lack of specific pathogens such as *L. monocytogenes*) under the "significant hazards" column is noteworthy. The identified Critical Control Points (CCPs) included: brine, cold smoking, vacuum pack/label, and finished product refrigerated storage. In addition to the "Segment Two HACCP plan" for CSS as described above (unpublished, developed 2016), another traditional HACCP plan was included in this research (unpublished, dated 1996). This HACCP plan had the original title, "Smoked fish processing HACCP plan form" (B. Blakistone, formerly with the National Fisheries

Institute, pers. comm., March 15, 2013), however, since the author was not specified in the document and to simplify its reference, it was referred to as "CSS Traditional HACCP plan" (Anonymous 1996). Section 7.2.2.3 covers a comparison of these two traditional HACCP plans and the risk-based HACCP plan for CSS.

7.2.2.2 Risk-based HACCP plans for cold-smoked salmon

The risk assessment derived HACCP plan (RB-HACCP) for CSS was developed based on the results of the sensitivity analysis (i.e., risk-based critical control points) for the CSS model. Table 7.5 shows the CSS RB-HACCP plan form for *L. monocytogenes*. The layout of the RB-HACCP plan form is similar to that of the traditional HACCP plan form as adopted in 1997 by the National Advisory Committee of Microbiological Criteria for Foods (NACMCF 1998). However, the first column includes the RB-CCPs instead of the CCPs as previously mentioned in section 7.2.1.2. Similarly to USDA generic HACCP plans (USDA 1999, 2004), which assign a "location" to each traditional CCP with a number and letter (e.g., 3B represents CCP#3 and a biological hazard) in the first column of the traditional HACCP plan, the risk-based HACCP plans include a priority level under the RB-CCP column based on the results of the sensitivity analysis. Thus, the RB-CCPs for CSS were grouped in accordance with their priority levels in six categories and numbered 1RB through 6RB. For example, the RB-CCP with the highest priority level was the "consumer module" (1RB), which included the duration and the temperature of the refrigerated storage of CSS at the consumer level.

The second column of the RB-HACCP plan form was called "specific significant hazard(s)" instead of "significant hazard(s)." The word "specific" was added because sometimes traditional HACCP plans include a generic description of the

hazards and not scientific names of pathogens. For example, although other pathogens such as *C. botulinum* or *S. aureus* are clearly identified with their scientific names as "significant hazards" in traditional HACCP plans, *L. monocytogenes* was not specifically mentioned in the studied traditional HACCP plans, and was only grouped under "pathogenic bacteria growth, survival, and/or recontamination" categories (see FDA 2011, Table 3-4, p. 71). Since the hazard of interest in this study was *L. monocytogenes*, the second column of the risk-based HACCP plan mentions specifically this pathogen. Including *L. monocytogenes* specifically would advance HACCP plans by addressing the particular intrinsic and extrinsic characteristics related to particular food-pathogen pairs (e.g., growth under refrigeration), its challenges in the processing environment, and its potential for causing severe health outcomes in at-risk populations.

The third column of the RB-HACCP, risk-based critical limits (RB-CLs), shows the possibility of having parameters, values, performance objectives (POs), performance criteria (PCs), food safety objective (FSO), and/or distributions depending on the RB-CCP. While the RB-CCP results for both case studies were analyzed in detail in Chapter 6, the RB-CLs were implied as the values obtained for the most stringent scenarios. These values were included in the corresponding sections of the RB-HACCP plans for frankfurters and CSS (see Tables 7.2 and 7.5, respectively). For example, in the case of duration of the refrigerated storage, three scenarios achieved a low risk for listeriosis, and thus determined risk derived critical limits based on public health impact. These scenarios achieved reductions of 1%, 10%, and 50% of the duration of the refrigerated storage, producing RB-CLs of 3.4 hours, 1.4 days, or one week,

respectively. Thus, to lower the risk of listeriosis in at-risk populations, CSS modeled under the specified conditions needs to be stored under refrigeration for a maximum of one week after purchase. From an international perspective, Ross (2010) suggested that a shelf life of 3-8 days, depending on the assumptions made in the model, could usually be expected to limit *L. monocytogenes* to less than 100 cfu per gram on RTE foods that support the growth of this pathogen up to the time of consumption. If longer shelf life is needed at the consumer level, it would be advisable to freeze the product as this scenario (scenario 23) achieved the lowest risk of listeriosis. This is in agreement with Gallagher and others (2016) who also found that the consumer level is critical to deliver risk mitigation strategies that could lower the risk of listeriosis.

The following columns of the RB-HACCP plans include monitoring (what, how, frequency, who), corrective actions, verification, and records. As previously explained, they were found to be in principle very similar to the traditional HACCP plan form. The fourth, fifth, sixth, and seventh columns correspond to monitoring each RB-CCP, and are particular to each food processing facility. An example of a monitoring activity would be “hold and test” for the final product. It is noteworthy that this is considered a monitoring and not a verification activity (Buchanan and Schaffner 2015). The presence of *L. monocytogenes* in the finished product can also indicate the lack of control of *L. monocytogenes* in the processing environment, which needs to be addressed with a *Listeria* environmental monitoring plan. The eighth column corresponds to corrective action(s), which in the case of the RB-HACCP would likely be lower in number because changes in the input variables of the process could be accessed in real time to determine the effects on the outputs, resulting in a proactive

hands-on system to address potential deviations of the process in a timely manner. The ninth column "verification" for the risk-based system as it relates to *L. monocytogenes* could also be referred to as process control verification testing of the finished product (sometimes referred to as "cross-lot" or "between-lot" testing), which has been used to detect changing patterns or trends of contamination. This type of testing could allow a distinction between occasional "in control" positive samples and an emerging loss of control. This ongoing evaluation of collected data over time could result in corrective action investigations even before a loss of control occurs. The specific testing requirements of the process control sampling plan depend on the type of process control analysis approach being employed (e.g., CUSUM, Moving Window) (ICMSF 2002). Various statistical process control (SPC) tools have been applied to the food industry to enhance the collection and analysis of HACCP data (Buchanan and Williams 2013). For example, Hayes and others (1997) used CUSUM and Individual charts to obtain trend analysis and advance warnings in a dairy operation to prevent the failure of CCPs. Tokatli and others (2005) demonstrated the utility of multivariate process monitoring and fault diagnosis techniques to HACCP programs involving food pasteurization processes. Srikaeo and Hourigan (2002) demonstrated that SPC techniques could be used to enhance the validation of CCPs related to shell egg washing. Srikaeo and others (2005) used SPC techniques to examine biscuit baking and found that a number of the parameters that influenced the adequacy of the baking process were not in control. Although SPC of microbiological data provides a vital additional aid to monitoring changes in a manufacturing process, end-point testing of manufactured foods is effective only as a means of retrospective monitoring of process, distribution, and

storage conditions. The distribution of organisms in foods and the statistical variation associated with methods of detection and enumeration lead to the conclusion that, at present, microbiological criteria (MC) should be used primarily as guidelines and specifications, except for the monitoring of high-level contamination with pathogens (Jarvis 2016).

The tenth and final column corresponds to records. Although this column is the same as in the traditional HACCP plan, in a risk-based HACCP plan some records could be managed more efficiently directly from an Excel spreadsheet, as previously suggested. In addition, managing the data and records generated by HACCP programs can be a major challenge. However, considering the expense associated with the generation and archiving of the data, there are often minimal attempts to mine the data for additional information, i.e., if the monitoring or verification activity is satisfactory, it is recorded, archived, and ignored unless there is a problem later. However, this data can provide important information if properly analyzed and arrayed (Buchanan and Williams 2013), which could be facilitated by using directly a HACCP spreadsheet. The use of spreadsheet software combined with assessment tools (e.g., @Risk, SPC) could enhance HACCP not only to store records more efficiently and make them readily available for further analyses but also to serve as a hands-on tool at the food processing level.

7.2.2.3 Comparison between traditional and risk-based HACCP plans for cold-smoked salmon

A comparison between the CCPs from the two traditional HACCP plans described in section 7.2.2.1 and the RB-CCPs for the CSS baseline scenario and scenarios 20, 21, 22, and 23 was depicted in Table 7.6. The comparison shows that all

CCPs found in the traditional “Segment Two HACCP plan” for CSS (unpublished, developed 2016) were included in the “CSS Traditional HACCP plan” (unpublished, dated 1996, shared by B. Blakistone, pers. comm., March 15, 2013) except for thawing (see Table 7.4), which the current FDA (2011) guidelines do not recognize as a CCP. The RB-CCPs for the baseline and the selected scenarios included all the CCPs from the “Segment Two HACCP plan” and other RB-CCPs, but not thawing. The other RB-CCPs included in the CSS baseline scenario as a result of the sensitivity analysis were the temperature of the consumer storage, the duration of the consumer storage, the slicing step of the process, the temperature during the retail distribution, and the duration of the retail distribution. For more details regarding each selected scenario results, refer to Table 7.6.

In addition, labeling is typically considered to be a critical control point (CCP) for CSS due to the fact that fish is considered an allergen (National Seafood HACCP Alliance 2011, 2017a) with the potential to cause “cross contact” (National Seafood HACCP Alliance 2017a, 2017b). Although labeling did not appear as a critical factor in the tornado graphs with regards to *L. monocytogenes*, it was considered an essential intervention in the consumer module in the RB-HACCP specific for *L. monocytogenes*, as it would be critical for consumers to have clear labeling storage instructions such as temperature and duration of the storage, expiration dates, preparation instructions prior to consumption, thawing instructions, and other relevant information.

7.2.3 Comparison of risk-based HACCP plans for frankfurters and cold-smoked salmon

The risk-based critical control points (RB-CCPs) for frankfurters and cold-smoked salmon (CSS) are summarized in Table 7.7. The table includes the RB-CCPs for the baseline models for both products as well as relevant selected scenarios. Five main RB-CCP categories were created based on the results found for both ready-to-eat (RTE) products. Although not identical, these categories shared commonalities that are noteworthy. These categories were summarized in accordance with the process sequence (see Table 7.7) but following the level of priority ("RB") found by the sensitivity analysis: (1RB) consumer module, (2RB) food contact surfaces, (3RB) retail distribution module, (4RB) thermal treatment/smoking at processing, and (5RB) raw product initial contamination. An additional category named "Other RB-CCPs" was created to compile all other RB-CCPs that were particular to one of the products (e.g., CSS brining (6RB)).

The five main categories included commonalities between the processes for the two selected RTE products. For example, the category "raw product initial contamination" includes: "initial level of *L. monocytogenes* in meat" for the frankfurter case study, and "raw salmon *L. monocytogenes* incidence" for the CSS case study. The sensitivity analysis for all the scenarios for both products found the category summarized "raw product initial contamination" to be a RB-CCP in all cases. For a detailed account of commonalities between these two products, see Table 7.7. An explanation of each of the summarized categories follows:

(1RB) consumer module: The consumer module category refers to the "time/duration of the consumer-refrigerated storage" and the "temperature of the consumer-

refrigerated storage." These two parameters were consistently found to be the main RB-CCPs for both RTE products. In addition, in the case of the frankfurter reheated (FR) scenario, the reheating log reduction at the consumer level was also found to be a RB-CCP. Thus, applying control measures such as reducing the mean and variance of key parameters (i.e., temperature and durations of storage) resulted in an effectively lower level of *L. monocytogenes* for the more stringent selected scenarios.

(2RB) food contact surfaces: This category refers to "handling at packaging" in the case of the frankfurters model, and to "slicing," "portioning," and "pinbone" in the case of the CSS model. Although these steps of the process are different in nature, they all occur after the thermal treatment/smoking and before the final product is packaged. Each of these steps involves direct contact of the final product with surfaces that have the potential to recontaminate the product. It is noteworthy that "handling at packaging" involves the manual selection of frankfurters using stainless steel mesh gloves by the operators to protect their fingers from the automated equipment. The operators remove broken frankfurters immediately prior to portioning and subsequent packaging at the end of the automated processing line while the conveyor belts are running. Thus, this step involves direct contact with the product and includes the portioning in packages of ten frankfurters each. It is noteworthy that the frankfurters processing facility was, for the most part, automated; by contrast, the CSS processing facility was not automated and required more operators using latex/nitrile gloves for the portioning and pinbone steps of the process. Although the slicing was semi-automated, there were two processing lines, one with old equipment and one with up-to-date equipment. Thus, the sanitary design conditions of these two slicers differed considerably.

(3RB) retail distribution module: The retail distribution category refers to the "time/duration during retail distribution refrigerated storage" and the "temperature during retail distribution refrigerated storage." These two conditions were consistently found as RB-CCPs for both RTE products.

(4RB) thermal treatment/smoking at processing: The thermal treatment/smoking category showed the temperature and time/duration of the thermal treatment at processing as the main RB-CCP for all frankfurter scenarios. In like manner, cold smoking, which involves a mild thermal treatment as well as other chemical changes (e.g., smoking with phenolic compounds) within the unit operation, was found to be an RB-CCP for all selected CSS scenarios.

(5RB) raw product initial contamination: This category refers to the incoming level of *L. monocytogenes* in both the raw meat pre-blend and the raw salmon. In the case of frankfurters, the "initial level of *L. monocytogenes* in cfu/g" was found to be the main RB-CCP at the ingredient module level. The frankfurters model was truncated, thus this step does not appear in subsequent tornadoes. However, it is an important step to consider. In similar fashion, the equivalent RB-CCP found for the CSS model was the "raw salmon Lm incidence" which was consistently found as an RB-CCP in all selected scenarios and refers to the incoming level of contamination with *L. monocytogenes*.

(6RB) other risk-based critical control points: The risk-based critical control points (RB-CCPs) included in this category were particular for each ready-to-eat (RTE) product. In other words, the "Other RB-CCPs" category compiles the RB-CCPs that were found for one but not the other RTE product of study. For example, "Other RB-CCPs" included the duration of transportation from processing to retail, and the

duration of the transportation from retail distribution to the consumer refrigerator for the frankfurter process and brining for the CSS process.

Buchanan and Whiting (1998) suggested that while HACCP systems work very well for microbiological hazards when there is an intervention step that provides a large reduction in the target microorganism (e.g., pasteurized milk, commercial sterilization of canned foods, cooked meats), it is less effective for foods that receive little if any reduction in microbiological populations. Instead, such foods (e.g., fresh produce, cold-smoked fish, fermented dairy products made from raw milk) rely on a series of controls that either partially reduce microbial populations or delay the growth of pathogenic microorganisms. Consequently, the ability to determine which steps in the manufacturing process are CCPs becomes much more difficult, particularly if there is substantial variability in the ingredients and processes. Typically, this results in a HACCP plan that is considerably more complex in terms of the number of identified CCPs (Buchanan and Whiting 1998). These statements were found to be true while comparing the results of the sensitivity analysis (SA) for the frankfurters and cold-smoked salmon processes. For example, the SA results identified more RB-CCPs in the cold-smoked salmon than in the frankfurter process.

7.2.4 Relating risk-based HACCP plans to food safety risk management metrics

The concept of food safety risk management metrics via the establishment of food safety objectives (FSOs) provides a link between public health outcomes and parameters that can be measured and controlled by food manufacturers and their regulatory agencies (ICMSF 2002). The original idea to overcome HACCP's inability to link stringency directly to public health impacts was developed by Buchanan and

Whiting (1998). This idea was used in this dissertation to develop risk-based HACCP (RB-HACCP) plans. For this purpose, risk assessment modeling techniques were used and combined with predictive microbiology (Buchanan 1992; Buchanan and Whiting 1996; Buchanan and others 1997; Whiting and Buchanan 1997; Buchanan and others 1998; Buchanan and Phillips 2000) and the FSO paradigm (ICMSF 2002; FAO/WHO 2006; Whiting and others 2006; van Schothorst and others 2009). The use of various risk assessment modeling tools has been of interest to a number of academic researchers who envisioned their application for identifying CCPs and CLs. For example, Domenech and others (2008) assessed the effectiveness of CCPs within a risk framework to more effectively identify CCPs and provide analyses needed to make informed decisions regarding CLs. Tenenhaus-Aziza and others (2014) used a case study, *L. monocytogenes* in soft cheese made from pasteurized milk, to show how QMRA could be used to direct potential intervention strategies at different food processing steps. Based on many assumptions, their model developed estimated the risk of listeriosis at the moment of consumption. SA and what-if scenarios were used and allowed for the identification of major parameters contributing to the risk of listeriosis. Using the same case study, Lamboni and others (2014) proposed a methodology, called multivariate factor mapping (MFM), that employed multivariate sensitivity analysis to identify the CCPs and CLs.

Using two case studies for RTE food products, frankfurters and cold-smoked salmon, the present research employed product pathogen pathway risk assessment models with a systems thinking approach in conjunction with sensitivity analyses and what-if scenarios to assist in the identification and prioritization of CCPs and the

establishment of CLs. This dissertation also presented risk-based HACCP plans for each of these RTE food products as they relate to *L. monocytogenes* and compared them with traditional HACCP plans. Risk assessment and systems modeling techniques were able to assess the relative importance of a series of control points, thereby establishing a means of assessing and prioritizing these points as critical control measures (Buchanan and Williams 2013). In addition, the use of systems modeling achieved a much higher degree of evidence to support the selection of CCPs than the current system of using a qualitative hazard analysis and simple decision trees. In this regard, the risk-based approach developed in this dissertation could be used and adapted by regulators to validate CCPs, HACCP plans, and/or PCHF food safety plans at different RTE facilities.

It is important to emphasize that the key food safety risk management metric that directly relates to the RB-HACCP plans is the FSO, which in turn could be used to define other risk-based metrics such as performance objectives (PO) and performance criteria (PC) along with more traditional food safety metrics such as microbiological criteria (MC), process criteria (PcC), and product criteria (PdC) (ICMSF 2002; Walls and Buchanan 2005; CAC 2008a). This section related the risk-based critical control points (RB-CCPs) to the FSO/PO paradigm (section 7.2.4.1), compared related microbiological criteria and risk-based sampling plans (section 7.2.4.2), related the stringency of RB-HACCP plans to MC (section 7.2.4.3), and compared related risk-based regulatory requirements (section 7.2.4.4).

7.2.4.1 Relating risk-based critical control points to the food safety objective/performance objective paradigm

The food safety objective/performance objective (FSO/PO) conceptual equation (ICMSF 2002) provided the underlying framework upon which the food safety risk management metrics were based in this study, as follows:

$$H_0 - \Sigma R + \Sigma I \leq FSO$$

Where:

H_0 = Initial level of the hazard

ΣR = Total (cumulative) reduction in level of the hazard

ΣI = Total (cumulative) increase in level of the hazard

FSO = Food Safety Objective

FSO, H_0, ΣR, and ΣI are expressed in $\log_{10}$ units

The ICMSF equation allowed the production pathway to be viewed as a series of inputs and outcomes that were consistent with a systems thinking approach. Given the complex production and cold chain requirements of ready-to-eat (RTE) products, the risk-based HACCP plans developed using QMRA models used a system-thinking approach. This general approach was used to describe individual subsystems (modules) to model the whole process and obtain, at the end of each module, an output that would identify the most critical factors. The output of the model for a module served as the input (H0) of the subsequent module. Using @Risk sensitivity analysis capabilities, the most critical parameters in each module were identified and considered to be the risk-based critical control points (RB-CCPs) associated with the two ready-to-eat (RTE) food products. Beginning with the latest version of @RISK, in addition to the “Inputs ranked by effect on output mean” tornado, @RISK 7.5’s has been updated with a “Contribution to variance” tornado graph capability that helps

explain how much of the variance in the output variable is attributable to each individual input (Palisade Corporation 2017). This is directly related to the working hypotheses explained in Chapter 2. Both of these specific tornadoes were selected for each module of the RTE products as explained and shown in Chapter 6.

In regards to linking RB-CCPs with public health goals, a risk ranking of the twenty-four CSS scenarios was conducted (see Table 6.6 in Chapter 6) based on criteria described by Carrington and others (2004) as <1 case per billion servings. This risk ranking and back-calculation allowed for the identification of the scenarios that "made the cut" to be defined as low risk for listeriosis. Chapter 6 also includes the description of the what-if scenarios for both RTE products. A summary of the frankfurters and CSS what-if scenario analyses can be found in Tables 6.1 and 6.4, respectively. Figures 7.1 and 7.2 depict a detail representation of the modules, including the steps of the process and the exact location of the RB-CCPs within each module for the frankfurters and CSS processes, respectively. The mean levels of *L. monocytogenes* (log CFU per gram) throughout each module of the process for the frankfurters not reheated (FNR) baseline scenario as well as for the frankfurters reheated (FR) scenario are shown in Figure 7.3. Similarly, the mean levels of *L. monocytogenes* (log CFU per gram) for the baseline model (scenario 1) compared to selected stringent what-if scenarios (scenarios 20, 21, 22, and 23) throughout each module of the CSS process are shown in Figure 7.4. This data shows the sensitivity of the modules, while the identification of the specific processes that need to be controlled was shown in Chapter 6.

While on one hand the food safety objective/performance objective (FSO/PO) paradigm relates the stringency of a food safety management system to its intended

public health outcome (Buchanan 2013), on the other hand the FSO relates to the food process itself via derived POs. These POs could be aligned with particular risk-based critical control points (RB-CCPs) identified by the sensitivity analysis. The location of the risk management metrics, such as FSOs at the consumer level and potential POs throughout the frankfurter and CSS processes, was shown in Figures 7.5 and 7.6, respectively. Figures 7.5 and 7.6 also include the prioritized RB-CCPs but, most importantly, show the prioritized corresponding food safety risk management metrics (i.e., food safety objective [FSO], performance objective [PO], performance criterion [PC], and microbiological criterion [MC]) in the systems approach modules for the frankfurter and CSS processes, respectively. Although each RB-CCP as identified by the sensitivity analysis (SA) could have a corresponding PO as represented in Figures 7.5 and 7.6, "final product POs" were determined and selected for frankfurters and CSS as shown in figures 7.7 and 7.8, respectively. It is important to note that in both case studies the selected POs were located at the end of the production line (PO2) and at retail distribution (PO3). In addition, for the CSS model, receiving raw salmon (PO5) was also found to be highly relevant as the cold-smoking step typically provides 1 log reduction (ICMSF 2011) compared to the cooking step of frankfurters which provides an overwhelming process lethality (>6D) for *L. monocytogenes*.

The detailed point estimate approach analyses of the FSO equations and comparison for each of the scenarios that met the FSO were summarized for the frankfurters and CSS scenarios in Tables 7.8 and 7.9, respectively. In addition, Table 7.8 summarizes the calculations for the frankfurter not reheated (FNR) and frankfurter

reheated (FR) scenarios after the thermal treatment at the processing level (model truncated) with respect to the components of the ICMSF equation at the consumer module level (PO1) for the mean levels of *L. monocytogenes* (log CFU per gram). Using the point estimate approach mean values of FNR and FR (without considering variability) for *L. monocytogenes* at the consumer level, the baseline scenario (FNR) resulted in 1.764 mean log CFU per gram whereas the reheated frankfurters (FR) at the consumer level resulted in -3.223 mean log CFU per gram of *L. monocytogenes*.

Table 7.9 summarizes the calculations for the CSS baseline model (scenario 1) as well as other selected more stringent scenarios which, based on the point estimate approach mean log CFU per gram values, were scenarios 20, 21, 22, and 23. It is important to note that scenario 23, representing the freezing of the product at the consumer level, achieved a PO1 of -1.716 mean log CFU per gram, which was the most stringent value. In addition, each of the scenarios 20, 21, and 22 achieved a PO1 of 0.229, -1.270, and -1.607, respectively. These values represent a reduction in the duration of the storage at the consumer level of 50%, 10%, and 1% of the original time/duration, respectively. As already noted, the maximum duration of storage under refrigeration to achieve a low risk of listeriosis (without considering variability) was calculated as 7 days for susceptible populations and the specific conditions of the model.

7.2.4.2 Relating the stringency of risk-based HACCP plans to microbiological criteria

While the concepts of sampling plans and microbiological criteria (MC) for foods have been proposed for decades (ICMSF 1986; ICMSF 1994), the efficiency of

a laboratory examination for pathogens or spoilage organisms affects the number and size of samples examined and the distribution of organisms in a product (Jarvis 2016). Zwietering and others (2016) have stressed the importance of understanding the nature of over-dispersed populations of pathogens in relation to food safety. The recent FAO/WHO (2016) publication provides a clear and extended description of MC related issues and a framework for evaluation of different strategies for the development and assessment of MC. However, the FAO/WHO (2016) publication does not relate MC with the stringency of a risk-based HACCP provided by the FSO/PO paradigm. The first use of the FSO/PO concepts have largely been around the development of risk-based microbiological criteria (MC) by the Codex Alimentarius Commission (CAC), having been used in the development of new MC for *Cronobacter sakazakii* in powdered infant formula (CAC 2008b), and *L. monocytogenes* in ready-to-eat (RTE) foods (CAC 2007a, 2009a). The CAC criteria for *L. monocytogenes* in RTE food (See Table 7.10) were developed through the step-wise consensus process within the Codex Committee for Food Hygiene (CCFH), including findings from the FAO/WHO (2004) quantitative risk assessment for *L. monocytogenes*. Previous qualitative assessments (ICMSF 1994, 2002) applicable to *L. monocytogenes* and susceptible populations "ICMSF cases" (selected cases 13-15) require a greater number of samples (n) than the Codex criteria for *L. monocytogenes* in RTE food (CAC 2007a, 2009) (see Table 7.11). However, the CAC microbiological criterion for RTE food products that support growth of *L. monocytogenes* was also recommended by ICMSF (2011) for the corresponding product categories (See Table 7.12). The Codex criterion uses 5 samples and has a stringent limit of absence in 25 g for each analytical unit. This criterion would

be able to reject a lot with a geometric mean concentration of 1 CFU in 55 g with 95% confidence (assuming a standard deviation of 0.8 log CFU/g). It should be noted that Codex used a standard deviation of 0.25 log CFU/g, whereas different standard deviations were used for the sampling plans calculations using output values for selected performance objectives (POs) from both case studies QMRA models (i.e., frankfurters and CSS). The standard deviations used were 0.2, 0.5, or 0.7 log CFU/g for frankfurters depending on the POs and 0.8 log CFU/g for CSS (See Table 7.13). The potential risk-based sampling plans depicted in Table 7.13 could be used to verify with a specified degree of confidence (P_{rej} 0.95) that the selected POs are not exceeded at the end of manufacture or retail distribution/point of sale. The ICMSF (2014, 2016) sampling tool was used to derive these calculations (See Table 7.13). These calculations relate the stringency of the risk-based HACCP plans (i.e., verification) to microbiological criteria. For example, for the CSS model, the calculated means at the end of the processing line and at the end of retail distribution of: -2.094 and -1.644 log CFU per gram, respectively, were used as inputs for the ICMSF (2014, 2016) sampling tool, assuming a standard deviation of 0.8 log CFU per gram in each case and the following conditions: m= absence in 25g (<0.04 cfu/g), c=0, and 2-class plan. The results obtained (i.e., n=9 and n=5, respectively) would have the following interpretation resulting from the ICMSF (2014) sampling tool: The sampling plan with an input mean of -2.094 log CFU per gram (CSS end of processing line) “would provide 97% confidence that a lot of food containing a median concentration of 1 organism in 124.2 g and an average concentration of 1 organism in 22.8 g (and having a standard deviation of 0.80 log cfu/g), would be rejected (i.e., more than 0 out of 9 samples of 25

grams giving detection of the organism).” The sampling plan with an input mean of -1.644388 log CFU per gram (retail distribution level) “would provide 97% confidence that a lot of food containing a median concentration of 1 organism in 44.1 g and an average concentration of 1 organism in 8.1 g (and having a standard deviation of 0.80 log cfu/g), would be rejected (i.e. more than 0 out of 5 samples of 25 grams giving detection of the organism).” Thus, the number of samples (n=5) for PO3 at the retail distribution level are similar to the Codex Alimentarius microbiological criteria for RTE foods in which growth of *L. monocytogenes* can occur (CAC 2007a, 2009).

In a similar manner, in the case of frankfurters, a calculated mean log CFU per gram half way to the retail distribution would have the following results and interpretations depending on the assumed SD. The sampling plan with an input mean of -1.82695 log CFU per gram and SD=0.2 log CFU per gram (frankfurters retail distribution level) “would provide 96% confidence that a lot of food containing a median concentration of 1 organism in 67.1 g and an average concentration of 1 organism in 60.4 g (and having a standard deviation of 0.20 log cfu/g), would be rejected (i.e. more than 0 out of 8 samples of 25 grams giving detection of the organism).” The sampling plan with an input mean of -1.82695 log CFU per gram and SD=0.5 log CFU per gram (frankfurters retail distribution level) “would provide 96% confidence that a lot of food containing a median concentration of 1 organism in 67.1 g and an average concentration of 1 organism in 34.6 g (and having a standard deviation of 0.50 log cfu/g), would be rejected (i.e. more than 0 out of 7 samples of 25 grams giving detection of the organism).”

Vidal and Cueva (2001) found it helpful to include, among other relevant food safety information, the microbiological criteria for *L. monocytogenes* and other pertinent pathogens within the "food safety data sheet" for a ready-to-eat product that supports the growth of *L. monocytogenes*. In a similar manner, the FSPCA (2017) product description form, part of the FSMA Preventive Controls-based food safety plan, was adapted to include, for example, microbiological criteria for *L. monocytogenes* in the case of frankfurters (see Table 7.14). Although frankfurters are a USDA regulated product, including MC in the product description forms may also be pertinent for their jurisdiction. Other information included in the "food safety data sheet" relevant to food safety was the shelf life together with the sampling plan recommended for end-product testing when there is suspicion that the HACCP plan is not performing as intended as recommended by ICMSF (2011). In addition, effective microbiological control comes from use of GMP, HACCP, and other control strategies at all stages of food production, distribution, and storage based on knowledge of the microbial ecology of particular foods under different process and storage conditions. Such control strategies also require effective introduction and monitoring of POs. The potential cost of intensifying testing schemes needs to be balanced against the costs of unnecessarily rejecting valuable food materials and/or of increasing consumer's risk by accepting defective products (Jarvis 2016).

7.2.4.3 Comparing HACCP related traditional and risk-based regulatory requirements

Although HACCP has become universally recognized as the primary system for food safety assurance (WHO 1997, 2017), a number of concerns have been raised about the way HACCP is implemented by industry and employed by regulatory

agencies, including “inadequate HACCP plan specificity for a given operation” and “inconsistencies in the approach taken by the U.S. Department of Agriculture (USDA) and the Food and Drug Administration (FDA) regarding HACCP implementation” (IOM/NRC 2003, p. 251). For example, the USDA generic HACCP model (USDA 1999) recommends a temperature of 158°F achieved instantaneously, whereas FDA (2011) recommends holding the product for 2 minutes at 158°F. Thus, the thermal treatment recommended by the FDA is more stringent than the one recommended by the USDA. With regards to the PRPs in relation to HACCP-based systems, the USDA “requires” the development of a *Listeria* Control Program to test for *L. monocytogenes* or an indicator organism on food contact surfaces (FCS), whereas the FDA “recommends” an environmental monitoring program in their draft guidance. It is noteworthy that this recommendation applies to the CSS case study because this product is under the jurisdiction of FDA (Seafood HACCP). In both cases (FDA and USDA), the detectable presence of *L. monocytogenes* in RTE foods or food contact surface (FCS) may result in the RTE food product being considered adulterated. Because of the FDA policy, most CSS processors do not test their end products for the presence of *L. monocytogenes* (IFT 2001). CSS processors may use environmental sampling in their plants in place of, and as predictors of, the presence of *L. monocytogenes*, although sampling of end products is often avoided (IFT 2001). FDA requests recall of any RTE food in which *L. monocytogenes* is detected using present methodology. FDA (2017) recommends that the listericidal process control established and used in food processes consistently destroy viable cells of *L. monocytogenes* and consistently lead to a food product that does not contain detectable *L. monocytogenes*

using a method that has a limit of sensitivity of detection of at least 0.04 CFU of *L. monocytogenes* per gram of food (<1 CFU per 25 g). This is consistent with the 1985 FDA established policy for *L. monocytogenes*: “detection of the organism in a ready-to-eat food by the FDA method is a violation of the Federal Food, Drug and Cosmetic Act, section 402(a) (1) and (4).” This is further explained in the paper “U.S. position on *Listeria monocytogenes* in Foods” (Shank and others 1996), which mentions that “public health and regulatory agencies in the USA have established zero tolerance for *Listeria monocytogenes* in cooked, ready-to-eat food.” In the case of frankfurters, which fall under the jurisdiction of USDA, the Pathogen reduction, HACCP systems, final rule (USDA 1996) and the *Listeria* rule (USDA 2014a, 2014b) apply and include (among other requirements) the establishment of control alternatives in regards to *L. monocytogenes* contamination of post-lethality exposed RTE products.

Although the Preventive Controls for Human Food (PCFH) final rule applies to products under FDA jurisdiction, it does not apply to CSS, which falls under the Seafood HACCP regulations. The requirement for PCHF within “FDA Food Safety Modernization Act (FSMA)” implies the use of such risk-based tools to prevent foodborne illness and protect public health. The now-required preventive controls incorporate controls beyond those managed as process-related CCPs in the HACCP framework. These preventive controls address not only CCPs, but also controls for hazards related to food allergens, sanitation, suppliers, and others requiring a preventive control (FSPCA 2016). The RB-HACCP plans were specific for *L. monocytogenes* identifying risk-based CCPs (RB-CCPs) quantitatively through sensitivity analyses and what-if scenario analyses. The risk-based HACCP plans

included not only RB-CCPs identified quantitatively through sensitivity analysis and their distributions from the risk assessment as critical limits but also risk management metrics as potential critical limits such as FSO at the consumer level and POs and PCs throughout the processes as depicted in Figures 7.5 and 7.6 for the frankfurter and CSS processes, respectively. For example, the reheating distribution for frankfurters based on the "quantitative assessment of relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat food" FDA/USDA (2003) data was included in the risk-based critical limit column of the RB-HACCP plan for frankfurters (See Table 7.2).

Although traditional HACCP plans are widely implemented, the main weaknesses are that the level of control needed is not stated clearly, and there is little or no guidance on what is expected of an adequately designed and implemented HACCP plan linked to public health outcomes. This omission is widespread in many documents in various sources including governmental regulations. An FSO would indicate the level of control needed for adequate GHP and HACCP systems (ICMSF 2002). This statement continues to be the case for the Preventive Controls final rule. Although it has placed the PRP under a preventive controls category and included more stringent requirements for them, it has not specified an FSO and thus it still needs to evolve into a quantitative system that would finally link public health with food processing control measures, intervention strategies, and/or mitigation strategies. The lack of use of risk-based tools such as quantitative microbial risk assessments in PCHF currently may be hampering the development of effective risk management programs and FSPs.

7.3 *Concluding Remarks on the Risk Assessment Derived HACCP Plans*

This research developed Risk-based Hazard Analysis Critical Control Point (RB-HACCP) plans linking public health goals with the internationally accepted food safety management system, Hazard Analysis Critical Control Point (HACCP), by using risk metrics such as the Food Safety Objective (FSO) equation and quantitative microbial risk assessment (QMRA) tools. These tools included sensitivity analysis (SA) and what-if scenario analyses. Besides the inherent limitations based on assumptions in the current QMRA model, several limitations were identified in regards to SA, and more research is needed to reduce the uncertainty associated with the present model. SA is a recommended practice to assess the robustness of the assessment with respect to uncertain model inputs or assumptions (e.g., EPA 2009). This means that we can ascertain if the inference of a model-based study is robust in light of the uncertainty in the underlying assumptions (Saltelli and D'Hombres 2010). Most importantly, "SA can be applied to learn not only about models but also about systems. If the model reasonably reflects real-world processes, the application of SA to the model can provide insights into the dominant controls of the system" (Pianosi and others 2016). In particular, this study found that it is possible to identify risk-based critical control points (RB-CCPs) by using SA and what-if scenario analyses. RB-CCPs were defined as the steps in the process that effectively reduced the mean and/or variance of a hazard and/or their associated public health risks. Thus, this dissertation demonstrated that QMRA modeling techniques provide an effective means of identifying the critical risk factors influencing the role of manufacturing practices, retail distribution, consumers, and beyond on the risk of *L. monocytogenes*

infections in two ready-to-eat products. The use of these techniques allowed for the development of risk-based HACCP (RB-HACCP) plans. Knowledge of the risk factors within the RB-HACCP plans is critical to the identification of meaningful intervention strategies to truly address with a risk-based approach the connection between the cold chain and public health outcomes. The results obtained from this quantitative risk assessment can be used as an innovative approach in microbial risk assessment models to determine the essential parts of a process that need prioritized attention through SA. On the basis of comparison between the results of the different what-if scenarios simulated, the risk of listeriosis could be effectively lowered if special attention is provided at the consumer level. It is important to recognize that because of the ubiquitous nature of *L. monocytogenes*, this psychrotrophic organism is present in RTE foods being consumed in the U.S. regardless of the zero tolerance policy (Chen and others 2003), the classification of the RTE product, or the utmost efforts made by RTE processing facilities to produce foods free of *L. monocytogenes*. Knowing that the probability of contracting listeriosis from the ingestion of a single cell of *L. monocytogenes* (approximately equal to the *R*-value) is extremely small, it could be reasonably suggested that in the unlikely event of contamination, maintaining low levels of this pathogen all the way through the FSO/consumer level will be likely effective in reducing the public health burden of listeriosis. In this regard, food processing manufacturers have an impact at the consumer level. Thus, there is a shared responsibility for ensuring food safety among consumers and industry. While industry has to achieve measurable POs even at the distribution and marketing levels, consumers need to be diligent to preserve the cold chain of RTE

food products by monitoring refrigerator and freezer temperatures, freezing the product if possible, and minimizing the storage time under refrigeration.

Although frankfurters and CSS are two distinctly different commercial ready-to-eat (RTE) food products, the quantitative risk assessment models for the frankfurters and CSS found not only similar categories of RB-CCPs but also similar priority levels for them (see Table 7.7) based on the SA results for the baseline scenarios of both RTE food products. These five categories in order of priority were (1) consumer module, (2) food contact surfaces, (3) retail distribution module, (4) thermal treatment/smoking at processing, and (5) raw product initial contamination. While these five categories of RB-CCPs and their priority order were found to be similar in the case of the baseline models for these RTE products, the priority order or risk ranking of RB-CCPs are dependent on each scenario and on particular conditions in each processing line, as explained in Chapter 6, and that the RB-CCPs for entire RTE product categories cannot be generalized since each processing line and RTE product have specific unit operations and variability conditions. Although, in some cases, RB-CCPs had variations in their ranking order depending on the specific scenario, the consumer module level was repeatedly seen as the module with the greatest potential to effectively reduce the median number of cases of listeriosis per serving due to the fact that this module has the greatest variability. While the consumer module was found to have the highest priority level based on the SA, there is a tendency to restrict the scope of HACCP to the processing facilities (i.e., before the product is shipped for retail distribution). A risk-based approach needs to be holistic and include all pertinent segments of the food chain, from farm to table. This would allow for the FSO concept

to be used at the facility level. As previously mentioned, the consumer module level for the frankfurter and CSS processes were found to have the highest priority among all other RB-CCPs based on the SA and were ranked as priority number one or "1RB-CCPs"or "1RB" within the RB-HACCP plans. In some cases, more than one "point" within each module was obtained based on the SA. For example, in the consumer module, not only the temperature but also the duration of the storage was found to be an essential "point" or parameter.

Overall, the present QMRA models quantified the risk of *L. monocytogenes* posed by consumption of frankfurters and CSS domestically produced in the U.S. on a per-serving basis in at-risk populations. Both of these models estimated that a considerable number of infection cases associated with the consumption of frankfurters and CSS per serving could be prevented by providing evidence for risk managers that more attention is warranted at the consumer level in regard to *L. monocytogenes*. The present study demonstrated that proper reheating at the consumer level is the most-effective mitigation strategy to reduce the risk of listeriosis from frankfurters. In the case of CSS, proper freezing and thawing of the product at the consumer level is an effective intervention strategy. In addition, proper refrigerated conditions reducing the refrigerated storage duration of CSS to a maximum of one week based on the specific conditions of the process modeled, and controlling the temperature were found to be the most-effective control measures to reduce the risk of listeriosis at the consumer level. Thus, the current risk models for frankfurters and CSS identified not only RB-CCPs throughout the food chain, but also provided a means for comparing the relative effectiveness of control measures that could lower the risk of listeriosis. Although in

the case of frankfurters reheating was identified as an effective mitigation strategy, this may not be the optimal risk management approach due to the propensity for frankfurters to be eaten non-reheated (FNR) by consumers since this product is already fully cooked and thus ready-to-eat. Other mitigation strategies could include reformulation of frankfurters with antimicrobials (Carrington and others 2004), high-pressure processing (Hayman and others 2004; Ferreira and others 2016), labeling of RTE food products with clear storage instructions (Newsome and others 2014), among many other options, including hurdle technology combinations. Section 7.4, which covers future research needs and recommendations, elaborates on potential mitigation strategies that could not only lower the risk of listeriosis but also limit the liability of food companies.

Finally, the present research addressed the two overall goals of this dissertation by (1) recommending the use of sensitivity analysis and what-if scenario analysis to identify "risk-based critical control points (RB-CCPs)" as risk assessment modeling tools that could be used to consider the quantitative impact of risk factors associated with the foodborne hazard of interest, and (2) using those tools to develop a means that allowed food producers to more effectively link their HACCP plans to food safety risk management metrics. The use of food safety risk management metrics, which is based on quantitative science-based evidence, can identify alternative technologies that may be equivalent and that may facilitate achieving the PO in specific selected scenarios for both RTE food products. This is important because an equivalent number of log reductions could be achieved using different technologies, thereby keeping the food industries competitive, flexible, and, more importantly, protecting public health by

allowing the achievement of the final PO and FSO. Thus, while HACCP works in combination with safe product design, PRPs, and an appropriate organizational culture (Wallace and Mortimore 2016), a risk-based HACCP system goes one step further by adding the risk assessment toolbox, specifically using quantitative modeling risk assessment modeling techniques, to provide consumers with an enhanced level of protection that can be measured and thus meet FSOs and public health goals.

7.4 *Future Research Needs and Recommendations*

Despite being recognized as a significant foodborne pathogen and accounting for 255 deaths, 1,455 hospitalizations, and 1,591 illnesses in the U.S. annually (Cartwright and others 2013), *L. monocytogenes* is largely unknown by the average consumer. In particular, "the need to educate consumers about *Listeria* and possible food vehicles exists, so consumers will know to safely handle and store RTE foods such as frankfurters" (Cates and others 2006) and cold-smoked salmon. Furthermore, consumers may have conflicting information about how long they should keep ready-to-eat products under refrigeration due to a lack of clear and specific label information. Indeed, the variation in date labeling terms and uses contributes to substantial misunderstanding by consumers and may lead to potential food safety risks (Newsome and others 2014). Particular attention should be paid to ready-to-eat (RTE) food labeling instructions and freezing option recommendations, especially for at-risk populations such as the elderly, pregnant women, and immune-compromised individuals, in order to help prevent listeriosis. Frankfurters are one of the major consumed meat products in the U.S. and, as such, consumers should be aware of the risks involved if they are not handled properly. Although CSS consumption is lower

than frankfurters, targeted consumer outreach and education in regards to how to properly store both of these RTE food products are essential. Updated studies at the consumer level concerning the application of HACCP to food preparation in domestic and professional kitchens (Griffith and Worsfold 1994; Tuominen and Maijala 2009) could also be beneficial. The development of other quantitative RB-HACCP plans including consumer modules for different food-pathogen pairs could help validate FSMA-type Food Safety Plans (FSPs).

In this research, the public health goal or ALOP considered to determine a low risk level of listeriosis in CSS was based on Carrington and others (2004). However, this degree of stringency could be varied depending on the concerns of society and policy makers and the effectiveness of new technologies (Buchanan 1995). For example, applying high-pressure processing (HPP) could minimize the risk of *L. monocytogenes* after ready-to-eat products are placed in their final packaging. If applied under optimal conditions to minimize the risk of listeriae for each specific food matrix, the HPP technology could be beneficial for most ready-to-eat products. For example, a letter-of-no-objection was issued for the use of HPP at 87,000 psi/600 MPa for a cycle time of 3-27 minutes as part of the Canadian guidance on the use of HPP post-lethality treatment to reduce *L. monocytogenes* levels in RTE meats and poultry products (Health Canada 2012). Optimal HPP post-processing and post-packaging intervention strategies, especially for minimally processed RTE food products, should be developed due to the current trends in food processing in order to reduce the extent of heating, minimize the use of chemical preservatives, and ensure foods that require

little or no preparation or are ready-to-eat and consequently not subjected to heating prior to consumption (ICMSF 2002).

Other new technologies—such as whole-genome sequencing—combined with epidemiological investigations can provide the discriminatory power to elucidate listeriosis outbreaks more effectively and link them to food products and production facilities more efficiently. Using this technology, two listeriosis outbreaks were defined in Denmark during the period 2013–15 (Gillesberg Lassen and others 2016). In 2011, Denmark had the highest rates observed (0.88 per 100,000) among 28 EU/EEA countries with an average case rate of 0.31 per 100,000 population (EFSA 2013). It is noteworthy that Denmark also had the highest yearly estimated consumption per capita of CSS on average among 20 countries (FAO/WHO 2004 p. 256). Cooperation at the national and international level, especially with states and countries with persistent high prevalence of top priority pathogens using the latest technology, is an important task that could benefit not only public health but also the scientific community. This is particularly true for *L. monocytogenes*, which has recently produced outbreaks attributed to foods considered to be "moderate risk" or "low risk" by existing risk assessments, including fruits, vegetables, and even ice cream (Buchanan and others 2017; Salazar and others 2016).

Table 7.1Frankfurters traditional HACCP plan form

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
Temperature during Thermal Processing	*Listeria monocytogenes*	Temperature 158°F	Monitor temperature of finished product in continuous oven with a calibrated thermometer	Randomly select three frankfurters from the hangers in the final cook zone. Measure the temperature in the geometric center of each frankfurter. Average.	Once per hour	Oven operator	If the temperature of product does not attain 158°F, then, 1) the operator notifies his supervisor and the Quality Assurance Department 2) line movement of the oven is suspended until proper	Check accuracy of temperature recording device (computer monitor from continuous system) a minimum of once per shift QA verifies accuracy of final product temperatures to within +/-2°F on a daily basis Calibrate recording devices annually Daily review of	Final cook zone temperature (CCP temperature) and internal temperature recording chart (Final Cook CCP Record) located by the final cook door zone of the house has the date and time recorded for each entry,

Table 7.1Frankfurters traditional HACCP plan form Continued

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							temperature can be attained (re-cook) 3) product "since last acceptable CCP check" is placed on "hold" 4) identify cause of deviation (power failure, steam loss, etc.) adjust and repair equipment and establish a preventive action plan 5) a recognized Processing	monitoring records	calibrated thermometer ID records, and is signed or initialed. *All documents maintained for at least one year

Table 7.1Frankfurters traditional HACCP plan form Continued

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							Authority will determine product disposition on a case-by-case basis *Corrective action records when applicable		

Source: Frankfurter HACCP plan (unpublished) from undisclosed private company (Anonymous 2015)
Note: This traditional HACCP plan is based on the HACCP plan at the undisclosed visited facility and not on the USDA Generic HACCP model for fully cooked, not shelf stable meat and poultry products (USDA 1999).

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
5RB Initial level of *L. mono-cytogene s* in raw meat pre-blend	*Listeria monocytogenes*	Supplier certificatio n that product meets Listeria performanc e standards and meets other establishm ent specificatio ns must accompany shipment	Receiving personnel will check each shipment for certification and microbiologica l state of the meat pre-blend	Verify packages are intact and lot number in the product shipment matches the Certificat e of Analysis (CoA)	Each shipment	Receivi ng person nel	Will not receive product unaccompani ed by Listeria certification. If company fails to meet Listeria performance standards, it will be an ineligible supplier until standards are again met	Every two months QA requests Listeria data results from meat suppliers Between-lot testing to verify Listeria performance standards meet requirements as intended	Receivin g Log Correctiv e Action Log

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
5RB Raw meat pre-blend refrigerated storage	*Listeria monocytogenes*	Raw product storage areas shall not exceed 40° F in refrigerated rooms or exceed 28° F in freezer rooms	Check raw product storage area temperature	Use a designated thermometer to monitor the temperature of the raw product storage cold room	Every 2 hours	Maintenance personnel	QA will reject or hold product dependent on time and temperature deviation. QA will identify the cause of the deviation and prevent reoccurrence. QA will assure that no product that may be adulterated has entered commerce	Maintenance supervisor will verify accuracy of the Room Temperature Log once per shift. QA will check all thermometers used for monitoring & verification for accuracy daily and calibrate to within 2° F accuracy as necessary	Room Temperature Log Thermometer Calibration Log Corrective Action Log

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
4RB Thermal treatment at processing, including: Temperature of the thermal treatment at processing Time or duration of thermal treatment	*Listeria monocytogenes*	Internal temperature 158°F (70°C) in the center of product for a length of time of 2 minutes (Gaze and others 1989). The time of 2 minutes at 158°F achieves a six logarithm reduction (6D) in the number of *L. monocytogenes* (FDA 2011). 6D reduction of *L. monocytogenes* in ready-to-eat chilled foods (Lund and others 1989) is an example of performance criteria (ICMSF 2002). Although D values are dependent upon	Monitor temperature of finished product in continuous oven with a calibrated thermometer QA will monitor time/temperature parameters to assure that critical limit was met. Continuous temperature recording chart for each smokehouse will be	Randomly select three frankfurters from the hangers in the coolest part of the final cook zone. Write the time at which samples were removed from cooker and immediately measure the temperature in the geometri	Continuous temperature recording chart for each batch. At the end of cooking, the internal temperature of three product samples will be taken and recorded In the continuous oven the monitoring will occur once per hour at the end of cooking zone.	Oven operator and Quality Assurance (QA) personnel	If the internal temperature does not attain specified cooking or chilling temperatures, then, 1) the operator notifies his supervisor and the Quality Assurance Department 2) line movement is suspended until proper temperature can be attained 3) All affected product "since last acceptable CCP check" is placed on "hold" (dependent on	QA supervisor will observe QA technician perform monitoring activities once per shift. QA taking internal temperature and sustained time once per day. Maintenance supervisor will verify accuracy of the oven and cooler temperature recording charts or device (computer monitor	Time/Temperature Log for final cook zone Internal Temperature Recording Chart (Final cook CCP record*) Product Temperature Log Oven Thermometer Calibration Log Corrective Action Log *located by the

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
at processing		the specific food being heated, the above-stated values are generally conservative and apply to all foods (FDA 2011, p. 419) Although this is a sufficient log reduction for this pathogen as indicated in validated time/temperature tables (lethal rate 1.000) (FDA 2011, p. 422), it is important to take into account the variability of the system under normal operating conditions (ICMSF 2002, p. 64). For example, if the thermal process must exceed 158°F 99.9% of	initialed for each batch.	c center of each frankfurter. Average the three temperatures.			time and temperature deviation) 4) A recognized Processing Authority or expert consultant will advise the plant about product deviation and determine product disposition on a case-by-case basis; on the basis of this advice product will be recooked or condemned. 5) QA always follows Process Authority recommendati	from continuous system) a minimum of once per shift. QA verifies accuracy of final product temperatures to within +/-2°F on a daily basis QA will check all thermometers used for monitoring and verification for accuracy daily and calibrate to within 2°F accuracy as necessary.	final cook door zone of the house has the date and time recorded for each entry, calibrated thermometer ID records, and is signed or initialed. **All documents maintained for at least one year

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		the time, the above-mentioned temperature (158°F) and pertinent standard deviation (SD) should be considered. Assuming SD=0.8, then the internal temperature of 160.4°F in the center of product should be reached to assure that the thermal process exceeds 158°F 99.9% of the time to achieve an acceptable level of safety. If process variation can be reduced to 0.5°F, then the CL could be set at 159.5°F to					on and identifies the cause of the deviation (power failure, steam loss, etc.) and establish a preventative action plan to prevent reoccurrence. 6) Cold spots in smokehouse, if detected, will be monitored and product temperature determined on these additional points and from each lot prior to release. →	Daily review of monitoring records → 7) Maintenance will review operation of the cooking or chilling equipment to adjust and repair as necessary. Maintenance schedule will be reviewed as necessary. Corrective action records when applicable.	

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		achieve the same degree of risk.							

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
0RB Brine chilling Chilling is usually part of the thermal treatment (4RB) to cool down the products immediately after cooking. In the case of the processing at the facility visited, the duration of this step was only 14 minutes.	*Listeria monocytogenes*	Immediately after cooking, the thermal treatment also includes a chilling step. Product to cool from 158°F to 30°F in 14 minutes. Chilling begins with a pasteurized potable water shower and continues with submersion of the product in pasteurized acid brine chill (citric acid >0.5M).	QA technician will observe chilling handling procedures to ensure critical limits are met. Cooler temperature will be monitored and recorded continuously using temperature record charts.	QA technician will select and check three samples per batch to ensure chilling time/temperature requirements have been met.	Time/Temperature charts will be reviewed for each product lot with time of observation recorded & initialed every hour.	QA technician	If the internal temperature does not attain specified cooking or chilling temperatures, then, 1) the operator notifies his supervisor and the Quality Assurance Department 2) line movement is suspended until proper temperature can be attained 3) all affected product "since last acceptable CCP check" is placed on "hold" (dependent on	QA will check all thermometers used for monitoring and verification for accuracy daily and calibrate to within 2°F accuracy as necessary. QA supervisor will review the product chilling log and cooler temperature recording chart once per shift.	Cooler Temperature Recording Chart Product Chilling Log Chilling Thermometer Calibration Log All documents maintained for at least one year.

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
In addition, the brine was pasteurized and the contact surface was peeled at the immediate subsequent step. Thus, in this particular case, brine chill did not represent a concern in regards to *L. monocytogenes.* Other facilities, however, may need to consider							time and temperature deviation) 4) a recognized Processing Authority or expert consultant will advise the plant about product deviation and determine product disposition on a case-by-case basis; on the basis of this advice product will be recooked or condemned. 5) QA always follows Process Authority recommendati	Daily review of monitoring records. Between-lot testing to verify Listeria performance standards meet requirements as intended → 7) maintenance will review	

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
this step if the conditions are different.							on and identifies the cause of the deviation (power failure, steam loss, etc.) and establish a preventative action plan to prevent reoccurrence. 6) cold spots in smokehouse, if detected, will be monitored and product temperature determined on these additional points and from each lot prior to release. →	operation of the cooking or chilling equipment to adjust and repair as necessary. Maintenance schedule will be reviewed as necessary. Corrective action records when applicable.	

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
2RB Handling at packaging	*Listeria monocytogenes*	No *Listeria monocytogenes* on product contact surfaces	Verify that sanitizer of demonstrated effectiveness against Lm is used and record results in the mid-shift cleanup log	Following SSOP procedures supervised cleaning crew uses sanitizer on all product contact surfaces	At mid-shift clean up	Cleaning crew supervisor	QA will address positive *Listeria* samples as detailed in the USDA/FSIS issuance "*Listeria* Guidelines for Industry." All products back to last cleanup will be held. No adulterated product will be shipped.	QA will observe cleaning crew supervisor; review log results, and once per week QA will verify that appropriate sanitizer is used according to manufacturer's instructions.	Listeria sampling log Corrective action log Mid-shift cleanup log (Revise Mid-shift procedures)

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
2RB Portioning (personnel is in direct contact with the product removing the defective frankfurters in their portion packs using stainless steel mesh gloves)	*Listeria monocytogenes*	No *Listeria monocytogenes* on product contact surfaces	Verify that sanitizer of demonstrated effectiveness against Lm is used and record results in the mid-shift cleanup log	Following SSOP procedures supervised cleaning crew uses sanitizer on all product contact surfaces	At mid-shift clean up	Cleaning crew supervisor	QA will address positive *Listeria* samples as detailed in the USDA/FSIS issuance "*Listeria* Guidelines for Industry." All products back to last cleanup will be held. No adulterated product will be shipped.	QA will observe cleaning crew supervisor; review log results, and once per week QA will verify that appropriate sanitizer is used according to manufacturer's instructions.	Listeria Sampling Log Corrective Action Log Mid-shift Cleanup Log (Revise Mid-shift procedures)

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
3RB Finished Product Storage (Cold) Finished product was assumed to be shipped immediately after production	*Listeria monocytogenes*	Finished product storage areas will not exceed 40° F.	Monitor finished product storage area temperatures and records results.	Continuous recording thermometer with visual checks	Continuous with visual check of recording every two hours	Cooler manager	If a deviation from a critical limit occurs, the following corrective actions will be taken: 1) The cause of the temperature exceeding 40° F will be identified and eliminated. 2) The CCP will be monitored hourly after the corrective action is taken to ensure that it is under control. 3) When the cause of the deviation is identified, measures will be taken to prevent it	Use appropriate microbiological criteria for *L. monocytogenes* in RTE products where growth can occur. Weekly review of monitoring and corrective action records. Check accuracy of time temperature recorder daily. Calibrate to within 2° F accuracy as necessary.	Room Temperature Log Thermometer Calibration Log Corrective Action Log

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							from recurring, e.g., if the cause is equipment failure, the preventive maintenance program will be reviewed and revised, if necessary. 4) If room temperature exceeds the critical limit, the Processing Authority will evaluate the product time/temperature deviation to ensure the present temperature is sufficient to preclude pathogen growth before release for	Maintenance supervisor will verify the accuracy of the room temperature log once per shift.	

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							shipment. If the temperature is not sufficient to preclude pathogen growth, recooking can be considered after computer modeling of *Listeria monocytogenes* growth in an assumed worst-case scenario.		

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
3RB Retail module Duration of refrigerated storage during retail Temperature of unopened package at retail distribution Duration of transportation from processing facility	*Listeria monocytogenes*	Cold room should be set to maintain a temperature of 40 °F or below. If product is received frozen, it is recommended to keep it that way for as long as possible. Follow producer recommendations and verify expiration date and appropriate label are included in the process. Follow a first in, first out (FIFO) approach with organized cold room storage.	Date Temperature FIFO Monitor duration of transportation from retail to consumer refrigerator	Verify packages are intact and lot number in the product shipment matches the Certificate of Analysis (CoA) Use designated thermometer Check for accurate receiving	Each shipment	Retail receiving personnel	If temperature in the cold room or product, then, - the retail operator notifies his supervisor - all affected product “since last acceptable CCP check” is placed on “hold” (dependent on time and temperature deviation) -identify the cause of the deviation (power failure, steam loss, etc.) and follow preventive action plan to prevent reoccurrence.	Every two months QA will request *Listeria* data results from retail companies Between-lot testing to verify Listeria performance standards meets requirements as intended. Shelf life testing.	Retail receiving logs including the following: -Temperature of transport at receiving at retail level -Date of receiving product Retail storage logs: -Temperature at the back and front of the room for both

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
to retail distribution				dates, expiration dates			-maintenance reviews operation of the →	→ cold room equipment to adjust and repair as necessary.	refrigerated and frozen rooms -Storage handling specifications -First in, first out (FIFO) Logs Corrective action log

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
1RB Consumer refrigerated storage: Duration of refrigerated consumer storage Temperature of unopened package at consumer refrigerator Temperature of opened	*Listeria monocytogenes*	"If there is no product date, hot dogs can be safely stored in the unopened package for 2 weeks in the refrigerator; once opened, only 1 week" (USDA 2013) Refrigerators should be set to maintain a temperature of 40 °F or below. When consumer leaves the grocery store with hot dogs, needs to head straight home and refrigerate or freeze them immediately. (USDA 2013)	Verify consumer refrigerator temperature Verify duration of refrigerated consumer storage for opened and unopened RTE products	Check temperature in build-in or appliance thermometer. Consumer should write date in the package immediately prior to refrigerated storage and again when the original package is opened. Check expiration dates.	Every time frankfurters are retrieved from refrigerator for consumption purposes	Consumers	If refrigerator does not have built-in thermometer to measure their internal temperature, keep an appliance thermometer in the refrigerator to monitor the temperature. This can be critical in the event of a power outage.	Verify the temperature of the refrigerator with a calibrated thermometer Verify refrigerator/ freezer doors are closed tightly at all times. Don't open refrigerator/ freezer doors more often than necessary and close them as soon as possible.	In the event of a power outage, do not open refrigerator. When the power goes back on, if the refrigerator is still 40 °F, the food is safe. Records may be needed depending on the duration of storage under temperatures

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
package at consumer refrigerator									above 40 °F. Foods held at temperatures above 40 °F for more than 2 hours should not be consumed.

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
1RB Duration of transportation from retail to consumer	*Listeria monocytogenes*	When consumer leaves the grocery store with hot dogs, needs to head straight home and refrigerate or freeze them immediately. (USDA 2013)	Monitor duration of transportation from retail to consumer refrigerator	Consumer will use a timer and will place the product in a thermal storage bag. Consumer will store the product in the refrigerator or freezer as soon as possible	Every time that RTE food products are purchased	Consumers	Consumer should never leave hot dogs at room temperature for more than 2 hours and no more than 1 hour when the temperature goes above 90 °F (USDA 2013)	N/A Verification at this level is not possible under normal conditions	N/A Records at this level are not collected under normal conditions

Table 7.2 Frankfurters risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazards**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
1RB Reheating log reduction at consumer level	*Listeria monocytogenes*	At least 4 log reduction (see distribution in Chapter 6 based on FDA/USDA 2003) =RiskCumul (4.49,6.68, {5.3,6.18,6.68}, {0.75,0.95,0.99 }, RiskName ("Reheating Log Reduction")) Reheating the frankfurters will effectively lower the risk for listeriosis in susceptible populations.	Monitor reheating to achieve acceptable *Listeria* log reduction	Reheat frankfurters until steaming hot before eating (USDA 2013)	Every time a frankfurter is consumed, immediately prior to consumption	Consumers, especially those at increased risk of foodborne illness	If product is not reheated until steaming hot, they should not be consumed by susceptible populations or should be reheated properly immediately.	N/A Verification at this level is not possible under normal conditions	N/A Records at this level are not collected under normal conditions

Table 7.3 Frankfurters HACCP plans comparison summary

Critical Control Points (CCPs) and Risk-based Critical Control Points (RB-CCPs))	**Traditional HACCP Plans**		**Risk-based CCPs Scenarios**	
	Facility visited HACCP[a]	USDA Generic HACCP[b]	Baseline (FNR)[c]	Frankfurters reheated (FR)[d]
Initial level of *L. monocytogenes* in meat pre-blend [e]	- [m]	+ [n]	+	+
Temperature of the thermal treatment at processing [f]	+	+	+	+
Time or duration of thermal treatment at processing [g]	-	+	+	+
Chilling or brine chill [j]	-	+	-	-
Reheating log reduction at consumer level [h]	N/A	N/A	N/A	+
Handling at packaging/portioning [i]	-	+	+	+
Duration of refrigerated consumer storage	-	-	+	+
Duration of transportation to retail distribution	-	-	+	+
Duration of transportation from retail to consumer	-	-	+	+
Duration of refrigerated storage during retail	-	-	+	+

Table 7.3 Frankfurters HACCP plans comparison summary (Continued)

Critical Control Points (CCPs) and Risk-based Critical Control Points (RB-CCPs)	Traditional HACCP Plans		Risk-based CCPs Scenarios	
	Facility visited HACCP[a]	USDA Generic HACCP[b]	Baseline (FNR)[c]	Frankfurters reheated (FR)[d]
Temperature of unopened package at consumer refrigerator	-	-	+	-
Temperature of opened package at consumer refrigerator	-	-	-	+
Temperature of unopened package at retail distribution [k]	-	-	+/- [o]	+/-

[a] The only CCP found for the traditional frankfurter HACCP plan form from the undisclosed facility visited (unpublished) was the temperature during "thermal processing."

[b] Traditional CCPs for frankfurters in this column were based on the "USDA Generic HACCP" model for fully cooked, not shelf stable meat and poultry products (USDA 1999).

[c] FNR represents the baseline "frankfurters not reheated" scenario. The model was truncated after the overwhelming thermal treatment at processing. This resulted in RB-CCPs of previous modules not being shown in the outputs of the subsequent modules.

[d] FR represents the "frankfurters reheated" scenario. The model was truncated after the overwhelming thermal treatment at processing. This resulted in RB-CCPs of previous modules not being shown in the outputs of the subsequent modules.

[e] The initial level of *L. monocytogenes (Lm)* in the raw meat pre-blend was not shown in the final tornadoes as RB-CCP because the model was truncated as stated above (c,d). However, the initial level of *Lm* ("cfu/g / IC") in the raw meat pre-blend was the most critical factor in the outputs at the end of the ingredients module level (see Figure 6.1.1).

[f,g] The temperature ("Tck1(°C)") and time ("tck1") of the thermal treatment for the baseline model was one of the most critical factors at the end of the raw product processing module (Figure 6.1.2).

[h] The reheating log reduction at the consumer level is specific to the FR scenario.

[i] Handling at packaging also involved the portioning of frankfurters. Portioning is considered a CCP in the USDA Generic HACCP plan for fully cooked, not shelf stable model.

[j] The USDA generic HACCP plan also included chilling as a CCP. However, the duration of this step in the visited facility was only 14 minutes on average which is a rapid treatment compared to the 4-6 hours required by the examples in the USDA Generic HACCP plan. In addition, the model assumed that the brine chill was pasteurized and not a likely source of *L. monocytogenes.*

[k] "Temperature of unopened package at retail distribution" is only shown at the bottom of Figure 6.1.7A (FR). However, it was also recorded (but not shown) at the bottom of Figure 6.1.5A (FNR).

[l] The baseline model assumed that retail distribution occurred immediately after finishing production. Thus the "Temperature (retail) distribution," "Time (retail) distribution," and "Finished product cooler storage" refer to the temperature of storage (at the facility or otherwise) after the product is finalized. The traditional CCP was referred to as "finished product cooler storage" and the equivalent RB-CCPs as "Temperature (retail) distribution" and "time (retail) distribution." The baseline model assumed that the final product was refrigerated.

[m] The sign - means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes did not show this step, condition, or parameter as RB-CCP.

[n] The sign + means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes showed this step, condition, or parameter as RB-CCP.

[o] The sign +/- means that the sensitivity analysis for the inputs ranked by effect on output mean tornado showed this step, condition, or parameter as RB-CCP. However, the contribution to variance tornado did not show this step, condition, or parameter as RB-CCP.

Table 7.4 Cold-smoked salmon traditional HACCP plan form

(1) Critical Control Point (CCP)	(2) Significant Hazards	(3) Critical Limits	(4) Monitoring	(5) Monitoring	(6) Monitoring	(7) Monitoring	(8) Corrective Action(s)	(9) Verification	(10) Records
			What	**How**	**Frequency**	**Who**			
Brine	Pathogenic bacteria growth temperature abuse	Fish are brined in refrigeration at a temperature of 40°F or less Minimum brining time of 24 hours Minimum of 60° salometer reading at start of the process Minimum water phase salt concentration of 3.5% in the final finished product	Cooler temperature Start and end time in brine Salinity of brine solution (degrees salt) Other conditions to be considered for each particular batch are Weight and size of fish, volume of brine	Continuous recording thermometer Visual check of time Salometer	Continuous with visual check once per batch Each batch Each batch at the start of the brine process	Brine operator Brine operator Brine operator	Move brine tanks to another cooler and fix cooler or adjust thermostat * If brining time is not met, then hold in brine until 24 hours is reached Add more salt and mix until salometer reads 60° Determine safety of product based on time and temperature exposure	Check accuracy of recording thermometer before initial use and then daily, and then annual calibration. Brine validation study for brine time. Quarterly lab analysis to verify that finished products have 3.5% water phase salt. Daily accuracy check of scale. Annual calibration of scale. Weekly review of brine logs, and corrective	Temperature recording chart with visual checks Brine logs including Time recording Brine validation study that demonstrates a minimum water phase salt concentration of 3.5% in the final finished product

Table 7.4 Cold-smoked salmon traditional HACCP plan form Continued

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
								action records.	
Cold smoking	Pathogenic bacteria growth temperature abuse	Internal fish temperature two options: 1) not to exceed 90°F (32.2°C) during a period of 20 hours or less or 2) not exceed 50°F (10°C) for a period of 24 hours or less (Otwell and others 2004)	Internal fish temperature at the thickest portion of three fish. Time fish are at target internal temperature.	Continuous temperature recording device with three temperature probes	Continuous with visual check of recording chart for each batch	Smoker operator	If proper time and temperature is not reached, then re-smoke: 1) not to exceed 90°F (32.2°C) during a period of 20 hours or less or 2) not exceed 50°F (10°C) for a period of 24 hours or less or destroy batch and adjust and	Weekly review of monitoring and corrective action records Check accuracy of temperature recording device before initial use and before each batch Calibrate recording device annually	Time and temperature recording chart and smoker log

Table 7.4 Cold-smoked salmon traditional HACCP plan form Continued

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							repair equipment		
Vacuum pack/ weight/label	Food Allergens	All product labels will contain "Salmon" in the ingredient list	Labels on finished product	Visual check of labels	A representative number of packages from each lot	Packing operator	If the label does not have "Salmon" in the ingredients list, then re-label any improperly labeled product and modify labeling procedures as appropriate	Weekly review of monitoring and corrective action records	Label check report
Finished product refrigerated storage	Pathogenic bacteria growth temperature abuse	Cooler temperature is 40°F or less (FDA 2011, Table A-2)	Cooler temperature	Continuous recording thermometer with visual checks	Continuous with visual check of recording once a day	Cooler manager	If cooler temperature is above 40F, then move product to another cooler or ice and hold for	Weekly review of monitoring and corrective action records. Calibrate temperature recorder	Cooler temperature log and time temperature recording chart.

Table 7.4 Cold-smoked salmon traditional HACCP plan form Continued

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Critical Control Point (CCP)	**Significant Hazards**	**Critical Limits**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							evaluation. Adjust or repair cooler as necessary. Evaluate product safety determining cumulative exposure temperature and time above 40°F. Destroy if necessary.	once per year. Check accuracy of time temperature recorder daily.	

Source: Adapted from National Seafood HACCP Alliance for Training and Education (Anonymous 2016)

Note: This traditional HACCP plan for cold-smoked salmon (Anonymous 2016) was not based on the HACCP plan at the visited CSS facility or on the Anonymous (1996) HACCP plan shared by Dr. Blakistone. Instead, this traditional HACCP plan for cold-smoked salmon was developed as part of the requirements for completion of the Seafood HACCP Alliance "Segment Two" Course using the Fish and Fishery Products Hazards and Controls Guidance (FDA 2011) and the Hazard Analysis and Critical Control Point Training Curriculum (National Seafood HACCP Alliance 2011). It is noteworthy that the "Segment Two HACCP Plan" was originally developed for hot-smoked salmon during the Seafood HACCP Alliance/AFDO Seafood Segment 2 course together with a group of seafood industry professionals (September 2016). It was then compared with a HACCP model for hot-smoked salmon (National Seafood HACCP Alliance, December 2016) and adapted for cold-smoked salmon.

Table 7.5 Cold-smoked salmon risk-based HACCP plan form for L. monocytogenes

(1) Risk-based Critical Control Point (RB-CCP)	(2) Specific Hazard(s)	(3) Risk-based Critical Limits (RB-CL)	(4)	(5)	(6)	(7)	(8) Corrective Action(s)	(9) Verification	(10) Records
			Monitoring						
			What	**How**	**Frequency**	**Who**			
5RB Raw Salmon Lm Incidence/ Fresh Raw Product Storage Initial Lm reference distribution	*Listeria monocytogenes*	Supplier certification that product meets Listeria performance standards and other establishment specifications must accompany shipment Raw product storage areas in the shipment truck shall not exceed 40°	Receiving personnel will check each shipment for certification and microbiological state of the meat pre-blend Receiving personnel will measure the temperature in the shipment	Receiving personnel will verify that lot number in the product shipment matches the certificate of analysis (CoA) Use appropriate thermometer to measure	Each shipment →Use a designated thermometer to monitor the temperature of the raw	Receiving personnel	If incoming product is not accompanied by *Listeria* performance standards certification in CoA and the temperature of required frozen or refrigerated product is not met, establishment will not receive the product. If supplier fails to meet	Every two months quality assurance (QA) personnel will request *Listeria* data results from the suppliers. Supplier verification program in place Process control verification testing or investigation testing, if necessary	Receiving log Thermometer calibration log Cold room temperature log Corrective action log

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		F in refrigerated rooms or exceed 28° F in freezer rooms	truck and incoming product Check raw product storage area temperature	temperature of the truck and temperature of the product and write the lot of the product and the truck license plate	product storage cold room		*Listeria* performance standards, it will be an ineligible supplier until standards are again met and they satisfy the internal requirements.		
6RB Brining	*Listeria monocytogenes*	Fish are brined in refrigeration at a temperature of 40°F or less Minimum brining time of 24 hours	Cooler temperature Start and end time in brine	Continuous recording thermometer Visual check of time	Continuous with visual check once per batch Each batch	Brine operator Brine operator	Move brine tanks to another cooler and fix cooler or adjust thermostat * If brining time is not	Check accuracy of recording thermometer before initial use and then daily, and then annual	Temperature recording chart with visual checks Brine logs including

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		Minimum of 60° salometer reading at start of the process Minimum water phase salt concentration of 3.5% in the final finished product	Salinity of brine solution (degrees salt) Other conditions to be considered for each particular batch are weight and size of fish, volume of brine	Salometer	Each batch at the start of the brine process	Brine operator	met, then hold in brine until 24 hours is reached. Add more salt and mix until salometer reads 60° *Determine safety of product based on time and temperature exposure	calibration. Brine validation study for brine time. Quarterly lab analysis to verify that finished products have 3.5% water phase salt Daily accuracy check of scale. Annual calibration of scale. Weekly review of brine	time recording Brine validation study that demonstrates a minimum water phase salt concentration of 3.5% In the final finished product

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
								logs, and corrective action records.	
4RB Cold Smoking	*Listeria monocytogenes*	Internal fish temperature two options: 1) not to exceed 90°F (32.2°C) during a period of 20 hours or less or 2) not exceed 50°F (10°C) for a period of 24 hours or less (Otwell and others 2004)	Internal fish temperature at the thickest portion of three fish. Time fish are at target internal temperature	Continuous temperature recording device with three temperature probes	Continuous with visual check of recording chart for each batch	Smoker operator	If proper time and temperature is not reached, then re-smoke: 1) not to exceed 90°F (32.2°C) during a period of 20 hours or less or 2) not exceed 50°F (10°C) for a period of 24 hours or less or	Use appropriate calculated microbiological criteria (MC4) for *L. monocytogenes* in RTE products where growth can occur following the calculated sampling plan for the cold smoking step based	Time and temperature recording chart and smoker log → Approximately 1 log bacterial count reduction is to be expected (ICMSF 2011, p. 125). Weekly review of

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							destroy batch and adjust and repair equipment	on the FSO/PO as follows: n=15 c=0 m=absence in 25g Cold smoking step is expected to achieve at least 1 log reduction of *L. monocytogenes.* This can be verified testing swabs of the fish before and after	monitoring and corrective action records. Check accuracy of temperature and time recording device before initial use and before each batch. Calibrate recording device as necessary.

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
								this processing step. →	
2RB Food Contact Surfaces: Slicing	*Listeria monocytogenes*	1.5 log reduction (Crandall and others 2012)	Cleaning and sanitizing monitoring Food contact surface testing for *Listeria*	Address as detailed in company SSOPs and procedures for testing.	Depending on each line, design of equipment and manufacturing specifications. Mid-shift cleaning.	QA personnel	If *L. monocytogenes* is detected on a food contact surface (FCS), the lot of RTE food needs to be either reprocessed with a validated listericidal control measure, diverted to a use in which the food will not be consumed	QA will observe cleaning crew supervisor; review log results, and once per week QA will verify that appropriate sanitizer is used according to manufacturer's instructions.	Listeria sampling log Corrective action log Mid-shift cleanup log (Revise Mid-shift procedures)

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							by humans or animals, sent for use in animals (where appropriate), or destroyed. In addition, consideration of potential contaminated product in commerce that should be recalled is needed (FDA 2017).		
2RB Portioning	*Listeria monocytogenes*	No *Listeria monocytogenes* on product	Verify that sanitizer of demonstrated effectiveness against *Lm* is	Following SSOP procedures supervised cleaning	at mid-shift clean up	Cleaning crew supervisor	QA will address positive Listeria samples as	QA will observe cleaning crew supervisor	Listeria Sampling Log

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		contact surfaces	used and record results in the mid-shift cleanup log Food contact surface testing for *Listeria*	crew use sanitizer on all product contact surfaces			detailed in the company procedures. Mid-shift procedures will be revised. All products back to last cleanup will be held. No adulterated product will be shipped.	; review log results, and once per week QA will verify that appropriate sanitizer is used according to manufacturer's instructions.	Corrective Action Log Mid-shift Cleanup Log
2RB Pinbone	*Listeria monocytogenes*	No *L. monocytogenes* on product contact surfaces	Verify that sanitizer of demonstrated effectiveness against *Lm* is used and record results in the mid-	Following SSOP procedures supervised cleaning crew use sanitizer on all product	at mid-shift clean up	Cleaning crew supervisor	QA will address positive Listeria samples as detailed in the company procedures. Mid-shift	QA will observe cleaning crew supervisor; review log results, and once	Listeria Sampling Log Corrective Action Log

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
			shift cleanup log Food contact surface testing for *Listeria*	contact surfaces			procedures will be revised. All products back to last cleanup will be held. No adulterated product will be shipped.	per week QA will verify that appropriate sanitizer is used according to manufacturer's instructions.	Mid-shift Cleanup Log
1RB Labeling (Related to *L. monocytogenes* and Consumer Module)	*Listeria monocytogenes*	Product labels will contain the following statement if susceptible populations are likely to consume product: "Freeze or keep refrigerated for a maximum of one week	Labels on finished product	Visual check of labels	Visually check a representative number of packages from each lot	Packing operator	If the label does not have "Freeze or keep refrigerated for a maximum of one week after purchase," then →	Verify label statement to be printed in the packaging materials or labels → Re-label any improperly labeled product modify labeling	Labeling logs

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		after purchase or thawing."						procedures as appropriate.	
3RB Finished Product Refrigerated Storage (This is part of retail as it was assumed that the product was shipped immediately after production)	*Listeria monocytogenes*	Cooler temperature is 40°F or less (FDA 2011)	Cooler temperature Duration of storage. Apply first in, first out approach. "Hold and Test" final product as a monitoring activity (Buchanan and Schaffner 2015)	Continuous recording thermometer with visual checks initialed by cold room personnel	Continuous with visual check of recording every two hours	Cooler manager	If cooler temperature is above 40F, then move product to another cooler or ice and hold for evaluation. Adjust or repair cooler as necessary. Evaluate product safety determining cumulative exposure temperature and time	Use appropriate microbiological criteria for *L. monocytogenes* in RTE products where growth can occur. Weekly review of monitoring and corrective action records. Check accuracy of time	Cooler temperature log and time temperature recording chart.

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
							above 40°F. Destroy if necessary.	temperature recorder daily. Calibrate as necessary.	
3RB Retail Distribution Module: Temperature (Retail) Distribution Time (Retail) Distribution	*Listeria monocytogenes*	Cold room should be set to maintain a temperature of 40 °F or below. If product is received frozen, it is recommended to keep it that way for as long as possible. Follow producer recommendations and verify expiration date and	Temperature of storage Duration of storage Location of the storage	Continuous recording thermometer with visual checks initialed by retail personnel Monitor the date and time the product is received for retail distribution. Apply first in, first out	Each shipment	Retail receiving personnel	If temperature in the cold room or product, then, - the retail operator notifies his supervisor - all affected product "since last acceptable CCP check" is placed on "hold" (dependent on time	Verify that the temperatures in the back and front of the cold room are not considerably different. Between-lot testing to verify Listeria performance standards met	Retail receiving logs including the following: - Temperature of transport at receiving at retail level -Date of receiving product Retail storage logs:

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
		appropriate label are included in the process. Follow a first in, first out (FIFO) approach with organized cold room storage.		(FIFO) approach. If possible, store cold smoked salmon in the back of the cold room which is typically colder.			and temperature deviation) -identify the cause of the deviation (power failure, steam loss, etc.) and follow preventive action plan to prevent reoccurrence. - maintenance reviews operation of the →	requirements as intended. Every two months QA will request *Listeria* data results from retail companies. → cold room equipment to adjust and repair as necessary.	- Temperature at the back and front of the room for both refrigerated and frozen rooms -Storage handling specifications -First in, first out (FIFO) Logs Corrective action log

Table 7.5 Cold smoked salmon risk-based HACCP plan form for Listeria monocytogenes *(Continued)*

(1)	(2)	(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
Risk-based Critical Control Point (RB-CCP)	**Specific Hazard(s)**	**Risk-based Critical Limits (RB-CL)**	**Monitoring**				**Corrective Action(s)**	**Verification**	**Records**
			What	**How**	**Frequency**	**Who**			
1RB Consumer Module: Temperature Consumer Time Consumer	*Listeria monocytogenes*	Refrigerators should be set to maintain a temperature of 40 °F or below. Freezing the product at this level will effectively lower the risk for listeriosis. Storing the product for a maximum of 1 week after purchase will effectively lower the risk for human invasive listeriosis in susceptible populations	Monitor the temperature and duration of the refrigerated storage for RTE food products	Using an appliance thermometer in the refrigerator to monitor the temperature. Writing the date the refrigerated storage began either after thawing or after purchase	Every time that cold smoked salmon is stored under refrigeration	Consumer	This can be critical in the event of a power outage. When the power goes back on, if the refrigerator is still 40 °F, the food is safe. Foods held at temperatures above 40 °F for more than 2 hours should not be consumed.	Verify the temperature of the refrigerator with an appliance thermometer Verify refrigerator/ freezer doors are consistently closed tightly Verify the date written in the package is legible	N/A Records at this level are not collected under normal conditions

Table 7.6 Cold-smoked salmon HACCP plans comparison summary

Risk-based Critical Control Points (RB-CCPs) [a]	**Traditional CCPs**		**Risk-based CCPs**				
	"CSS Traditional HACCP plan" (Anonymous 1996)[b]	"Segment Two HACCP plan" (Anonymous 2016)[c]	Baseline Scenario	Scenario 20	Scenario 21	Scenario 22	Scenario 23
Temperature Consumer	- [d]	-	+ [e]	+	+	+/- [f]	+
Time Consumer	-	-	+	+	+	+/-	+
Slicing	-	-	+	+	+	+	+
Temperature (**Retail**) Distribution [g]	+	+	+	+	+	+	+
Time (**Retail**) Distribution [h]	+	+	+	+	+	+	+
Cold Smoking	+	+	+	+	+	+	+
Raw Salmon Lm Incidence [i]	+	+	+	+	+	+	+
Initial Lm reference distribution	-	-	+/-	+	+	+	+/-
Portioning	-	-	+/-	+/-	+/-	+/-	+/-
Pinbone	-	-	-	+/-	+	+	+/-
Thawing [j]	+	-	-	-	-	-	-
Brining [k]	+	+	+	+	+	+	+

[a]

Table 7.6 Cold-smoked salmon HACCP plans comparison summary (Continued)

Risk-based Critical Control Points (RB-CCPs) [a]	**Traditional CCPs**		**Risk-based CCPs**				
	"CSS Traditional HACCP plan" (Anonymous 1996)[b]	"Segment Two HACCP plan" (Anonymous 2016)[c]	Baseline Scenario	Scenario 20	Scenario 21	Scenario 22	Scenario 23
Finished product labeling [l]	+	+	-	-	-	-	-

a All the results shown correspond to the tornadoes at the end of the consumer module level unless specified otherwise.
b The "CSS Traditional HACCP Plan" for cold-smoked salmon was titled "Smoked Fish Processing HACCP Plan Form." This 1996 document (unpublished) was obtained from a personal communication (email message, 15 March 2013) with Dr. Barbara Blakistone, formerly with the National Fisheries Industries (NFI).
c The "Segment Two HACCP Plan" (Anonymous 2016) was originally developed for hot-smoked salmon as part of the requirements for completion of the Seafood HACCP Alliance/AFDO Seafood Segment Two course together with a group of seafood industry professionals (September 2016), compared with a HACCP model for hot-smoked salmon (National Seafood HACCP Alliance for Training and Education, December 2016) and adapted for the cold-smoked salmon process.
d The sign "-" means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes did not show this step, condition, or parameter as RB-CCP.
e The sign "+" means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes showed this step, condition, or parameter as RB-CCP.
f The sign "+/-" means that the sensitivity analysis for the inputs ranked by effect on output mean tornado showed this step, condition, module or parameter as RB-CCP. However, the contribution to variance tornado did not show this step, condition, module, or parameter as RB-CCP. Thus, this symbol refers to variations in the results of the inputs ranked by effect on output mean and contribution to variance tornadoes.
g The baseline model assumed that retail distribution occurred immediately after finishing production. Thus the "Temperature (retail) distribution" and "Finished product cooler storage" refer to the temperature of storage (at the facility or otherwise) after the cold-smoked salmon product is finalized. The traditional CCP was referred to as "finished product cooler storage" and the equivalent RB-CCPs as "Temperature (retail) distribution." The baseline model assumed that the final product was frozen.
h The baseline model assumed that retail distribution occurred immediately after finishing production. Thus the "Time (retail) distribution" and "Finished product cooler storage" refer to the duration of storage (at the facility or otherwise) after the cold-smoked salmon product is finalized. The traditional CCP was referred to as "finished product cooler storage" and the equivalent RB-CCPs as "time (retail) distribution." The baseline model assumed that the final product was frozen.
i "raw salmon Lm incidence" and "fresh raw product storage" refer to the incidence after raw product storage and immediately before production. The traditional CCP was referred to as "fresh raw product storage" whereas the equivalent RB-CCP as "raw salmon Lm incidence." The baseline model assumed that the raw salmon was frozen at receiving.
j Although "Filleting" was shown at the bottom of the CSS baseline model tornado at the consumer level (Figure 6.2.7A), it was not found for scenarios 20, 21, 22, or 23.
k Brining shows as RB-CCP at the end of the Brining module for both, "inputs ranked by effect of output mean" and "contribution to variance" tornadoes. However, at the end of the "cold smoking processing," "post-cold smoking processing," and "distribution and marketing" modules, it only shows as a RB-CCP in the "inputs ranked by effect on output mean" tornado. The results shown for the selected scenarios including the baseline scenario represent the output of the brining module.
l The traditional CCP "finished product labeling" refers to fish as an allergen and is not related to *L. monocytogenes* as the hazard of interest.

Table 7.7 Risk-based HACCP plans comparison summary

Summary of risk-based critical control point (RB-CCP) common categories for selected ready-to-eat (RTE) products and prioritized RB-CCPs (#RB)	**Frankfurters model**		**Cold-smoked salmon model**				
	Baseline scenario (FNR)	**Franks reheated (FR)**	**Baseline scenario**	**Scenario 20**	**Scenario 21**	**Scenario 22**	**Scenario 23**
Raw product initial contamination (5RB)							
Initial level of *L. monocytogenes* (*Lm)* in meat	+ [a]	+	N/A	N/A	N/A	N/A	N/A
Raw salmon *L. monocytogenes* (*Lm)* incidence	N/A	N/A	+	+	+	+	+
Thermal treatment/smoking at processing [e] (4RB)							
Temperature of thermal treatment at processing	+	+	N/A	N/A	N/A	N/A	N/A
Time/duration of thermal treatment at processing	+	+	N/A	N/A	N/A	N/A	N/A
Cold smoking	N/A	N/A	+	+	+	+	+
Food contact surfaces [f] (2RB)							
Handling at packaging/Portioning	+	+	+/- [b]	+/-	+/-	+/-	+/-
Slicing	N/A	N/A	+	+	+	+	+
Pinbone	N/A	N/A	- [c]	+/-	+	+	+/-

Table 7.7 Risk-based HACCP plans comparison summary (Continued)

Summary of risk-based critical control point (RB-CCP) common categories for selected ready-to-eat (RTE) products and prioritized RB-CCPs (#RB)	Frankfurters model		Cold-smoked salmon model				
	Baseline scenario (FNR)	Franks reheated (FR)	Baseline scenario	Scenario 20	Scenario 21	Scenario 22	Scenario 23
Retail distribution module (3RB)							
Time/duration retail distribution refrigerated storage	+	+	+	+	+	+	+
Temperature retail distribution refrigerated storage	+/-	+/-	+	+	+	+	+
Consumer module (1RB)							
Time/duration consumer-refrigerated storage	+	+	+	+	+	+/-	+
Temperature consumer-refrigerated storage	+/-	-/+ [d]	+	+	+	+/-	+
Reheating log reduction at consumer level	N/A	+	N/A	N/A	N/A	N/A	N/A
Other RB-CCPs [g] (6RB)							
Initial *Lm* reference distribution for raw salmon	N/A	N/A	+/-	+	+	+	+/-
Brining (Brine chill for frankfurters: 0RB)	-	-	+	+	+	+	+/-
Duration of transp. from processing to retail	+	+	N/A	N/A	N/A	N/A	N/A
Duration of transp. from retail distrib. to consumer	+	+	N/A	N/A	N/A	N/A	N/A

[a] The sign "+" means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes showed this step, condition, or parameter as RB-CCP.
[b] The sign "+/-" means that the sensitivity analysis for the inputs ranked by effect on output mean tornado showed this step, condition, or parameter as RB-CCP. However, the contribution to variance tornado did not show this step, condition, or parameter as RB-CCP.
[c] The sign "-" means that the sensitivity analysis for both the inputs ranked by effect on output mean and the contribution to variance tornadoes did not show this step, condition, or parameter as RB-CCP.
[d] The sign "-/+" means that that the sensitivity analysis for the contribution to variance tornado showed this step, condition, or parameter as RB-CCP. However, the inputs ranked by effect on output mean tornado did not show this step, condition, or parameter as RB-CCP.

[e] Thermal treatment is assumed to include chilling with cold water in the case of frankfurters and cooling or equilibrating in the case of cold-smoked salmon.
[f] Other food contact surfaces (FCS) of interest could include: peeling and collate transportation.
[g] Although “Filleting” was shown at the bottom of the CSS baseline model tornado at the consumer level (Figure 6.2.7A), it was not found for scenarios 20, 21, 22, or 23.
[h] Retail includes the final storage as it was assumed that the product is immediately shipped to retail distribution after leaving the processing facilities.

Table 7.8 Food safety objective point estimate approach comparison between the risk assessment derived baseline model for frankfurters not reheated and the what-if scenario frankfurters reheated

Scenarios	**FSO $\geq$ $H_0 - \Sigma R + \Sigma I$**		**Process risk metrics for *L. monocytogenes***		
	$H_0 - \Sigma R + \Sigma I$	**(Mean Log CFU per gram)**	**H_0**	**ΣR**	**ΣI**
Frankfurters not reheated (FNR)	1.764		-3.493	0	5.257
Frankfurters reheated (FR)	-3.223		-3.493	-3.069	3.339

Where:
FSO $\geq$ H0 – ΣR + ΣI: ICMSF equation
FSO: Food Safety Objective at the consumer level (PO1)
H0: Initial level of the hazard after the thermal treatment step at the processing facility (model truncated).
ΣR: Total (cumulative) log reduction of the hazard after the thermal treatment step at the processing facility (model truncated).
ΣI: Total (cumulative) log increase of the hazard due to growth and/or recontamination after the thermal treatment step at the processing facility (model truncated).
FSO, H0, ΣR, and ΣI are expressed in log10 units.

Table 7.9 Cold-smoked salmon point estimate approach comparison of stringent scenarios

Scenarios	**FSO ≥ H_0 – ΣR + ΣI**		**Process risk metrics for *L. monocytogenes***		
	H_0 – ΣR + ΣI	**(Mean Log CFU per gram)**	**H_0**	**ΣR**	**ΣI**
Scenario 1 (Baseline)	1.961		0.044	-2.580	4.498
Scenario 20	0.229		0.044	-2.580	2.766
Scenario 21	-1.270		0.044	-2.580	1.267
Scenario 22	-1.607		0.044	-2.580	0.930
Scenario 23	-1.716		0.044	-2.652	0.892

Where:
FSO ≥ H_0 – ΣR + ΣI: ICMSF equation
FSO: Food Safety Objective at the consumer level (PO1)
H_0: Initial level of the hazard
ΣR: Total (cumulative) log reduction of the hazard
ΣI: Total (cumulative) log increase of the hazard due to growth and/or recontamination
FSO, H_0, ΣR, and ΣI are expressed in $\log_{10}$ units.

Table 7.10 Codex Alimentarius Commission microbiological criterion for L. monocytogenes *in ready-to-eat foods*

Product	Microorganism	Point of application	Analytical method	Sampling plan and limits			Class Plan	Case
				n	c	m		
Ready-to-eat foods that support growth	*Listeria monocytogenes*	From the end of manufacture or port of entry (for imported products) to the point of sale	ISO 11290-1	5[a]	0	Absence in 25 g (<0.04 cfu/g)[b]	2[c]	NA[d]
Ready-to-eat foods that do not support growth	*Listeria monocytogenes*	From the end of manufacture or port of entry (for imported products) to the point of sale	ISO 11290-2	5	0	10^2	2	NA

Source: CAC 2009a

Where n = number of samples that must conform to the criterion; c = the maximum allowable number of defective sample units in a 2-class plan; m = a microbiological limit which, in a 2-class plan, separates acceptable lots from unacceptable lots.

[a] National governments should provide or support the provision of guidance on how samples should be collected and handled, and the degree to which compositing of samples can be employed.

[b] Absence in a 25-g analytical unit. This criterion is based on the use of the ISO 11290-1 method. Other methods that provide equivalent sensitivity, reproducibility, and reliability can be employed if they have been appropriately validated (e.g., based on ISO 16140).

[c] Assuming a lognormal distribution, this sampling plan would provide 95% confidence that a lot of food containing a geometric mean concentration of 0.023 cfu/g and an analytical standard deviation of 0.25 log cfu/g would be detected and rejected based on any of the five samples are positive for *L. monocytogenes.* Such a lot may consist of 55% of the 25g samples being negative and up to 45% of the 25g samples being positive. 0.5% of all the samples from this lot could harbor concentrations above 0.1 cfu/g.

[d] NA=not applicable as Codex criterion used in place of ICMSF cases.

The typical actions to be taken where there is a failure to meet the above criterion would be to (1) prevent the affected lot from being released for human consumption, (2) recall the product if it has been released for human consumption, and/or (3) determine and correct the root cause of the failure.

Table 7.11 International Commission on Microbiological Specifications for Foods suggested sampling plans stringency in relation to the degree of risk and conditions of use (selected cases 13-15)

Degree of concern relative to level of health hazard (*Listeria monocytogenes)*	**Conditions in which RTE food is expected to be handled prior to consumption and consumed after sampling in the usual course of events***		
	Conditions **reduce** degree of concern (e.g., reheating RTE food product)	Conditions cause **no change** in concern (e.g., Lm growth will not occur)	Conditions **may increase** concern (e.g., Lm growth can occur)
Severe hazard: for general or restricted populations, causing life-threatening or substantial chronic sequelae or illness of long duration: m = 0/25 g (Human invasive listeriosis)	Case 13 n=15, c=0 Two-class Mean level 1 CFU/330 g	Case 14 n=30, c=0 Two-class Mean level 1 CFU/850 g	Case 15 n=60, c=0 Two-class Mean level 1 CFU/2,000 g

Adapted from ICMSF (2002), ICMSF (2011), and FDA (2017)
*More stringent sampling plans would generally be used for sensitive foods destined for susceptible populations (e.g., baby food, food for hospitals, foods for AIDS patients, dietetic food, and relief foods)

Where
n = number of samples to be tested that must conform to the criterion;
c = the maximum allowable number of defective sample units (positives) in a 2-class plan;
m = a microbiological limit which, in a 2-class plan, separates acceptable lots from unacceptable lots
Lm = *Listeria monocytogenes*

Table 7.12 Useful microbial testing for cooked meat products and lightly preserved fish as recommended by the International Commission on Microbiological Specifications for Foods including sampling plans comparison for end-product testing for L. monocytogenes

Testing	**Product**	**Relative importance**	**Useful testing (relative to *Listeria monocytogenes*)**
Critical Ingredients	Cooked meat	Low	These products do not contain nonmeat ingredients of significance for microbiological safety or quality
	Lightly preserved fish	Low	If brine injection is used, the brine should be prepared freshly for each batch or checked for presence of *L. monocytogenes*, which should be absent
In-process	Cooked meat	High Medium	Monitoring the cooking parameters is essential For products that support *L. monocytogenes* growth, postcook samples can assess control of *Listeria* spp. Typical levels encountered postcook: • *Listeria* spp. – absent
	Lightly preserved fish	Low	In-process Low In-process samples are not routinely collected
Processing environment	Cooked meat	High Medium	For products that support *L. monocytogenes* growth, during production sample product contact surfaces where cooked products are exposed to potential contamination before packaging. Sponge or swab samples from floors, drains and other nonproduct contact surfaces can provide an early indication of the level of control and a potential risk of contamination for equipment and product. Typical levels encountered: • *Listeria* spp. – absent Sample equipment surfaces before start-up to verify efficacy of cleaning and disinfecting.
	Lightly preserved fish	High	Swab product contact surfaces and close surfaces, and test for aerobic colony count and *L. monocytogenes.* Typical levels encountered after cleaning and disinfection: • Aerobic colony counts – <10–10^2 CFU/cm2 • *L. monocytogenes* – absent
Shelf life	Cooked meat	Medium	Shelf-life testing may be useful for refrigerated products with extended code dates. Shelf-life testing of frozen cooked meats is not necessary.

Table 7.12 Useful microbial testing for cooked meat products and lightly preserved fish as recommended by the International Commission on Microbiological Specifications for Foods including sampling plans comparison for end-product testing for L. monocytogenes *(Continued)*

Testing	**Product**	**Relative importance**	**Useful testing (relative to *Listeria monocytogenes*)**
	Lightly preserved fish	Medium	Shelf-life Medium Shelf-life testing through sensory assessment may be useful for products with longer shelf life. The potential for growth of *L. monocytogenes* during shelf life should be determined
End product	Cooked meat	Medium	End-product Medium Test for indicators for ongoing process control and trend analysis
		Medium	Routine sampling for pathogens is not recommended. If application of GHP or HACCP is in question, the following sampling plans are recommended
	Lightly preserved fish	Medium	End-product Medium Routine sampling for pathogens is not necessary. If application of GHP and HACCP is in question, sampling for *L. monocytogenes* may be considered in lot acceptance

					Sampling plan & limits/25 g			
	Product	Microorganism	Analytical Method	Case	*n*	*c*	*m*	*M*
	Cooked meat: supports growth	*L. monocytogenes*	ISO 11290-1	NA	5	0	0	--
	Lightly preserved fish: supports growth	*L. monocytogenes*	ISO 11290-1	NA	5	0	0	--

Source: Adapted from ICMSF (2011)
Where
n = number of samples to be tested that must conform to the criterion;
c = the maximum allowable number of defective sample units (positives) in a 2-class plan;
m = a microbiological limit which, in a 2-class plan, separates acceptable lots from unacceptable lots
Note: More stringent sampling plans would generally be used for sensitive foods destined for susceptible populations (e.g., baby food, food for hospitals, foods for AIDS patients, dietetic food, and relief foods)

Table 7.13 Potential microbiological criteria for selected performance objectives to verify with a specified degree of confidence that the selected POs are not exceeded at the end of manufacture or retail distribution/point of sale

RTE food products (i.e., CSS or frankfurters)[a]	PO [Log (CFU/g)][b]	Mean level of *L. monocytogenes* [Log (CFU/g)][c]	SD [Log (CFU/g)][d]	Sample size (g)[e]	Sensitivity of the method (m) [Log (CFU/g)][f]	Number of samples (n) required to achieve specific probability of rejecting (P_{rej}) the lot [g]	Amount of food that contains one pathogen (g)[h]
						Desired P_{rej} 0.95	
Cold Smoked Salmon	PO3 (POS)	-1.644	0.8	25	-1.4	5	44.1
	PO3 (Retail)	-1.869	0.8	25	-1.4	6	74.0
	PO2 (End of manufacture)	-2.094	0.8	25	-1.4	9	124.2
Frankfurters	PO3 (Retail)	-1.827	0.5	25	-1.4	7	67.1
			0.2	25	-1.4	8	
	PO3 (POS)	-0.154	0.5	25	-1.4	1	1.4
			0.5	5	-0.7	2	
	PO2 (End of manufacture)	-3.495	0.7	25	-1.4	111	3126.1

a RTE refers to the ready-to-eat food products of case study one (i.e., frankfurters) or case study two (i.e., cold-smoked salmon or CSS)

b PO refers to performance objective (PO) in Log (CFU/g). PO3 refers to the retail distribution module and PO2 refers to the end of manufacture or end of the processing line for all cases. However, PO3 (POS) refers to the point of sale (POS) calculated at the output level (end of) the retail distribution module whereas PO3 (Retail) was calculated half way or in the middle of the retail distribution module (assuming linearity) and not at the output level of this module.

c The mean level of *L. monocytogenes* was obtained from the risk assessment modules. For example, the mean level of *L. monocytogenes* for CSS at PO3 (POS) [-1.644 Log (CFU/g)] was obtained at the output level of the retail distribution module.

d SD refers to the standard deviation (sigma) in Log (CFU/g). SD was assumed to be 0.8 Log (CFU/g) in the case of CSS and 0.2 or 0.5 Log (CFU/g) in the case of frankfurters. However, the SD for the frankfurters PO2 (End of manufacture) [0.71 Log (CFU/g)] was calculated following procedures by Zwietering and others (2010) with data obtained from the output module at that level.

e The sample size was for the most part 25g following the Codex standard for *L. monocytogenes*. A sample size of 5 grams was used only in one case to show the effect on the sensitivity of the method and number of samples (n) to be tested.

f The sensitivity of the method (m), also known as the microbiological limit, separates acceptable lots from unacceptable lots in a 2-class plan. In the case of the Codex standard for *L. monocytogenes* for foods that support growth, requires m=absence in 25g [<0.04cfu/g or -1.4 Log (CFU/g)].

g The number of samples (n) required to achieve specific probability of rejecting (P_{rej}) the lot was calculated using the ICMSF (2014, 2016) sampling tool for the desired P_{rej} which would provide 95% confidence that a lot of food would be rejected under the specified conditions.

h Amount of food that contains one pathogen (g) refers to the geometric median. This was calculated for *L. monocytogenes* using the ICMSF (2014, 2016) sampling tool.

Table 7.14 Food safety data sheet for frankfurters

Product Name(s)	Frankfurters, Beef Frankfurters (standard), franks, hot dogs (common name), wieners, or bologna (USDA 2013)
Product Description, including Important Food Safety Characteristics	RTE hotdog (or hot dog) products are defined in 9 CFR 430.1 as "a ready-to-eat meat or poultry frank, frankfurter, or wiener, such as a product defined in 9 CFR 319.180 and 319.181" (9 CFR 430.1). Beef trimmings mixed with minor ingredients (see below) blended and emulsified to a smooth consistency to produce a high moisture, fully cooked, not shelf stable, ready-to-eat (RTE) product. Applicable standards of identity: 9 CFR 319.180 (Frankfurters). Cooked sausages (e.g., bratwurst), as defined in 9 CFR 319.140, would be considered RTE, but would not be considered to be deli or hotdog products. In other words, "hotdog products" or "typical hotdogs" as defined in the *Listeria* rule (i.e., post-lethality exposed RTE) "are RTE meat or poultry franks, frankfurters, wieners per 9 CFR 319.180 standard (thus, does not include products like bratwurst, polish sausage, other cooked sausages covered by 9 CFR 319.140)." (USDA/FSIS 2003)
Ingredients	Beef pre-blend, water, ice, modified food starch (ingredient in excess amount permitted in regular beef frankfurters), contains 2% or less of: salt, sodium lactate, hydrolyzed soy protein, spice blend (including flavorings, paprika), potassium chloride, sodium diacetate, sodium erythorbate, and sodium nitrite.
Allergens	Soy (hydrolyzed soy protein)
Packaging Used	Vacuum packed; hermetically sealed; barrier film packaging material
Intended Use	Consumed as purchased (Ready-to-eat [RTE]). RTE product that is intended to be refrigerated and consumed with or without a reheating* step at the consumer level
Intended Consumers	General population At-risk populations (e.g., pregnant woman, elderly populations, and immune-compromised individuals) are encouraged to reheat frankfurters prior to consumption.
Shelf Life	Varies with packaging and storage temperature. Preferred refrigerated storage temperature 30-40°F. Range of 90, 95, 100 days from date of pack kept at 36 degrees F or less
Labeling Instructions	Keep refrigerated printed on immediate product container and shipping container. Keep frozen (if necessary).
Storage and Distribution	Wholesale to retail distributors Product shipped and stored refrigerated (or frozen). Keep frozen (if necessary).
Sampling Plan for *L. monocytogenes*	n=5, c=0, m= absence in 25 g (<0.04 cfu/g), 2-class plan, analytical method (ISO 11290-1) at the end of production

Source: Format adapted from FSPCA (2017)

Figure 7.1 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach risk assessment derived flowchart baseline model for the frankfurters process and *L. monocytogenes*

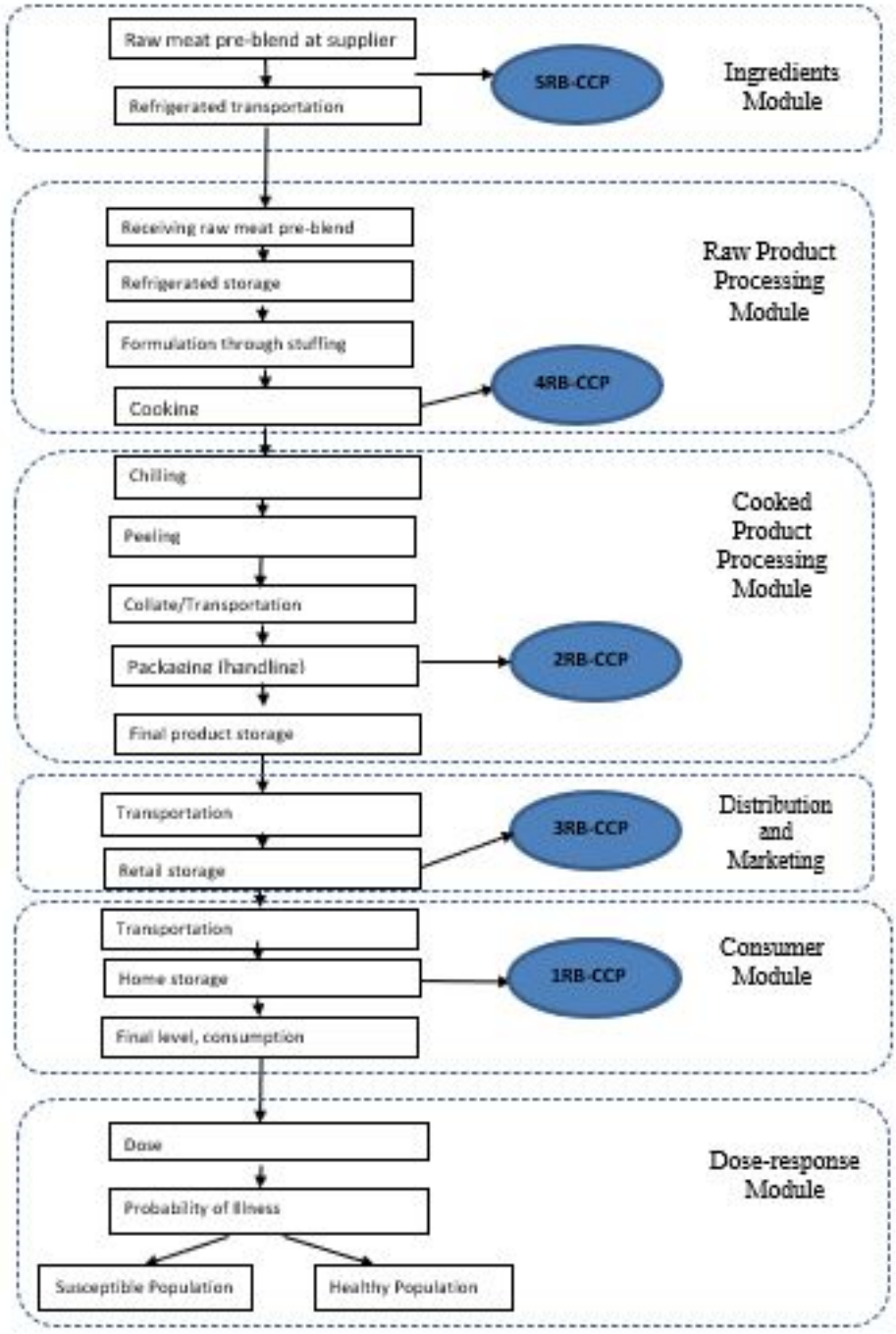


Figure 7.2 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach risk assessment derived flowchart model for the cold smoked salmon process and *L. monocytogenes*

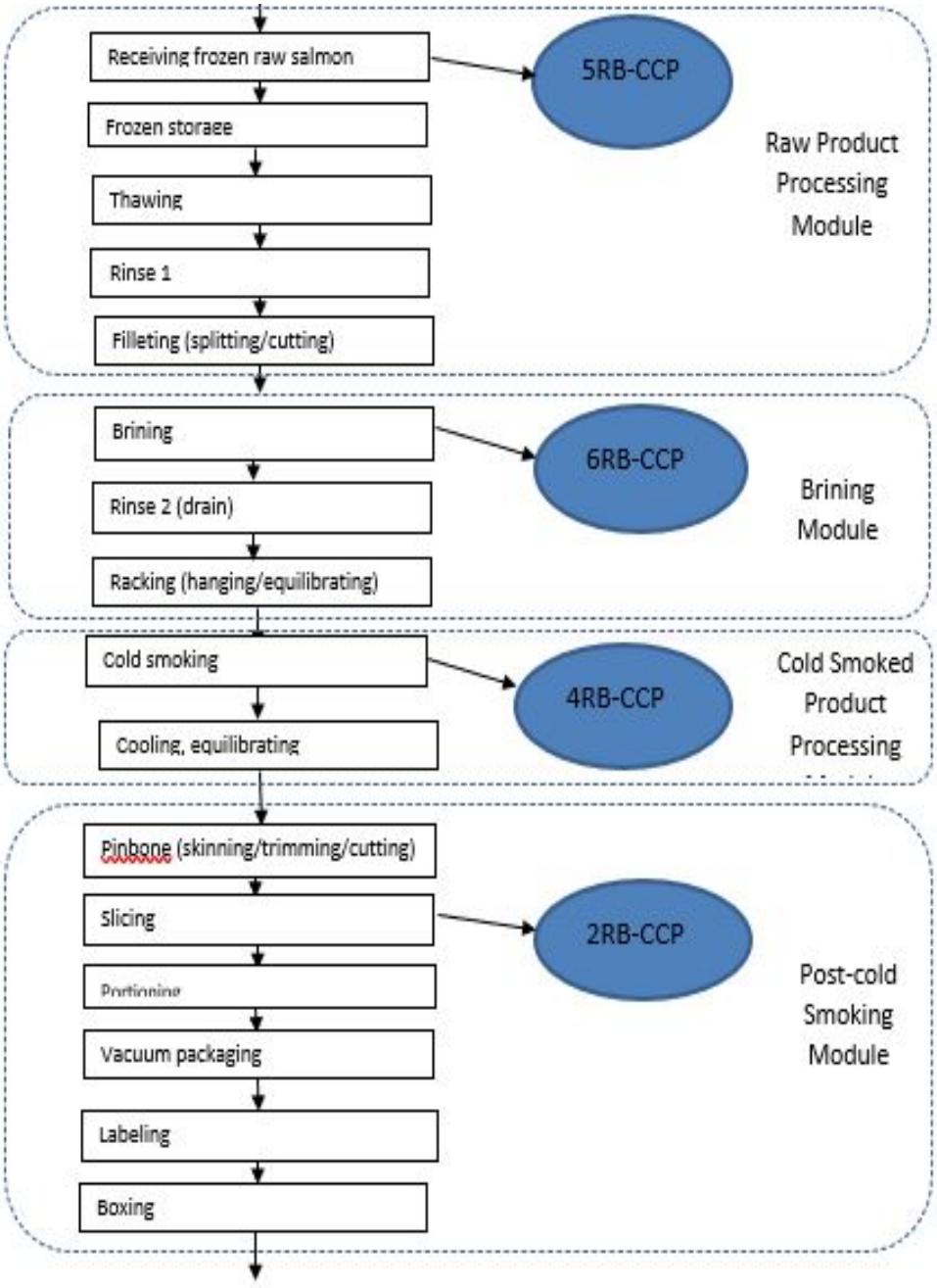


Figure 7.2 Continued

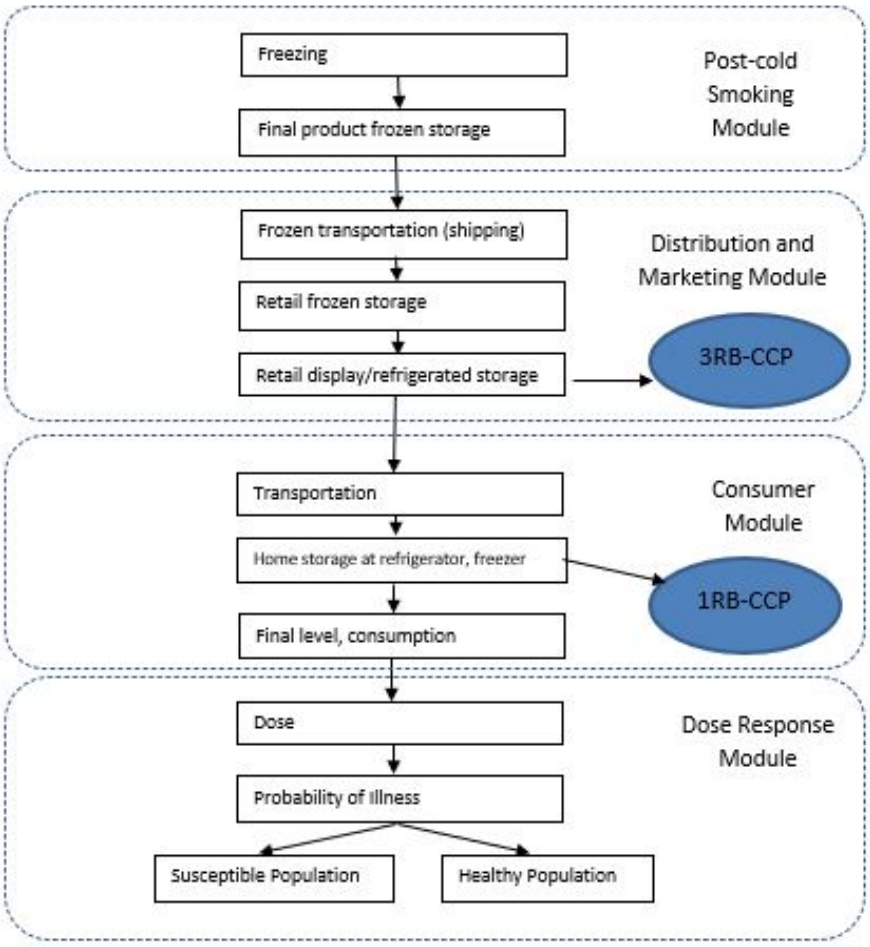


Figure 7.3 Comparison between the risk assessment derived baseline model for the frankfurters not reheated (FNR) and the what-if scenario frankfurters reheated (FR)

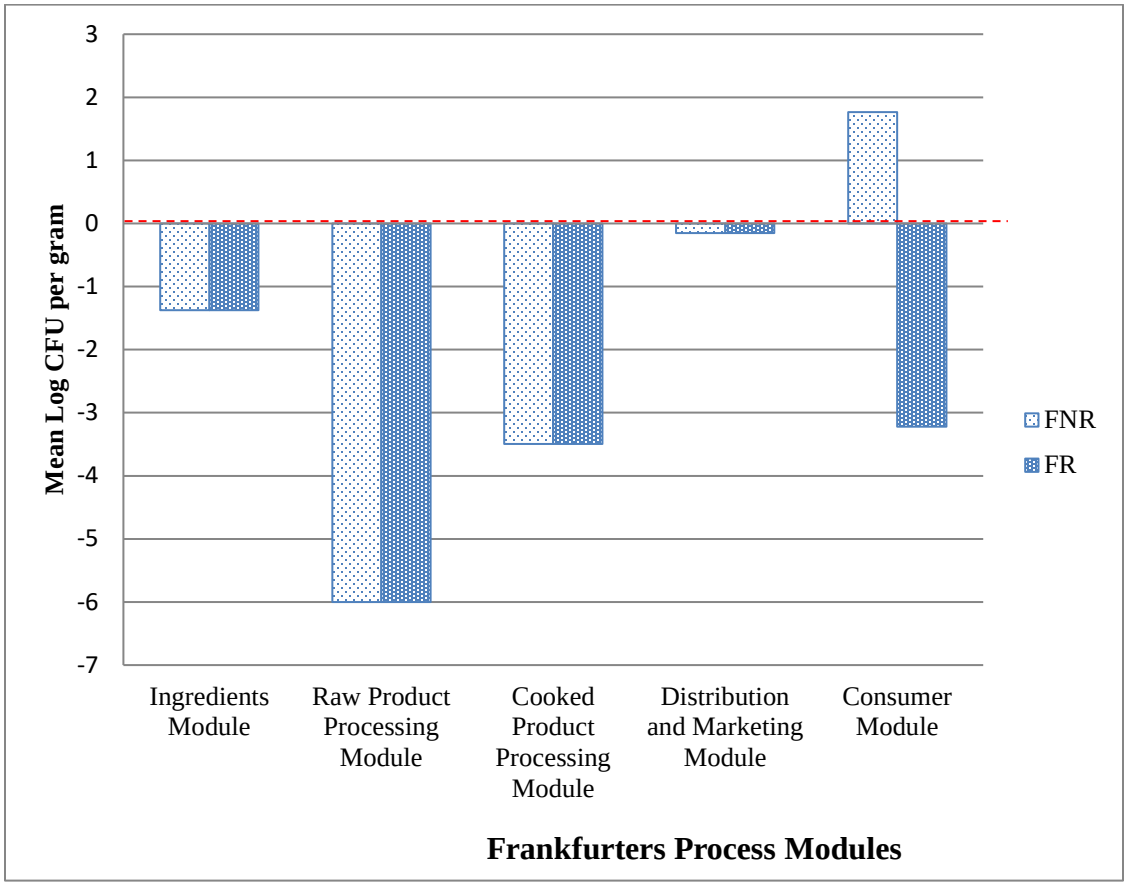


Figure 7.4 Comparison between the risk assessment derived baseline model for the cold-smoked salmon (CSS) process and more stringent what-if scenarios

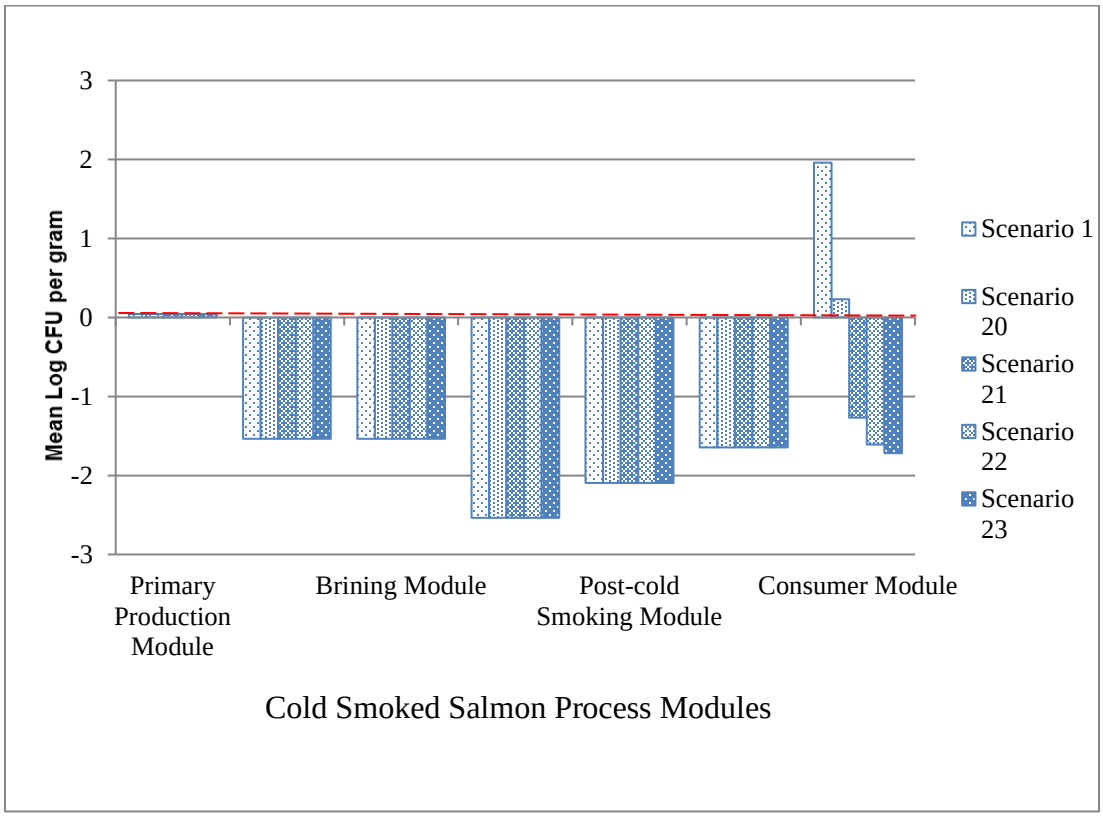


Figure 7.5 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach modules for the risk assessment derived model for the frankfurters process and *L. monocytogenes*

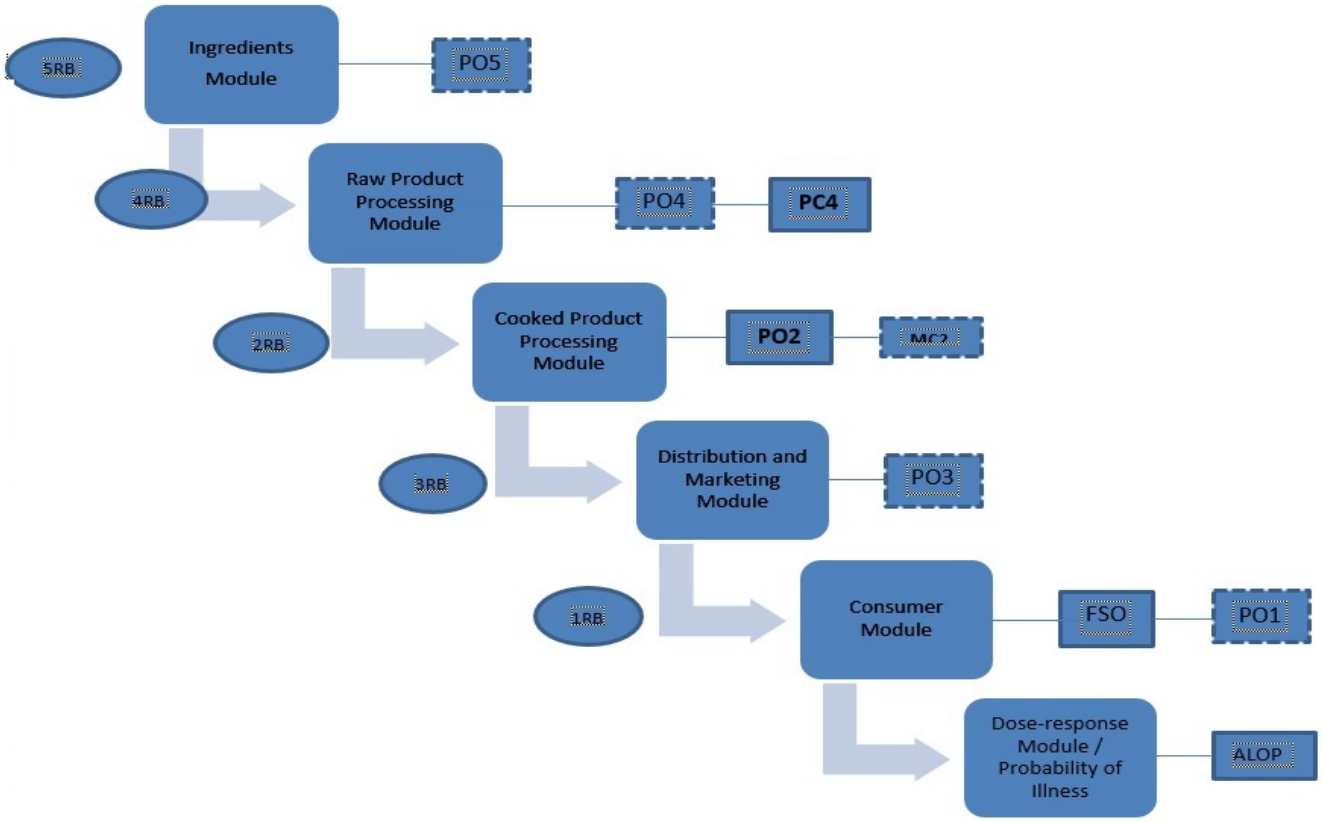


Figure 7.5 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach modules for the risk assessment derived model for the frankfurters process and *L. monocytogenes* (Continued)

$H_0 - \Sigma R + \Sigma I \leq FSO$: ICMSF equation

H_0: Initial level of the hazard

ΣR: Total (cumulative) reduction of the hazard

ΣI: Total (cumulative) increase of the hazard

$\leq$: Preferably less than, but at least equal to

FSO: Food Safety Objective

FSO, H_0, ΣR, and ΣI are expressed in $\log_{10}$ units.

PO: Performance Objective

PC: Performance Criterion (e.g., PC4=6D log R)

MC: Microbiological Criterion

ALOP: Appropriate Level of Protection

Derived ICMSF equations for prioritized risk management metrics

corresponding to each prioritized risk-based critical control point (#RB-CCP or #RB)

1RB-CCP (1RB): $FSO \geq PO1 \geq H_{0\text{-}1} - \Sigma R1 + \Sigma I1$

2RB-CCP (2RB): $PO2 = H_{0\text{-}2} - \Sigma R2 + \Sigma I2$

3RB-CCP (3RB): $PO3 = H_{0\text{-}3} - \Sigma R3 + \Sigma I3$

4RB-CCP (4RB): $PO4 = H_{0\text{-}4} - \Sigma R4 + \Sigma I4$

5RB-CCP (5RB): $PO5 = H_{0\text{-}5} - \Sigma R5 + \Sigma I5$

Figure 7.6 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach modules for the risk assessment derived model for the cold-smoked salmon process and *L. monocytogenes*

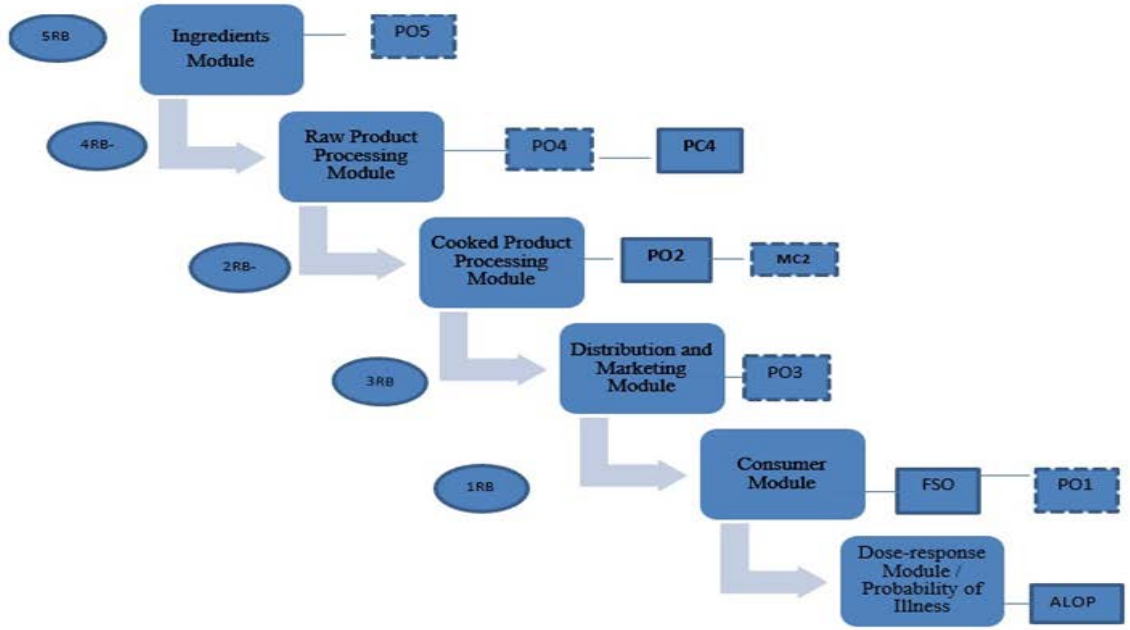


Figure 7.6 Risk-based critical control points (RB-CCPs) located in order of priority within the systems approach modules for the risk assessment derived model for the cold-smoked salmon process and *L. monocytogenes*

Where:

$H_0 - \Sigma R + \Sigma I \leq FSO$: ICMSF equation

H_0: Initial level of the hazard

ΣR: Total (cumulative) reduction of the hazard

ΣI: Total (cumulative) increase of the hazard

$\leq$: Preferably less than, but at least equal to

FSO: Food Safety Objective

FSO, H_0, ΣR, and ΣI are expressed in $\log_{10}$ units.

PO: Performance Objective

PC: Performance Criterion (e.g., PC4=1D log R)

MC: Microbiological Criterion

ALOP: Appropriate Level of Protection

Derived ICMSF equations for prioritized risk management metrics corresponding to each prioritized risk-based critical control point (#RB-CCP or #RB)

1RB-CCP (1RB): $FSO \geq PO1 \geq H_{0\text{-}1} - \Sigma R1 + \Sigma I1$

2RB-CCP (2RB): $PO2 = H_{0\text{-}2} - \Sigma R2 + \Sigma I2$

3RB-CCP (3RB): $PO3 = H_{0\text{-}3} - \Sigma R3 + \Sigma I3$

4RB-CCP (4RB): $PO4 = H_{0\text{-}4} - \Sigma R4 + \Sigma I4$

5RB-CCP (5RB): $PO5 = H_{0\text{-}5} - \Sigma R5 + \Sigma I5$

6RB-CCP (6RB): $PO6 = H_{0\text{-}6} - \Sigma R6 + \Sigma I6$

Figure 7.7 Selected food safety risk management metrics in the systems approach modules for the risk assessment derived model for the frankfurters process and *L. monocytogenes*

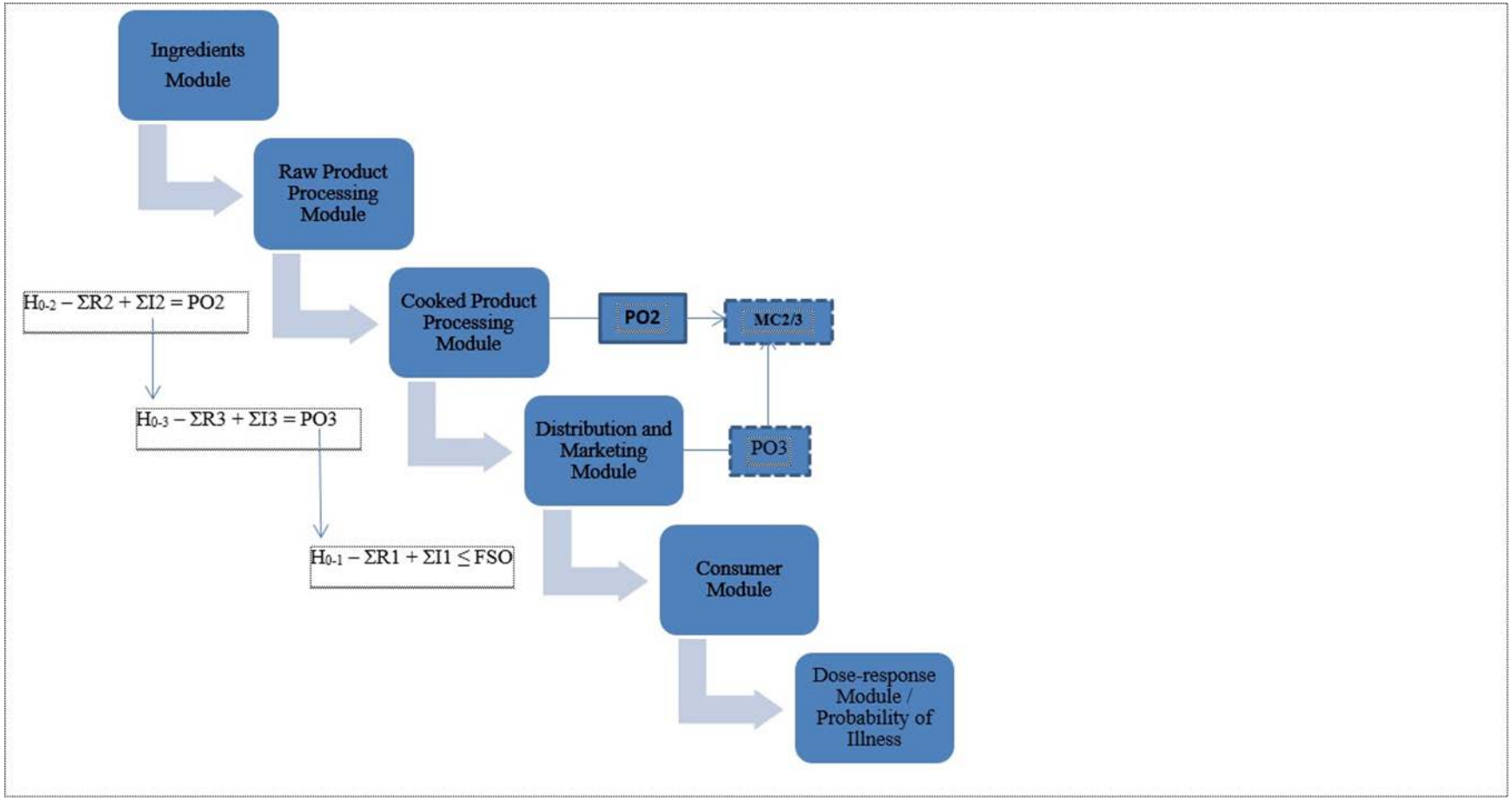


Figure 7.7 Selected food safety risk management metrics in the systems approach modules for the risk assessment derived model for the frankfurters process and *L. monocytogenes*

Where:

PO: Performance Objective

MC: Microbiological Criterion

$H_0 - \Sigma R + \Sigma I \leq FSO$: ICMSF equation

H_0: Initial level of the hazard

ΣR: Total (cumulative) reduction of the hazard

$\leq$: Preferably less than, but at least equal to

ΣI: Total (cumulative) increase of the hazard

FSO, H_0, ΣR, and ΣI are expressed in $\log_{10}$ units.

Please note that the PO of one point of the food chain may be the H_0 of the following one (e.g., $H_{0\text{-}1}$ = PO3) Thus, the input for the consumer module is the output for the retail distribution module.

Derived FSO/PO equations for each RB-CCP based on sensitivity analysis derived priority level

1RB-CCP (1RB): $FSO \geq H_{0\text{-}1} - \Sigma R1 + \Sigma I1$

2RB-CCP (2RB): $PO2 = H_{0\text{-}2} - \Sigma R2 + \Sigma I2$

3RB-CCP (3RB): $PO3 = H_{0\text{-}3} - \Sigma R3 + \Sigma I3$

4RB-CCP (4RB): $PO4 = H_{0\text{-}4} - \Sigma R4 + \Sigma I4$

5RB-CCP (5RB): $PO5 = H_{0\text{-}5} - \Sigma R5 + \Sigma I5$

RB-CCP = RB = Risk-based Critical Control Point

#RB-CCP = #RB = number of priority for the RB-CCP or RB

Figure 7.8 Selected food safety risk management metrics in the systems approach modules for the risk assessment derived model for the cold-smoked salmon process and *L. monocytogenes*

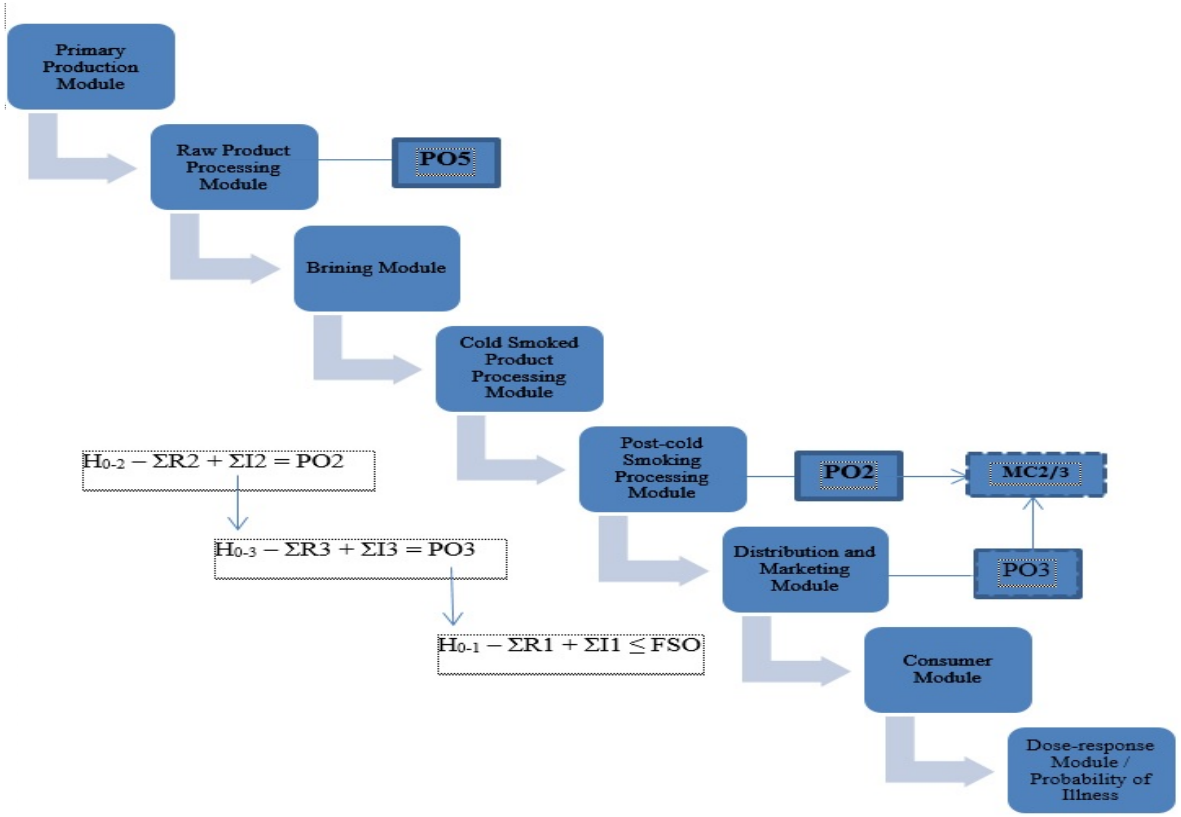


Figure 7.8 Selected food safety risk management metrics in the systems approach modules for the risk assessment derived model for the cold-smoked salmon process and *L. monocytogenes* (Continued)

Where:

PO: Performance Objective

MC: Microbiological Criterion

$H0 - \Sigma R + \Sigma I \leq FSO$: ICMSF equation

H0: Initial level of the hazard

ΣR: Total (cumulative) reduction of the hazard

$\leq$: Preferably less than, but at least equal to

ΣI: Total (cumulative) increase of the hazard

FSO, H0, ΣR, and ΣI are expressed in log10 units.

Please note that the PO of one point of the food chain may be the H0 of the following one (e.g., H0-1 = PO3) Thus, the input for the consumer module is the output for the retail distribution module.

Derived FSO/PO equations for each RB-CCP based on sensitivity analysis derived priority level

1RB-CCP (1RB): $FSO \geq H0\text{-}1 - \Sigma R1 + \Sigma I1$

2RB-CCP (2RB): $PO2 = H0\text{-}2 - \Sigma R2 + \Sigma I2$

3RB-CCP (3RB): $PO3 = H0\text{-}3 - \Sigma R3 + \Sigma I3$

4RB-CCP (4RB): $PO4 = H0\text{-}4 - \Sigma R4 + \Sigma I4$

5RB-CCP (5RB): $PO5 = H0\text{-}5 - \Sigma R5 + \Sigma I5$

6RB-CCP (6RB): $PO6 = H0\text{-}6 - \Sigma R6 + \Sigma I6$

RB-CCP = RB = Risk-based Critical Control Point

#RB-CCP = #RB = number of priority for the RB-CCP or RB.

Appendices

Appendix 1: Summary of standardized data survey used during three data collection visits to request available data

In review of the scientific literature and data collection visits, the steps of the frankfurter process at the facility level were divided into the following four modules: Module I Ingredients, Module II Raw product processing, Module III Cooked product processing, and Module IV Distribution and transport. Data related to the variables associated with the manufacturing steps included in each of these modules was acquired as they were likely to affect the outputs regarding survival, growth, and thermal inactivation of *Listeria monocytogenes*. The data collected was from the specific processing lines when available. When data was not available, data and/or models from the literature were used and substituted. If these were likewise unavailable, then realistic assumptions were made in consultation with experts.

The data requested in the standardized survey for Modules I-IV included:

Module I Step 1: Receiving Raw Materials

- Initial microbial profiles, including prevalence of *L. monocytogenes* and/or *Listeria* spp. in the meat pre-blend and other ingredients

- Water activity (a_w) and pH in the meat pre-blend and other ingredients

- Antimicrobials and salt percentages (NaCl and nitrate) in the meat pre-blend and other ingredients

- Temperature and time history of meat pre-blend prior to arrival at reception (shipment conditions, seasonality)

- Temperature, water activity, storage history, and microbiological profile of spices, paprika, and hydrolyzed soy protein and other ingredients used in frankfurter formulation

Module II Step 2: Refrigerated Storage

- Temperature of ingredients and/or circular temperature monitoring charts from the raw meat room

- Duration of storage

Module II Step 3: Formulation through Stuffing

- Fat content and variation on fat content (measurements) to develop a distribution

- Antimicrobials and salt percentages (NaCl, sodium erythorbate, and nitrite)

- Fat content and variation on fat content (measurements) to develop a distribution

- Antimicrobials and salt percentages (NaCl, sodium erythorbate, and nitrite)

- Meat temperature and duration of holding of between formulation and processing

- Variation of formulation from batch-to-batch, i.e., how does the proportion of each ingredient (hydrolyzed soy protein, spices, garlic, paprika, flavorings) vary from batch-to-batch

- Conditions under which the mixture of ingredients are held during this period (Temperature, relative humidity, time)

- Diameters of frankfurters, including an estimate of variability

- Storage Temperature and Times for the meat pre-blend (after formulation and scaling)

- Salt percentages in Lactate blend

- Quality of the water utilized to make the ice and to incorporate directly in product. Sanitary data for the ice-maker/ equipment in contact with this water (*E.coli* data collected).

- Monitoring procedures/forms at Vacuum/Magnets/Emulsify step in the process.

Module II Step 4: Cooking

- Temperature and relative humidity profile/map in the smoking equipment, including an estimate of the run-to-run variability. Actual reading variability from 4 different seasons collected. Data collected for all the ovens and processes using SAS 9.1.

- Time in the oven (cooking-smoking equipment), including estimate of run-to-run variability

- Time and temperature profile of the product including data on the internal temperature reached in the product during heat treatment and an estimate of the variability of that value. Data collected and processed using SAS 9.1 shown the minimum, most likely and maximum values for temperatures as: 71,1, 73,3, 75,5 °C

- pH monitoring charts for liquid smoke

- Time of the smoking shower (liquid smoke zone)

Module III Step 5: Cooling

- Microbial data of cooling water: Coliforms and *E. coli* (5 samples tested monthly - state labs). Chlorine levels tested daily.

- Initial and final temperature of product

- Cooling curve/rates/time

Module III Step 6: Brine Chilling

- Microbiological quality of brine

- pH and acidulant (citric acid) concentration

- Salt concentration of brine including variability

- Temperature of brining water including variability

- Time of product in contact with brine

- Frequency of sanitation of the brine chill chamber and any environmental microbial data post-cleaning

Module III Step 7: Peeling

- Temperature on the surface before and after peeling. The equipment operates with a hot blade and it has a steaming system.

- Sanitation swabs/food contact surface testing

Module III Step 8: Chilling

- Cooling curves and final temperatures reached by product

- Holding times

-Area in contact with the product collected for one production line including: peeler tables, conveyors, and “short slipstick”

Module III Step 9: Final Product Composition and Storage Conditions

- Product and room temperatures

- Inventory holding times

- Product composition (a_w, fat, salt concentration, pH)

- Change in product temperature during storage

- Temperature/time profile/map of the pallets within the cold room

Module III Step 10: Distribution

- Temperature mapping of refrigerated compartment of trucks

- Time records

In addition to the data requested for the specific variables modeled for each steps of the process, additional information was collected for the sole purpose of research. These data were not included in Appendix 1 as it was considered proprietary in accordance with the Non-Disclosure Agreement.

Appendix 2: Incidence Studies for *Listeria monocytogenes* in Raw Ingredients and Ready-to-Eat Products

Appendix 2.1 Weighted Scheme Determination for the Incidence Studies of Listeria monocytogenes

The estimation of incidence from the literature employed a scheme for weighting the relative importance of the studies based on several criteria. This allowed use of literature data to be maximized while considering changing practices in the meat and seafood industries.

Factors considered included number of samples (n), geographic location (gw), and study date (dw) using the following formula: **Study weight = n * gw * dw**

Based on these criteria, literature studies were weighted according to the following criteria and rationale:

Number of samples (n): Larger studies with more representative samples may provide a better estimate of the incidence. Incidence data was presented as both the numerator and denominator to allow for differences in the size of the populations upon which the proportions are based. Rationale: All pertinent publications were included, regardless of the number of samples. Publications with a larger number of samples were given more weight proportionally to the total number of samples (n). The specific study weight was the number of samples (n) of each study. For example, in the case of frankfurters, a study with 117 samples (i.e., Wang and Muriana) was considered to have n=117 and a study with 32800 samples (i.e., Wallace and others) was considered to have n=32800.

Geographic weight (gw): Pertinent publications were considered regardless of the country of origin. Based on the globalized market, publications from regions that contribute to the United States food supply were assigned more weight. Rationale: A value of 1 was

used unless the study was conducted in a region and for foods (i.e., smoked fish) for which there is little or no contribution (importation) to the United States food supply—in which case a value of 0.5 was used.

Weight for the date of the study (dw): The year of publication of the study (i.e., not the year the samples may have been processed) were considered to weight the incidence studies. All pertinent publications were included regardless of the year of publication. Rationale: *L. monocytogenes* and other *Listeria* spp. have been isolated from seafood on a regular basis since 1987 (Ben Embarek 1994). Evidence exists that improved sanitation and HACCP programs have reduced the contamination of foods since the recognition of the public health problem from *L. monocytogenes* in the 1980's (FDA/USDA 2003). Therefore, recent publications were assigned more weight. A value of 1 was used for the most recent studies published (2006-2016); a value of 0.8 was used for studies published between 2000 and 2005; a value of 0.6 was used for studies published between 1994 and 1999; and a value of 0.4 was used for studies published in or before 1993. The weighted scheme was applied in the same manner to all pertinent data collected from the literature. When needed, weighted means and standard deviations were calculated using the formulas described in Table A2.1.

Appendix 2.2 Incidence Studies for Listeria monocytogenes *in Raw Ground Beef and Frankfurters*

Appendix 2.2.1 Incidence Studies for *Listeria monocytogenes* in Raw Ground Beef

The USDA Baseline Survey results for ground beef as corrected and cited by the ICMSF (2002) were used to estimate the incidence of *L. monocytogenes* in raw ground beef. The calculated weighted occurrence of *L. monocytogenes* in ground beef was 14.4%. Table

A2.2 summarized the results of the 563 samples analyzed by the USDA and included the calculations for the weighted occurrence of *L. monocytogenes* in ground beef.

Appendix 2.2.2 Incidence Studies for *Listeria monocytogenes* in Frankfurters

The literature for the incidence of *L. monocytogenes* on frankfurters was reviewed and available studies were consolidated in Table A2.3. Wallace and others (2003) was considered to be the most relevant source of data for vacuum-sealed packages of frankfurters because of the number of samples collected (n=32,800) and because this data was volunteered and collected specifically for frankfurters and *L. monocytogenes* from 12 commercial manufacturers over a two-year period in the United States. The 12 producers included nine large and three small plants located in 10 states. In total, 532 of 32,800 (1.6%) packages of frankfurters tested positive for *L. monocytogenes.* This incidence considered all the individual processing facilities with minimum and maximum incidence of 0% and 16% (plant 133), respectively (Wallace and others 2003). The cumulative frequency for the incidence of *L. monocytogenes* on frankfurters based on Wallace and others (2003) is depicted in Figure A2.1.

The calculated total weighted incidence of *L. monocytogenes* from all studies including Wallace and others (2003) was 1.88%, which is slightly higher than the incidence calculated from the Wallace and others study alone of 1.62%. This calculated weighted value is higher because the other studies are older than Wallace and others (2003) and therefore, had higher incidence of *L. monocytogenes*. As explained in Chapter 2, there has been a reduction in the occurrence of *L. monocytogenes* in the meat industry including frankfurters. The weight applied due to the date of publication (dw) reflected the reduction of contamination due to the improved sanitation and HACCP programs in more recent years. The incidence of studies other than Wallace and others (2003) have a minor effect on the

overall incidence due to their combined low number of samples which represents approximately 10% compared to Wallace and others (2003).

It is noteworthy that Wallace and others (2003) is not a quantitative study and other studies have been considered to determine the distribution of the levels (CFU/g) of *L. monocytogenes* in frankfurters (e.g., Wang and Muriana 1994). The probability distribution for the level of *L. monocytogenes* in contaminated frankfurters is reported in Figure A2.2.

Appendix 2.3 Incidence Studies for Listeria monocytogenes *in Raw Salmon and Cold-Smoked Salmon*

Appendix 2.3.1 Incidence Studies for *Listeria monocytogenes* in Raw Salmon

The level of *L. monocytogenes* contamination for raw salmon was estimated from pertinent studies found in the literature (Table A2.4). These studies revealed a relatively high but variable incidence of *L. monocytogenes.* The incidence of *L. monocytogenes* in raw salmon varied from 0% to 100% with a weighted mean of 20.34% and a weighted standard deviation of 20.83. The formulas for these calculations were described in Table A2.1.

Appendix 2.3.2 Incidence Studies for *Listeria monocytogenes* in Cold-Smoked Salmon

The level of *L. monocytogenes* contamination for cold-smoked salmon was estimated from numerous studies and surveys carried out worldwide during the last 20 years (Table A2.5). These studies revealed a relatively high but variable incidence of *L. monocytogenes* in smoked fish ranging from 0% to 78.7%. The incidence of *L. monocytogenes* in cold-smoked salmon had a weighted mean of 12.36%. The cumulative frequency for the incidence of *L. monocytogenes* in cold-smoked salmon is depicted in Figure A2.3. In addition, the probability

distribution for the level of *L. monocytogenes* in contaminated cold-smoked salmon is reported in Figure A2.4.

Table A2.1 Formulas for the weighted mean and weighted standard deviation

Formula	Reference
Weighted Mean = $\bar{X}_w = \frac{\sum_{i=1}^{n}(Wi*Xi)}{\sum_{i=1}^{n}(Wi)}$	NIST DATAPLOT Reference Manual (P. 2-65)
Weighted SD = $sd_w = \sqrt{\frac{\sum_{i=1}^{n} Wi(Xi-\bar{X}w)^2}{\frac{(N'-1)\sum_{i=1}^{n} Wi}{N'}}}$	NIST DATAPLOT Reference Manual (P. 2-66)
where: Xi= incidence percentage Wi=weight of the ith observation N' = number of non-zero weights $\bar{X}$w = weighted mean of the observations	

Table A2.2 USDA Baseline Survey Results for Occurrence of *Listeria monocytogenes* in ground beef

	Ground Beef	Weighted occurrence of *Listeria monocytogenes* in ground beef
Incidence based on 25 g samples	(563 samples)	
	18%	$\frac{(563 * 1 * 0.8) * 18\%}{563} = 14.4\%$

These data for ground beef indicates that only 99 samples out of 563 were found to contain *L. monocytogenes* which revealed an incidence of 18% based on 25 g samples. Samples found positive by the qualitative method were further analyzed to determine the number of *L. monocytogenes* per gram of ground beef. The results for the positive samples were as follows:

Number of *L. monocytogenes* per gram	Ground beef (99 samples)
< 0.03	45.2
0.03-0.29	0.0
0.3-2.9	30.2
3.0-29.9	15.0
30-299.9	9.6
300 or higher	3 samples had > 110/g

Upon further analysis of the 99 samples, this data show that 90.4% had fewer than 30 CFU per gram and only 3 of the total 563 samples had greater than 110 *L. monocytogenes* per gram, which was the upper limit of detection used for testing.

Table A2.3 Incidence Studies for *Listeria monocytogenes* in Frankfurters

Study Reference	Meat Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Hayes and others 1992	Hotdogs	USA	40	28	1	0.4	R	16	30	12
Levine 2000	Hotdogs	USA	1593	1516	1	0.8	P	1274.4	4.83	3.86
Levine 2001	Hotdogs	USA	1800	1766	1	0.8	P	1440	1.88	1.50
Ng and Seah 1995	Franks, chicken & pork	Singapore	78	73	1	0.6	P/R	46.8	6.41	3.85
Oregon State Dept. of Agriculture 2001	Hotdog	USA	11	11	1	0.8	R	8.8	0	0
Oregon State Dept. of	Hotdogs, beef	USA	3	3	1	0.8	R	2.4	0	0

Study Reference	Meat Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Agriculture 2001										
Oregon State Dept. of Agriculture 2001	Hotdogs, turkey	USA	3	3	1	0.8	R	2.4	0	0
Qvist and Liberski 1991	Frankfurter	Denmark	64	56	1	0.4	R	25.6	12.5	5
Samelis and Metaxopoulos 1999	Frankfurter, sausage type	Greece	8	8	1	0.6	P	4.8	0	0
Wallace and others 2003	Frankfurter	USA	32800	32268	1	0.8	P	26240	1.62	1.30
Wang and Muriana, 1994	Hotdogs	USA	117	93	1	0.6	R	70.2	20.51	12.31
Wenger and others 1990	Wieners, turkey	USA	46	25	1	0.4	P	18.4	45	18

Study Reference	Meat Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
TOTAL			36563	35850					1.95	1.88

Table A2.4 Incidence Studies for *Listeria monocytogenes* in Raw Salmon

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Dass 2011	Raw salmon	Ireland	60	43	1	1	P	60	28	28
Dauphin and others 2001	Salmon, cut & raw	France	1	0	1	0.8	P	0.8	100	80
Dauphin and others 2001	Surfaces, raw salmon (wholE)	France (origin Scotland)	8	1	1	0.8	P	6.4	88	70.40
Dauphin and others 2001	Surfaces, raw salmon (fillets)	France (origin Norway)	7	1	1	0.8	P	5.6	86	68.80
Dauphin and others 2001	Surfaces, raw salmon	France	18	16	1	0.8	P	14.4	11	8.8
Davies and others 2001	Salmon/raw seafood	Great Britain	5	5	1	0.8	P	4	0	0
Guyer and Jemmi, 1990	Raw salmon	Switzerland	236	168	1	0.4	P	94.4	29	11.60
Kamat and Nair, 1994	Salmon	India (origin Norway)	1	1	0.5	0.6	P	0.3	0	0
Oregon State Dept of Agric., 2001	Salmon	USA	8	8	1	0.8	R	6.4	0	0

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Soriano and others2001	Salmon, raw	Spain	4	4	1	0.8	R	3.2	0	0
Vogel and others 2001	Salmon, raw	Denmark	30	30	1	0.8	P	24.0	0	0
Vogel and others 2001	Salmon, raw	Denmark	185	169	1	0.8	P	148.0	9	7.20

Table A2.4 Continued

Study Reference	Seafood Product	Count ry of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Yamazaki and others 2000	Salmon, raw	Japan	12	11	1	0.8	R	9.6	8	6.40
Ben Embarek and others 1997 cited/by Miettinen 2006, Dass 2011, and FDA Web	Salmon, live & farmed (skin and belly cavity swabbed)	Norway	10	10	1	0.6	P	6.0	0	0
Medrala and others 2003 cited/by Miettinen 2006 and Dass 2011	Salmon, raw	Norway	46	44	1	0.8	P	36.8	4	3.20
Eklund and others 1995 cited/by Miettinen 2006 and Dass 2011	Salmon, skin 25g	USA	46	16	1	0.6	P	27.6	65	39

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Eklund and others 1995 cited/by Miettinen 2006	Salmon, flesh under skin (1 cm thick) 25g	USA	22	22	1	0.6	P	13.2	0	0
Eklund and others 1995 cited/by Miettinen 2006	Salmon, Belly-cavity lining 25g	USA	7	7	1	0.6	P	4.2	0	0
Eklund and others 1995 cited/by Miettinen 2006	Salmon, head 25g	USA	17	9	1	0.6	P	10.2	47	28.20
Eklund and others 1995 cited/by Miettinen 2006 and Dass 2011	Salmon, tail 25 g	USA	9	3	1	0.6	P	5.4	67	40.20

Table A2.4 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Eklund and others 1995 cited by Miettinen	Salmon, trimmings (belly cavity and belly flaps) 25 g	USA	15	14	1	0.6	P	9.0	7	4.20

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
2006 and Dass 2011										
TOTAL			747					489.5	22.18	20.34

Table A2.5 Incidence Studies for *Listeria monocytogenes* in Cold-Smoked Salmon

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Oregon State Dept of Agriculture, 2001	Unpublished data	USA	168	167	1	0.8	R	134.4	0.60	0.48
Garland, 1995	Smoked salmonids	Tasmania	285	284	1	0.6	P	171	0.35	0.21
Thimothe and others 2004	Smoked fish	USA	233	230	1	0.8	P	186.4	1.29	1.03
Hartemink and Georgsson, 1991	Smoked fish (salmon, minced salmon, herring, trout)	Iceland	31	30	1	0.4	R	12.4	3.23	1.29
Lappi and others 2004	Cold-smoked salmon	USA	36	35	1	0.8	P	28.8	2.78	2.22
Baek and others 2000	Frozen smoked mussels	South Korea	68	65	1	0.8	R	54.4	4.41	3.53

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Hatakka and others 2001	Cold-smoked vacuum-packed fish products	Finland	232	222	1	0.8	R	185.6	4.31	3.45
Gombas and others 2003	Smoked seafood	USA	2644	2530	1	0.8	R	2115.2	4.31	3.45
Inoue and others 2000	Smoked salmon	Japan	92	87	1	0.8	R	73.6	5.43	4.35
Guyer and Jemmi, 1990	Smoked salmon	Switzerland	64	60	1	0.4	P	25.6	6.25	2.50
Teufel and Bendzulla, 1993	Smoked fish	Germany	380	353	1	0.4	R	152	7.11	2.84
Cabedo and others 2008	Smoked salmon	Spain	89	82	1	1	R	89	7.87	7.87

Table A2.5 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Johansson and others 1999	smoked fish products	Finland	78	72	1	0.6	R	46.8	7.69	4.62
Bull and others 2002	Smoked Fish	Australia	388	355	1	0.8	R	310.4	8.51	6.80
Jemmi and others 1992-1993	Smoked fish	Switzerland	1125	1018	1	0.4	R	450	9.51	3.80
Beaufort and others 2007	Cold-smoked salmon	France	1010	906	1	1	R	1010	10.30	10.30
Norton and others 2001	Smoked fish (salmon, sablefish, sea bass)	USA	96	85	1	0.8	P	76.8	11.46	9.17
LaTorre and others 2007	Smoked salmon	Italy	104	11	1	1	R	104	10.58	10.58
Jemmi, 1990 (a)	Smoked salmon	Switzerland	820	732	1	0.4	R	328	10.73	4.29

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Loncarevic and others 1996	Smoked Fish (rainbow trout and salmon)	Sweden	26	23	1	0.6	R	15.6	11.54	6.92
Miettinen and others 2001	Cold-smoked rainbow trout	Finland	25	22	1	0.8	R	20	12.00	9.60
Cordano and others 2001	Shellfish	Chile	268	237	1	0.8	R	214.4	11.57	9.25
Nakamura and others 2004	Cold-smoked salmon	Japan	95	83	1	0.8	R	76	12.63	10.11
Jemmi and others 2002	Smoked fish	Switzerland	1285	1114	1	0.8	R	1028	13.31	10.65

Table A2.5 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Scoglio and others 2000	Smoked fish	Italy	21	18	1	0.8	R	16.8	14.29	11.43
Heinitz and Johnson 1998	Smoked fish and shellfish	USA, Canada, Norway, the Philippines, and UK	1080	929	1	0.6	P	648	13.98	8.39
Norton and others 2000	Cold-smoked salmon	USA	38	32	1	0.8	P	30.4	15.79	12.63
Dillon and others 1994	Smoked seafood Products	Newfound-land	258	215	1	0.6	R	154.8	16.67	10.00
Van Coillie and others 2004	Smoked salmon	Belgium	42	34	1	0.8	R	33.6	19.05	15.24
Cortesi and others 1997	Vacuum-packed sliced cold-smoked salmon	Italy	165	133	1	0.6	R	99	19.39	11.64

Table A2.5 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Dass 2011	Vacuum-packed cold-smoked salmon	Ireland	120	94	1	1	P	120	21.67	21.67
Dominguez and others 2001	Cold-smoked fish (salmon and trout)	Spain	170	132	1	0.8	R	136	22.35	17.88
Yamazaki and others 2000	Smoked seafood (Salmon, herring)	Japan	13	10	1	0.8	R	10.4	23.08	18.46
Jemmi and others 1990 (b)	Smoked/marinated salmon	Switzerland	100	76	1	0.4	R	40	24.00	9.60
Vitas and others 2004	Smoked salmon	Spain	100	72	1	0.8	R	80	28.00	22.40
Uyttendaele and others 2009	Smoked fish	Belgium	90	64	1	1	R	90	28.89	28.89

Table A2.5 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Aguado and others 2001	Smoked salmon	Northern Spain	52	36	1	0.8	R	41.6	30.77	24.62
Farber 1991b	Salmon	USA, Chile, Scotland, Norway, Canada	32	22	1	0.4	P	12.8	31.25	12.50
Di Pinto and others 2010	Smoked salmon	Italy	132	87	1	1	R	132	34.09	34.09
Jørgensen and Huss 1998	Cold-smoked salmon and halibut	Denmark	420	257	1	0.6	R	252	38.81	23.29
Ng and Seah 1995	Smoked mussels	Singapore	2	1	1	0.6	R	1.2	50.00	30.00
Hudson and others 1992	smoked salmon	New Zealand	12	4	1	0.4	R	4.8	66.67	26.67

Table A2.5 Continued

Study Reference	Seafood Product	Country of Origin	No. of samples (n)	No. of negative samples	Geo-graphic weight (gw)	Date of publica-tion weight (dw)	Collection (Processor or Retail)	Study weight (n*dw*gw)	Incid-ence %	Weigh-ted Incid-ence %
Ericsson and others 1997	Gravad or cold-smoked rainbow trout and salmon	Sweden (Outbreak strains)	9	3	1	0.6	R	5.4	66.67	40.00
Vogel and others 2001	Cold-smoked salmon	Denmark	200	56	1	0.8	P	160	72.00	57.60
Eklund and others 1995	Cold-smoked salmon	Seattle, WA, USA	61	13	1	0.6	P	36.6	78.69	47.21
TOTAL			12759					9013.8	12.43	12.36

Table A2.6 Probability Distribution to estimate the initial level of *Listeria monocytogenes* in the raw meat pre-blend destined for frankfurter production

Level in the Process	Unit	Distribution	Reference
Initial Concentration of *L. monocytogenes* in raw meat pre-blend	(CFU/g)	RiskGamma(0.015386,67.306 ,RiskShift(0.019861))	Best Fit distribution developed from USDA Baseline Survey Results for Ground Beef, Ground Turkey, and Ground Chicken (Source: Nationwide Federal Plant Raw Ground Beef Microbiological Survey, August 1993 – March 1994) cited by ICMSF (2002)

Figure A2.1 Cumulative Frequency for the Incidence of *Listeria monocytogenes* on Frankfurters

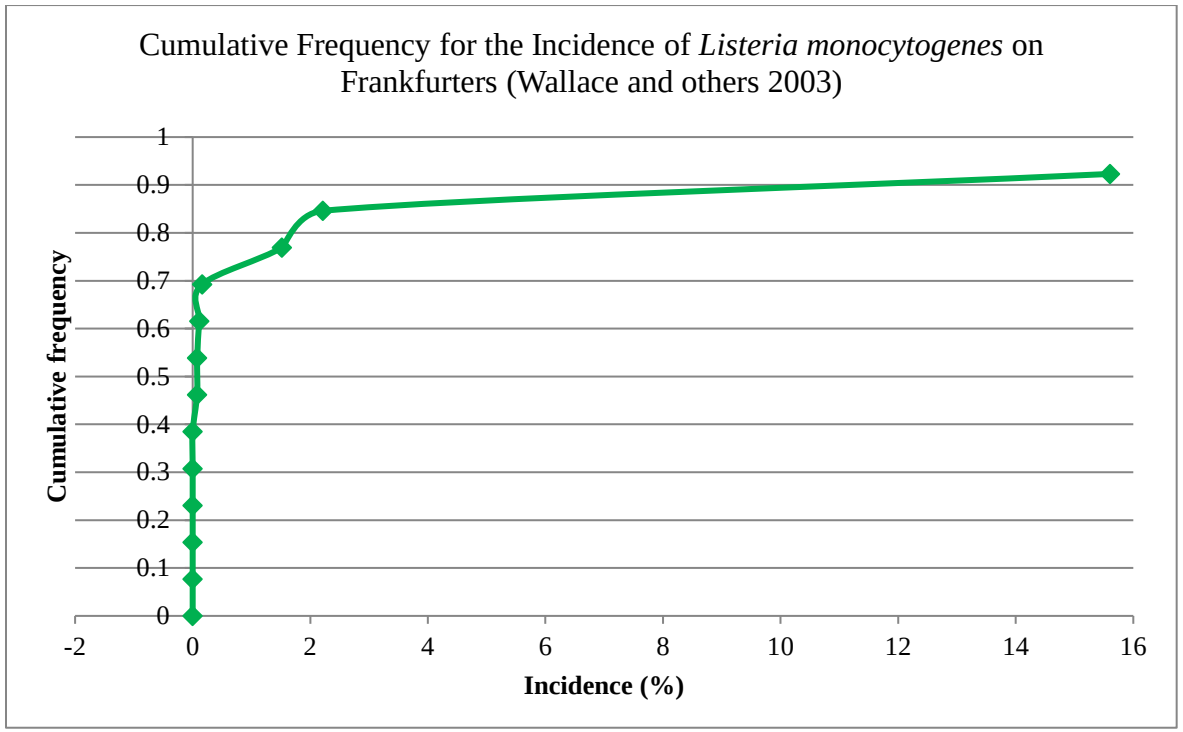


Figure A2.2 Probability Distribution of Different Concentration Levels of *Listeria monocytogenes* in Contaminated Frankfurters

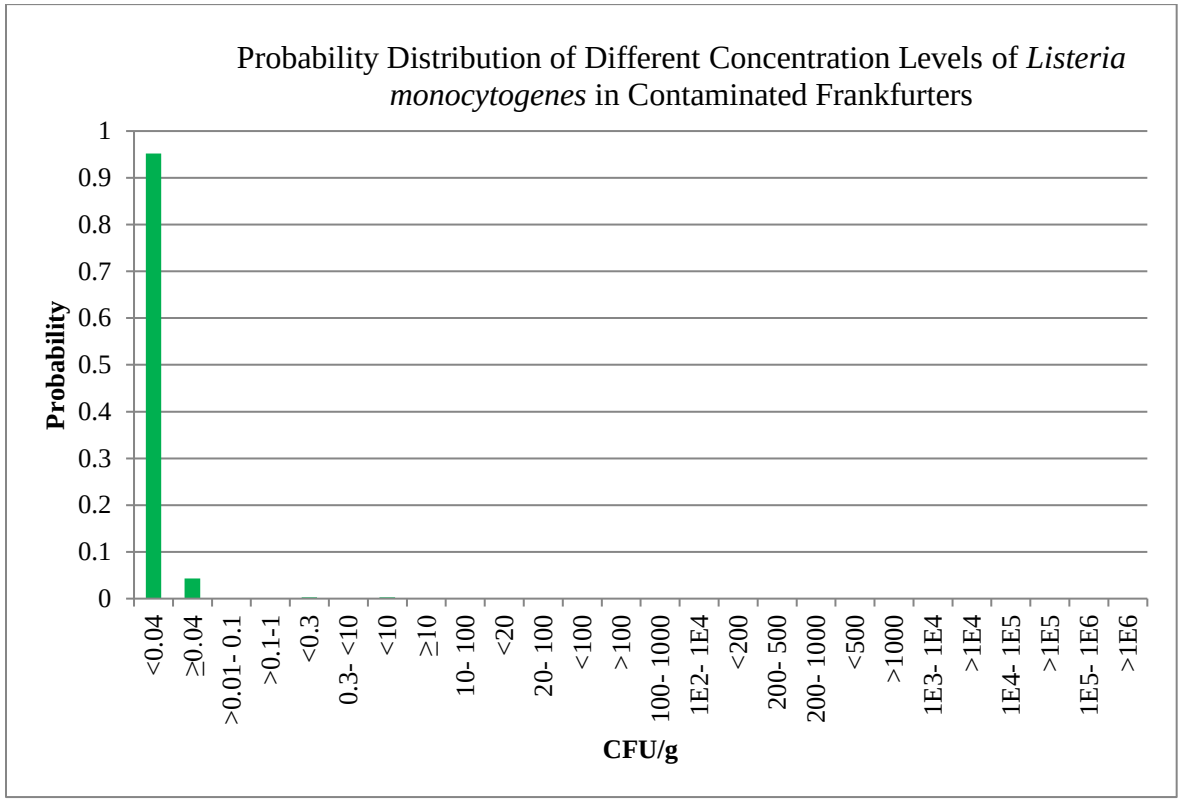


Figure A2.3 Cumulative Frequency for the Incidence of *Listeria monocytogenes* in Cold-Smoked Salmon

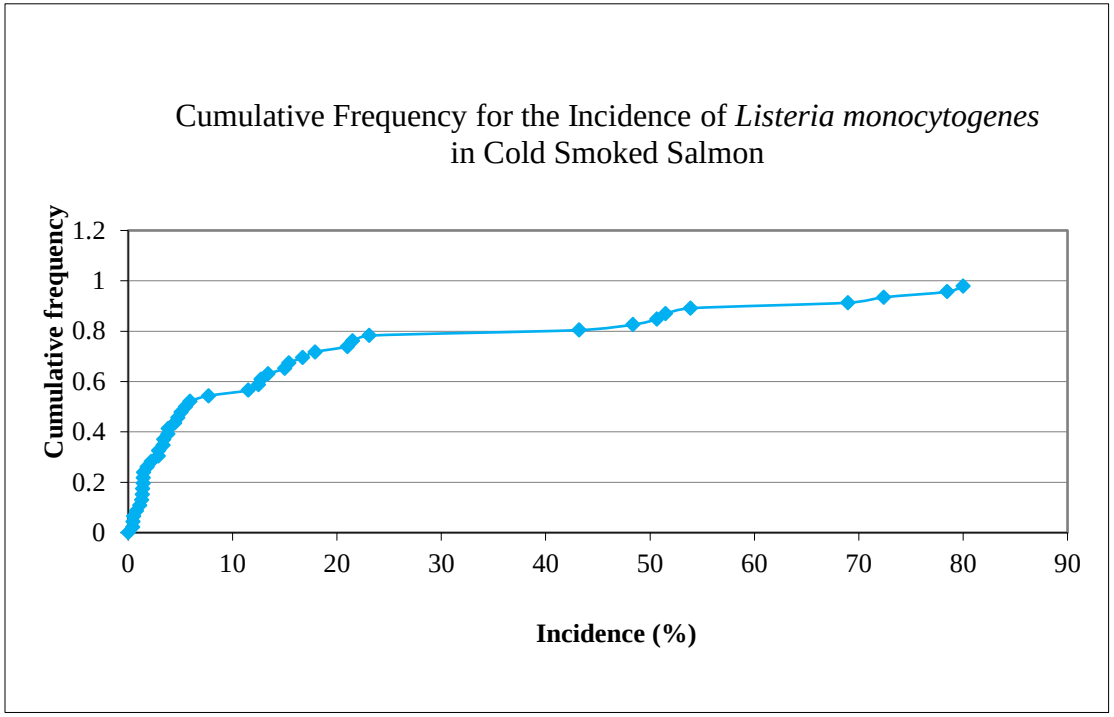
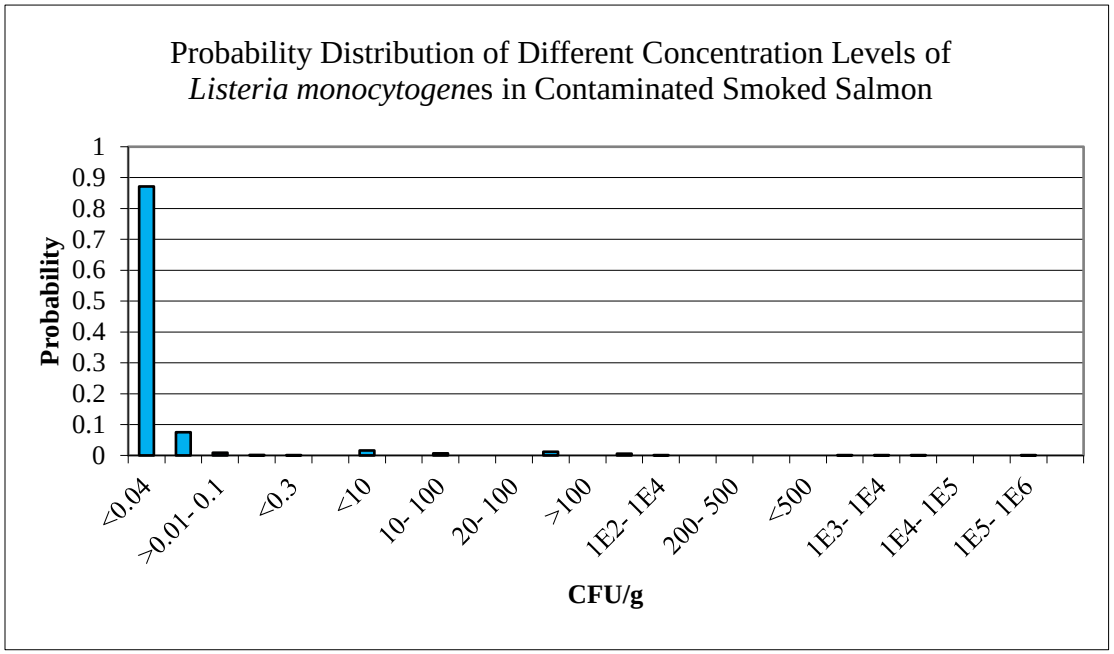


Figure A2.4 Probability Distribution of Different Concentration Levels of *Listeria monocytogenes* in Contaminated Smoked Salmon



Glossary

Risk Management Metrics	Definition (CAC 2015a, 2016)
Food Safety Objective (FSO)	The maximum frequency and/or concentration of a hazard in a food at the time of consumption that provides or contributes to the appropriate level of protection (ALOP).
Performance Criterion (PC)	The effect in frequency and/or concentration of a hazard in a food that must be achieved by the application of one or more control measures to provide or contribute to a PO or an FSO.
Performance Objective (PO)	The maximum frequency and/or concentration of a hazard in a food at a specified step in the food chain before the time of consumption that provides or contributes to an FSO or ALOP, as applicable.

Traditional Food Safety Metrics	Definition (CAC 2007a)
Process Criterion (PcC)	"A PcC specifies the conditions of treatment that a food must undergo at a specific step in its manufacture to achieve a desired level of control of a microbiological hazard. For example, a milk pasteurization requirement of a heat treatment of 72°C for 15 seconds specifies the specific time and temperature needed to reduce the levels of *Coxiella burnetii* in milk by 5 logs. Another example would be specifying the times and temperatures for refrigerated storage which are based on preventing the growth of mesophilic pathogenic bacteria such as *Salmonella* Enterica in raw meat. Underlying a PcC should be a transparent articulation of the factors that influence the effectiveness of the treatment. For the milk pasteurization example, this would include factors such as the level of the pathogens of concern in raw milk, the thermal resistance among different strains of the microorganisms, the variation in the ability of the process to deliver the desired heat treatment, and degree of hazard reduction required."
Product Criterion (PdC)	"A PdC specifies a chemical or physical characteristic of a food (e.g. pH, water activity) that, if met, contributes to food safety. Product criteria are used to articulate conditions that will limit growth of a pathogen of concern or will contribute to inactivation, thereby decreasing the potential for risk to increase during subsequent distribution, marketing and preparation. Underlying a PdC is information related to the frequency and level of the contamination in the food and/or raw ingredients that is likely to occur, the effectiveness of the control measure, the sensitivity of the pathogen to the control measure, the conditions of product use, and related parameters that ensure that a product will not have the pathogen at an unacceptable level when the product is consumed. Ideally, each of these factors that determine the effectiveness of a PdC would be transparently considered when the criterion was being established."

Microbiological Criterion (MC)	"A microbiological criterion is a risk management metric which indicates the acceptability of a food, or the performance of either a process or a food safety control system following the outcome of sampling and testing for microorganisms, their toxins/metabolites or markers associated with pathogenicity or other traits at a specified point of the food chain." (CAC 1997).
Notes: "The definition of a MC included in CAC/GL 21-1997 (CAC 1997) shall apply too." (CAC 2007a) "CAC/GL 21-1997 was revised and renamed in 2013" (CAC 1997)	"An MC is based on the examination of foods at a specific point in the food chain to determine if the frequency and/or level of a pathogen in a food exceed a pre-established limit (e.g., the microbiological limit associated with a 2-class sampling plan). Such microbiological testing can either be employed as a direct control measure (i.e., each lot of food is tested and unsatisfactory lots removed) or, in conjunction with a HACCP plan or other food safety control system, as a periodic means of verifying that a food safety control system is functioning as intended. As a technological and statistically-based tool, an MC requires articulation of the number of samples to be examined, the size of those samples, the method of analysis and its sensitivity, the number of "positives" and/or number of microorganisms that will result in the lot of food being considered unacceptable or defective (i.e., has a concentration or percentage of contaminated units exceeding the pre-determined limit), and the probability that the pre-determined limit has not been exceeded. An MC also requires articulation of the actions that are to be taken if the MC is exceeded. The effective use of an MC is dependent on a selection of a sampling plan based on the above parameters to establish the appropriate level of stringency. Since the levels of a pathogen in many foods can change over the course of their manufacture, distribution, marketing and preparation, an MC is generally established at a specific point in the food chain and that MC may not be pertinent at other points. Underlying an MC should be a transparent articulation of the pre-determined limit and the rationale for the sampling plan chosen." (CAC 2007a)

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