THE EFFECT OF KITCHEN-SCALE PRODUCE PREPARATION TECHNIQUES ON THE RISK OF LISTERIOSIS IN CANCER PATIENTS

By

Carly Gomez

A THESIS

Submitted to,
Michigan State University
in partial fulfillment of the requirements
for the degree of

Biosystems Engineering — Master of Science

2020

ABSTRACT

THE EFFECT OF KITCHEN-SCALE PRODUCE PREPARATION TECHNIQUES ON THE RISK OF LISTERIOSIS IN CANCER PATIENTS

By

Carly Gomez

The foodborne pathogen *L. monocytogenes* generally infects immunocompromised individuals, but cancer patients in particular are infected more frequently, with higher morbidity and mortality. Because of the known risk of *L. monocytogenes*, and other pathogens, in produce, immunocompromised individuals are often placed on neutropenic diets that exclude fresh produce. Therefore, this study aimed to first evaluate several kitchen-scale treatments as potential interventions to reduce the population of *L. monocytogenes* in prepared produce (apples, cucumbers, and celery), then develop a data-driven risk model for listeriosis in cancer patients who consume ready-to-eat (RTE) salads, as influenced by the kitchen-scale treatments. Surface blanching and surface blanching followed by peeling were the most effective treatments in both cucumbers (mean reductions of 5.1 and 5.9, respectively) and apples (mean reductions of 3.5 and 4.2 log cfu/g, respectively) ($P < 0.05$). All treatments lacked efficacy for celery, with reductions significantly less ($P < 0.05$) than in other products, likely due to considerable inoculum internalization. For refrigerated salads with no treatment, the median risk of invasive listeriosis over a period of one chemotherapy cycle was predicted to be at most 5.6×10^{-10}. This decreased to 7.3×10^{-11} when salad components were surface blanched. Results from this study can be used to develop improved risk management strategies and risk communication materials for cancer patients and their caretakers.

ACKNOWLEDGEMENTS

The unique, cross-disciplinary nature of this project would not be possible without the guidance and support of my tireless advisor, Dr. Bradley Marks. Throughout my many years in his lab, Dr. Marks encouraged me to pursue projects in areas that intrigue me and address pertinent societal needs. This fostered my passion for research, and I am grateful for his continued expertise. My committee members, Dr. Jade Mitchell and Dr. Elliot Ryser, have also served as significant mentors and improved my abilities in not only risk modeling and microbiology, but also critical thinking.

I would also like to thank my lab mates and fellow BAE graduate students, whose assistance and friendship I greatly appreciate. Our lab managers, Michael James and Nicole Hall, were a tremendous help throughout all of my experiments and always kept things running smoothly. Additionally, I would like to acknowledge the Farrall Hall faculty and staff who help to make Farrall Hall and the BAE program an excellent place to work and study. Finally, I am incredible grateful for the love and encouragement of my family and friends, who enthusiastically support all of my endeavors and make time outside the lab so much fun.

This material is based upon work supported by the National Science Foundation Graduate Research Fellowship Program under Grant No. DGE-1848739. Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the National Science Foundation.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER 1: INTRODUCTION

1.1 The Problem

The incidence of listeriosis has increased in recent years *(4, 38, 70, 105)*, with *Listeria monocytogenes* having been repeatedly isolated from fresh, ready-to-eat (RTE), and minimally processed produce *(13, 82, 132, 134, 137, 153, 174, 203)*. Fruits and vegetables can become contaminated with *L. monocytogenes* during preharvest from multiple environmental sources in the field *(13, 193)*. Fresh produce also can become contaminated with *Listeria monocytogenes* postharvest from contact with contaminated wash water and various surfaces, such as shredders, conveyors, and coolers *(6, 147)*. Consequently, *L. monocytogenes* has been implicated in several large outbreaks involving produce, including packaged salads *(33)*, ready-to-eat (RTE) salad *(172)*, caramel apples *(6)* and cantaloupe *(122)*. In the outbreak linked to cantaloupe, data on concurrent medical conditions were available for 123 victims, 108 of which were immunocompromised in some way *(122)*. Foodborne outbreaks also have occurred in hospitals, prompting discussion on food safety and prophylaxis for susceptible populations *(69, 85, 91, 117, 160, 163, 197)*.

Healthy individuals typically develop a noninvasive form of listeriosis, termed febrile gastroenteritis, which causes self-limiting diarrhea and fever *(4, 82, 173, 198)*. Immunocompromised members of the population, including cancer patients, pregnant women, the elderly, those with HIV/AIDS, and those with autoimmune diseases such as lupus develop invasive listeriosis, which manifests with more severe symptoms, such as meningoencephalitis and septicemia, and a high mortality rate of 20-40% *(4, 35, 38, 58, 77, 82, 168, 173, 198)*. Cancer patients are particularly vulnerable *(71, 84, 106, 161, 162)*, with higher mortality rates than other groups *(77, 78, 162)*. The relative risk of listeriosis for cancer patients can be up to

1,139 *(77)* and 17.6 *(130)* when compared to healthy individuals less than 65 years old and individuals with other immunocompromising conditions, respectively.

Cancer patients are markedly more susceptible to listeriosis because their treatment undermines several of the body's natural barriers against infection *(63)*. Chemotherapy inhibits neutrophil generation, leading to neutropenia, a state of low neutrophil count in which patients are at an increased risk of infection *(42)*. Additionally, acid-neutralizing drugs cause hypochlorhydria (stomach pH > 4.0), leaving patients susceptible to listeriosis and other bacterial infections *(11, 39, 85, 120, 178)*. Cancer treatments also invoke inflammatory responses and vascular changes that disrupt gastrointestinal tissue *(8)*. Patients may develop gastrointestinal mucositis - inflammation and ulcers throughout the digestive tract - which creates a pathway for bacteria to translocate from the digestive system to the bloodstream *(50, 188, 194)*. Finally, cancer patients are frequently treated with broad-spectrum antibiotics *(67, 99)*, the wide target range of which leads to the death of endogenous gut bacteria, eliminating natural competition as a defense mechanism *(31, 41, 63, 126)*.

Neutropenic diets (NDs) are a common risk management strategy for foodborne illness in cancer patients, despite never being proven to reduce rates of infection *(46, 83, 103, 129, 180, 189)*. NDs vary greatly between institutions *(20, 27, 68, 164)*, but most commonly eliminate the consumption of high-risk foods that are not cooked or pasteurized prior to consumption, including fresh produce and RTE salad products *(83, 166)*. This diet remains controversial because these food groups are vital sources of fiber and vitamin C, which support intestinal integrity *(76)*, reduce bacterial translocation *(30, 44, 167)* and improve immune function *(5, 26, 72, 107, 116, 124)*. Adherence to the diet is notoriously difficult *(83, 103)*, and has negative effects on quality of life *(123, 128)*. Thus, it may be more beneficial to implement food safety

strategies that focus on improved food handling practices, such as proper storage and kitchen-scale preparation. However, the efficacy of such strategies in reducing pathogen levels and illness risk needs to be assessed through experimental testing and subsequent risk analysis.

The risk to human health posed by foodborne pathogens can be assessed using quantitative microbial risk assessment (QMRA), a process comprised of the following steps: hazard identification, dose-response assessment, exposure assessment, risk characterization, and risk management *(79)*. The purpose of hazard identification is to describe the pathogen and its known effects on its host. Dose-response assessment quantifies the probability of an adverse outcome of interest, based on applied dose. For foodborne pathogens, these models depend on the amount of pathogen consumed, variations in strain virulence, and host susceptibility *(184)*. Exposure assessment determines the amount of pathogens ingested - the frequency at which consumers ingest the foods and the level of contamination on implicated foods. Survival of the pathogen is also considered, for example in various storage conditions and after any pathogen control treatments. Exposure and dose-response assessments are combined in the risk characterization step, in which risk of a designated outcome and associated uncertainty are estimated. These estimates are used to guide action decisions in risk management.

1.2 Research Gaps

Multiple risk assessments have been developed for listeriosis in generalized low-risk and high-risk populations *(12, 49, 59, 88, 111, 145, 150, 152, 181, 186, 195, 199)*, but current literature lacks a data-driven model that is specific to cancer patients. Existing dose-response models rely on studies in mice, which may not be applicable to humans *(104, 186)*, studies in guinea pigs *(195)* and primates *(165)* with an endpoint of stillbirth in pregnant animals, which is

not applicable to the target population of cancer patients, and epidemiological data *(22, 59, 111, 199),* which lack detailed exposure assessments specific to cancer patients.

Institutional discrepancies in the implementation and administration of the neutropenic diet highlight a clear misunderstanding of its purpose in mitigating foodborne illness. There is a critical need for studies that specifically address the role of the ND in preventing foodborne diseases, as existing studies only address general infections, the cause of which are much more difficult to attribute to diet.

Data on alternative risk management strategies, such as hyper-hygienic produce preparation methods, are limited. Several studies have been conducted on reduction of *L. monocytogenes* in produce; unfortunately, many of these studies were focused on large-scale commercial manufacturing practices *(2, 18, 36, 64, 121, 131, 139, 140, 142, 149, 169, 202),* which do not translate directly to preparation in healthcare or home settings. In order to inform risk models and develop accurate food safety preparation guidelines, it is important to quantify the *L. monocytogenes* reductions that may be achieved using kitchen-scale produce preparation techniques.

1.3 Goal and Objectives

Ultimately, the goal of this work is to assess the risk for listeriosis in cancer patients who consume fresh produce prepared by several differing kitchen-scale methods. This will be achieved through two main objectives: i) describe *L. monocytogenes* survival on fresh produce subjected to hyper-hygienic preparation processes; and ii) develop a risk model for listeriosis in cancer patients who consume fresh produce, in the form of RTE salads, as affected by hyper-hygienic produce preparation and modified neutropenic diets. These findings will provide a basis for evidence-based food safety measures in this vulnerable population.

CHAPTER 2: LITERATURE REVIEW

The literature review performed for this study examined foodborne pathogen reduction techniques and existing listeriosis risk models.

2.1 Pathogen Reduction on Produce

2.1.1 Chemical Treatments

The treatments that have been studied for kitchen-scale foodborne pathogen reduction can be grouped into five categories: chemical treatments, flash boiling/blanching, water soaks and rinses, organic acid soaks and rinses, and physical removal methods (Figure 1). Produce soaks and scrubs are often used commercially and have been shown to result in moderate pathogen reductions; however, these surfactants, detergents, and solutions, such as sodium hypochlorite, chlorine, and hydrogen peroxide are not recommended by the FDA for consumer use *(2, 18, 64, 142, 169, 185, 202)*. Recently, fruit and vegetable washes that target microbes, dirt, wax, and pesticides have become available to consumers. Existing studies on such products report contradictory efficacy results *(16, 62, 81, 98, 113, 115, 139, 176)*, indicating a need for further validation of consumer produce washes as antimicrobial treatments.

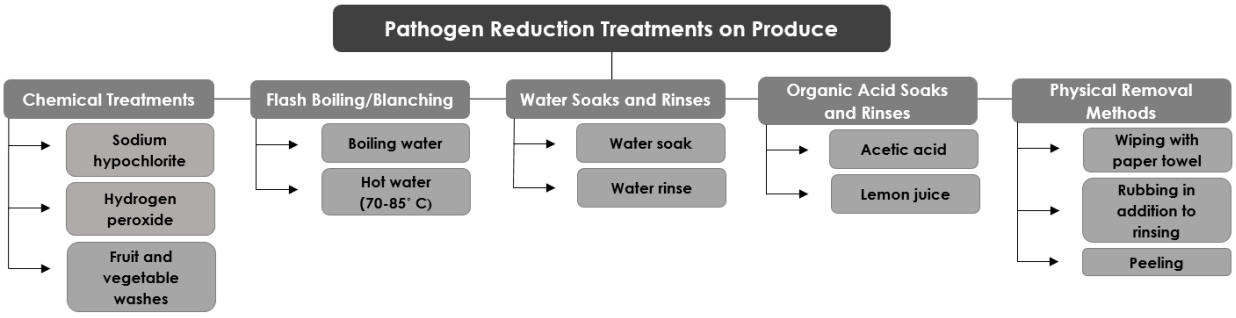

Figure 1: Kitchen-scale pathogen reduction treatments

The antimicrobial effects of organic acid soaks (propionic, acetic, lactic, malic, and citric acids) have also been tested in produce, with less to similar efficacy as water soaks and rinses *(131, 140, 169)*. Unfortunately, the most significant reductions occurred at acid concentrations

greater than 1.0%, at which point smell, taste, and texture may be compromised *(131)*. Additionally, these approaches may not be ideal for kitchen-scale treatment, as consumers may not have the proper training or equipment to handle high concentrations of chemicals.

2.1.2 Flash Boiling/Blanching

Blanching (flash boiling), which is frequently used in commercial food processing to inactivate enzymes and expel gasses, can also have antimicrobial benefits *(121)*. Blanching of produce can reduce foodborne pathogen populations by as much as 8 log CFU/g *(21, 36, 114, 121)*. However, conflicting results concerning product integrity have been reported following treatment (when trying to preserve a functionally raw product); thus, further work is needed to validate surface blanching treatments aimed specifically at reducing bacterial surface contamination (rather than enzyme inactivation).

2.1.3 Water Soaks and Rinses

Most consumers and food service personnel report using water treatments to clean and sanitize their produce *(108, 113, 142)*, as water washing/rinsing is the method recommended by the U.S. Food and Drug Administration (FDA) *(183, 185)*. Additionally, while water soaking treatments may moderately reduce pathogen levels on contaminated product, previously uncontaminated products can readily became contaminated from the same wash water *(90, 141)*. Rinsing treatments have shown similar efficacy in removing *L. monocytogenes* from produce *(62, 98)*, and do not involve handling potentially contaminated wash water.

2.1.4 Physical Processes

Various physical processes also have been investigated as potential microbial reduction strategies for contaminated produce *(53)*. In general, the frictional forces of rubbing or scrubbing in addition to rinsing greatly improve pathogen reduction, but again efficacy depends largely on

product surface characteristics *(53, 98, 115, 141)*. Peeling the surface of firm fruits and vegetables has also been investigated as a means of pathogen reduction and was found to have moderate efficacy, although allowing for considerable survivors *(53).* Although not tested on products inoculated with *L. monocytogenes*, these results suggest that friction-based treatments can also decrease pathogen populations

2.1.5 Factors Affecting Pathogen Removal

Storage temperature affects the efficacy of pathogen-reduction treatments on produce. Nastou et al. *(131)* reported that water soaking resulted in significant pathogen reductions when lettuce and parsley were stored at 5°C and 15°C, but not at 30°C. Francis and O'Beirne *(64)* reported that the efficacy of antimicrobial treatments decreased faster when produce was stored at 8°C opposed to 3°C. These results demonstrate that proper storage temperatures are crucial to support kitchen-scale treatments.

Storage time also influences the effectiveness of pathogen removal treatments. Sapers *(155)* and Lopez et al. *(113)* explained that increased time between a contamination event and antimicrobial treatment increased the resistance of pathogens to remove from produce. Francis and O'Beirne *(64)* also reported that *L. innocua* regrew in as few as four days after treatment. Longer storage times can make it more difficult to reduce *L. monocytogenes* during treatment and can reduce the positive effects of the treatment.

The morphology of fruits and vegetables is highly variable, with the calyx, stem, and other structures more conducive to attachment and growth of *L. monocytogenes*. Kilonzo-Nthenge *(98)* demonstrated that in broccoli, higher inoculation was achieved for the calyx than the stem, and, in apples, water treatments were not as effective in *L. innocua* removal from the calyx and stem ends compared to the remaining apple surface. Sapers et al. *(156)* also reported

that *E. coli* cells remaining on apples after treatment were clustered in the stem and calyx ends. Removing these portions of the fruit during preparation may reduce the consumable dose.

Some produce types are anti-listerial by nature, whether it be due to the composition of their tissues or the antagonistic nature of their native microflora *(51)*. In 2007, Liao *(109)* demonstrated that carrots were anti-listerial for both of these reasons. When sliced carrots were dipped into a *L. monocytogenes* suspension, a decrease of greater than 2 log cfu was observed in the suspension. Additionally, when the carrots were sanitized to curtail native microflora, *L. monocytogenes* growth increased by greater than 2 log cfu, and when carrot native microflora was introduced onto bell peppers (a product with no inherent antilisterial activity), *L. monocytogenes* growth on the peppers was inhibited. Erickson *(51)* hypothesized that similar characteristics may exist in other types of produce, as certain varieties of cabbage have been shown to facilitate *L. monocytogenes* growth during storage, and others have caused a decline in *L. monocytogenes* population. For these reasons, *L. monocytogenes* contamination may vary greatly between products, and inoculation levels should be verified prior to treatment testing. Additionally, testing on a wide array of produce types would improve understanding of this characteristic.

2.2 Existing Listeriosis Risk Studies

Risk estimates depend heavily on dose-response models, which relate amount of pathogen consumed to the occurrence and severity of detrimental health outcomes *(92)*. Because risk resulting from a single exposure is often so low (e.g., 1 in 10,000) that experiments would require an impractical number of subjects (> 10,000), a dose-response model is crucial for estimating risk at low doses *(79)*. Response data can be collected from either animal studies or epidemiological data and used to derive a model, from which responses at low doses can be

extrapolated. In animal studies, the effects of various high pathogen doses are observed in a surrogate animal having similar pathology to humans. It can be difficult to find such an animal, as animal exposure routes, morbidity, and mortality rates often differ from humans. Epidemiological data (cases, illnesses, and deaths) are combined with consumption and contamination data to attribute recorded cases to an estimate of pathogen dose consumed. These models often include a lot of uncertainty, as estimates are dependent on the quality of case, consumption, and contamination data reported. Existing dose-response models for listeriosis are based on either approach; both, as well as an alternative approach, will be explored in this review. Exposure assessments are another crucial element of risk analysis, as they determine the dose of pathogen subsequently used in the dose-response model. Unfortunately, current risk studies do not consider the consumption of salad products by cancer patients specifically; however, relevant alternatives will be examined.

2.2.1 Animal Study Models

In 1989, Golnazarian et al. *(73)* developed dose-response models for infection and death in healthy and immunocompromised mice that ingested *L. monocytogenes* suspended in food. These data were used in the 2003 U.S. Food and Drug Administration (FDA) and Food Safety and Inspection Service (FSIS) risk assessment *(186)* to establish the dose-response curve. Because *L. monocytogenes* colonization of mouse organs may not translate directly to invasive listeriosis in humans, death was chosen as the endpoint for this model. The variation in virulence was determined by performing feeding studies in mice with multiple strains of *L. monocytogenes* isolated from food. However, when the resulting mouse model was implemented with the exhaustive exposure assessment data, the calculated LD_{50} (dose that is lethal for 50% of the population) was overestimated by a factor more than 1,000,000 *(186)*. Consequently, a dose-

response scaling factor derived from FoodNet surveillance data was applied to the mouse curve, so that model predictions agreed with public health data.

Lecuit et al. *(104)* suggested that mice may not be an adequate surrogate for human listeriosis studies. They note that in humans, E-cadherin (an epithelial surface receptor protein) is a receptor for internalin (an *L. monocytogenes* surface protein), which allows *L. monocytogenes* to translocate through the intestinal epithelium and cause systemic infection. Mouse E-cadherin is not a receptor for internalin, thus preventing translocation of the bacterium through the intestinal epithelium. Because mice cannot model the suspected route of systematic infection in humans, caution is needed when using the data from mice to predict human illness *(199)*.

Since this discovery, other animals have been explored as models for dose-response studies. In 2007, Williams et al. *(196)* determined an effective dose for stillbirths and maternal organ infection in pregnant guinea pigs that consumed whipping cream inoculated with *L. monocytogenes*. Later, Smith et al. *(165)* created a dose-response model for stillbirths in orally exposed pregnant rhesus monkeys. These models predicted LD_{50} values that were much lower than those predicted by the FDA/FSIS model, and are similar to those predicted by the Food and Agriculture Organization of the United Nations (FAO) and World Health Organization (WHO) model (reviewed in the following section). Unfortunately, stillbirth is not the endpoint of interest in this study, so these models are not applicable to cancer patients.

2.2.2 Epidemiological Data Models

In 1996, Farber et al. *(59)* assessed the risk of listeriosis from pâté and soft cheese in Canada using a "Weibull-Gamma" dose-response model, which was chosen for its flexibility and capacity to be transformed to other well-known models. Parameters were characterized based on chosen estimates for ID_{10} and ID_{90} (doses that will result in illness for 10 and 90% of the

population), that resembled approximate doses that caused illness in outbreaks. The prevalence of *L. monocytogenes* reported in pâté and soft cheese, combined with disappearance data (food that "disappears" from the supply chain), were used to assess exposure. This model relied on many generalizations, particularly for exposure, which greatly influences overall risk characterization. Therefore, a risk model with a more credible exposure assessment and dose-response parameterization approach would likely be more accurate.

Using food survey data that quantified *L. monocytogenes* in smoked fish along with detailed data on national listeriosis cases, Buchanan et al. *(22)* developed a conservative dose-response model for listeriosis. This model relied on the following assumptions: i) all cases of listeriosis were in immunologically high-risk populations and due to consumption of RTE smoked fish; ii) an exponential dose-response model can be used for listeriosis; and iii) only servings with *L. monocytogenes* populations greater than 4 logs caused illness. These assumptions prevented the model from accounting for differences in prevalence and growth potential between foods and in susceptibility due to various immunocompromising conditions. Lindqvist and Westöö *(111)* used an approach with similar assumptions, but with a Weibull-gamma model for high- and low-risk groups, with reported results much more conservative than Buchanan's. The sizable difference in estimated risk between the two approaches revealed inadequate dose-response data. In both cases, improved information on prevalence, level of contamination, and strain virulence would enhance risk model validity, as would specificity to fresh produce and cancer patients for the target application in this study.

In 2004, FAO/WHO developed an exponential dose-response model with an endpoint of invasive listeriosis *(199)* using the exposure data from the 2003 FDA/FSIS risk assessment *(186)*. This dose response model utilized two values of parameter *k* (the probability that one

organism will survive to cause illness) to account for variations in host susceptibility, corresponding to general high- and low-risk populations. Being based on thorough epidemiological data, the dose-response model in this risk assessment is considered highly reliable and has been used in several subsequent risk assessments *(88, 145, 150, 152)*. However, this risk assessment did not calculate risk due to fresh produce, but instead targeted pasteurized milk, ice cream, fermented meat, and smoked fish.

More recently, Pouillot et al. *(146)* developed specialized exponential listeriosis dose-response models by adjusting the *k* parameter for host susceptibility and strain virulence. Using strain characterization from the FDA/FSIS risk assessment *(186)*, as well as epidemiological data, they created dose-response parameters for 11 population subgroups, including hematological and nonhematological cancer patients. The characterizations of this dose-response parameters are valuable for the current work.

2.2.3 Alternative Model

The most recent listeriosis dose-response model was developed by Rahman et al. *(148)*. This model utilizes a novel mechanistic approach to more accurately assess risk based on host-pathogen dynamics. Several different parameters account for host-specific physiological characteristics, such as stomach acid, commensal bacteria in the small intestine, and host immune cells. Values for these parameters were derived from a butter outbreak in Finland that primarily affected immunocompromised patients (blood and organ transplant patients). This is a promising start for a model applicable for cancer patients, but because the group's sensitivity analysis proved that risk of infection is most dependent on the host's immune response, additional parameters are needed to address specific cancer physiologies (hypochlorhydria,

impaired neutrophil generation). Unfortunately, this is not currently possible due to the lack of clinical data.

2.3 Summary

This literature review identified several important gaps regarding this project. First, most commercial pathogen reduction strategies for produce are not applicable to home or healthcare settings, due to the use of pernicious chemicals. Studies on applicable treatments, such as commercial produce washes and water soaks and rinses, have yielded conflicting results. Other approaches, such as blanching and peeling have potential for effective removal but have not been tested on products inoculated with *L. monocytogenes*. Therefore, the efficacy of these treatments needs to be experimentally assessed on a variety of produce.

Risk models are contingent on their dose-response model and exposure assessment, but many existing dose-response models for listeriosis cannot be applied to cancer patients due to their use of controversial animal surrogates (mice), inappropriate endpoints (stillbirth), or susceptibility generalizations. Additionally, some risk models rely on exposure assessments based on consumption of other foods (soft cheese, smoked fish), which differs from fresh produce. One study characterized dose-response parameters for cancer patients but did not conduct a full risk assessment. Thus, there is a need for a listeriosis risk model specific to raw produce and cancer patients. This thesis addresses the aforementioned literature gaps through systematic testing of kitchen-scale microbial reduction strategies and the development of a listeriosis risk model specific to cancer patients who consume fresh produce.

CHAPTER 3: KITCHEN-SCALE TREATMENTS FOR REDUCTION OF *L. MONOCYTOGENES* IN PREPARED PRODUCE FOR IMMUNOCOMPROMISED POPULATIONS

Immunocompromised individuals are typically placed on diets that exclude raw produce due to the expected risk of foodborne pathogens, one of the most dangerous being *L. monocytogenes*. Pathogen reduction and sanitization treatments may be effective in raw produce, but related studies are few, with conflicting results. The goal of this study was to assess the efficacy of several kitchen-scale treatments, suitable for use by hospital staff, caretakers, and patients themselves, for the reduction of *L. monocytogenes* on fresh produce, using cucumbers, apples, and celery as representative products. This study directly addresses the research gap for *L. monocytogenes* reduction on produce in healthcare and home settings which is needed to develop the risk assessment in Objective 2. The results of this study will help in the development of data-driven food safety guidelines for immunocompromised individuals and later be used to inform risk models.

3.1 Materials and Methods

3.1.1 Produce

Three different products tested were chosen based on surface morphology: (i) miniature cucumbers (rough surface), ii) apples (smooth surface with stem and calyx ends), and iii) celery (porous, rigid surface). Fresh miniature cucumbers and apples (cv. "Gala") were purchased from a local supermarket, stored in a walk-in cooler at 4°C no longer than one week before treatment. Pre-cut, ready-to-eat (RTE) celery sticks were purchased packaged from a local supermarket and stored in a walk-in cooler at 4°C, with all treatments completed within 10 days of purchase. All products were visually inspected, and any samples showing surface/structural damage or decay were discarded.

3.1.2 Utensil and Supply Preparation

All metal utensils, including cooling racks, knives, stirring spoons, tongs, spatulas, apple slicers/corers, and manual peelers, were autoclaved at 121°C for 15 min before use. Plastic cutting boards and containers with non-metal components, such as the apple peeler, were thoroughly cleaned, disinfected in 75% ethanol, and dried for 5 min before use. The same protocol was used if utensils had to be reused within the same replication.

3.1.3 Bacterial Strains

Three avirulent *Listeria monocytogenes* strains (J22F serotype 4b *(56)*, J29H serotype 4b *(56)*, and M3 serotype 1/2a *(96)*) were acquired from Dr. Sophia Kathariou from the Department of Food, Bioprocessing, and Nutrition Sciences at North Carolina State University. The stock cultures were stored at -80°C in tryptic soy broth (TSB; Difco, BD, Sparks, MD) containing 10% (v/v) glycerol. Working cultures were streaked onto tryptic soy agar containing 0.6% yeast extract (TSAYE; Difco, BD), incubated at 37°C for 24 h, and transferred monthly.

3.1.4 Inoculum Preparation

For each strain, an isolated colony from the working stock was subjected to two 37°C/ 24 h transfers, in 9 and 1000 mL of tryptic soy broth containing 0.6% yeast extract (TSBYE; Difco, BD), respectively. Subsequently, the three strains were combined in a sterile container to yield 3000 mL of inoculum. The *L. monocytogenes* population in the inoculum was ~10^9 log cfu/ml, as determined by plating samples diluted in sterile 0.1% phosphate-buffered saline (PBS) (MP Biomedicals, Irvine, CA) on TSAYE containing 0.025% (w/v) esculin (97% esculin hydrate; Sigma-Aldrich, St. Louis, MO) and 0.05% (w/v) ammonium iron citrate (Sigma-Aldrich). This medium (eTSA) is a non-selective, differential medium that produces gray-green, black-haloed

Listeria colonies with indented black centers. Because preliminary work revealed low bacterial attachment to apples, the wax was removed before inoculation by very briefly submerging each apple in boiling water for 5 s and wiping with a paper towel.

3.1.5 Dip Inoculation

Whole products were removed from storage, equilibrated to room temperature, and submerged in the three-stain *L. monocytogenes* cocktail for 10 min, while being agitated with a large sterile spoon. After inoculation, the samples were placed on a sterile stainless-steel rack in a large plastic tub, partially covered, and stored in a walk-in cooler at 4°C for 24 h.

3.1.6 Treatments

Treatments included were: (i) submerging in a commercial produce sanitizer (1 oz sanitizer, 3,800 mL tap water) with active ingredients dodecylbenzene sulfonic acid sodium salt (1.23%) and lactic acid (17.29%) (Monogram Clean Force, Ecolab, St. Paul, MN) for 90 s ("sanitizer soak"), (ii) rinsing under running tap water for 15 s under running tap water with hand scrubbing, ("tap water rinse"), (iii) soaking in tap water for 90 s with frequent agitation ("tap water soak"), (iv) surface blanching in boiling water for 25 s ("blanch"), (v) rinsing under running tap water for 15 s followed by peeling ("rinse+peel"), and (vi) surface blanching in boiling water for 25 s followed by peeling ("blanch+peel"). The tap water temperature was 24.5 ± 0.2°C, as verified by a thermometer (Omega, Norwalk, CT). Blanching was performed in water with a temperature of 100 ± 0.2°C. After blanching, the whole products were placed in appropriately sized Whirl-Pak bags (Nasco, Fort Atkinson, WI) and immersed in an ice bath for 1 min to lower the surface temperature. Standard, handheld vegetable peelers were used for cucumbers and apples, and crank apple peelers (CucinaPro Apple Peeler, CucinaPro, Trumbull, CT) also were used for apples. Two pieces of produce were examined for numbers of *Listeria* in

each of three replicated trials per treatment as described below. Sample assay methods are also described below.

3.1.7 Additional Experiments

Additional experiments were performed for celery and apples to evaluate product-specific attributes of inoculation and recovery. Inoculum uptake experiments were performed on pre-cut celery sticks to elucidate surface vs. internal bacterial inactivation. Prior to inoculation, celery sticks were labelled with ink marker 1 mm from the root end at nine 1-cm increments. The inoculum was pipetted into a sterile glass beaker to a height of 1 mm. Thereafter, the beaker was covered with aluminum foil containing holes through which the celery sticks were inserted to the bottom of the beaker. After 10 min in the 1 ml of inoculum, the celery sticks were removed and placed on a sterile stainless-steel rack in a walk-in cooler (4°C for 24 h) in the same manner as the other products. Three replications were performed, with two subsamples in each replication.

For apples, the efficacy of peeling with a crank apple peeler designed to remove an apple peel in a continuous rotational peeling action was compared with a standard, handheld vegetable peeler. Apples were inoculated as previously described, rinsed for 15 s with hand scrubbing under running tap water, and then peeled using either the apple peeler or a standard handheld vegetable peeler. Again, three replications were performed, with two subsamples in each replication.

3.1.8 Peeling

Miniature cucumbers were peeled vertically from top to bottom with the standard handheld vegetable peeler until no skin remained. The ends were not peeled and were removed during sampling. Apples were also manually peeled from top to bottom. leaving the skin at the calyx and stem ends (about 10% of the total) remaining. The peelers were sterilized between

products. Apples peeled by the apple peeler were secured to the core prongs and were peeled until about 5% of the total apple skin remained at the calyx and stem ends.

3.1.9 Post-Treatment Sample Preparation, Recovery, and Enumeration

The treated samples were placed on a sanitized cutting board in a biological safety cabinet. Apples were cored and sliced into 12 equal size wedges using a sterile apple slicer/corer (Vremi, New York, NY), with every fourth piece (three slices total, the first chosen at random) placed into a Whirl-Pak sampling bag (Nasco). Cucumbers were cut into 1-cm thick slices (measured with a sterile ruler) using a sterile knife. End pieces were discarded. and the next closest pieces, as well as the center piece (three slices total), were placed in a sampling bag. The same procedure was repeated for celery using segments 3-cm in length. In the inoculum uptake experiment for celery, the stalk was cut into pre-labelled 1-cm long segments with a new sterile knife used for each cut to eliminate inoculum transfer. All samples were diluted 1:5 (w/v) in sterile 0.1% PBS and stomached for 180 s (IUL Masticator Silver, 400 ml, IUL S.A., Barcelona, Spain). Thereafter, 1 mL of the homogenized sample was serially diluted in 9 mL sterile 0.1% PBS with appropriate dilutions plated on eTSA (1 mL for suspected low populations, 0.1 otherwise). After 48 h of incubation at 37°C. all colonies resembling *L. monocytogenes* were counted and compared to the untreated control (log cfu/g). The limit of detection was 2.5 cfu/g. These methods were also applied to untreated products (two per replication, totaling six of each) to determine product-specific mesophilic bacteria populations.

3.1.10 Statistical Analyses

All experiments were performed in triplicate with duplicate subsamples for each treatment. Statistical tests included analysis of variance (generalized linear model ANOVA) followed by a Tukey pairwise comparison, and two-sample t-tests for comparison between

individual treatments (all completed using Minitab 19, State College, PA), with a significance level of $\alpha = 0.05$ applied for all tests. In treatments that resulted in undetectable levels of *L. monocytogenes*, the limit of detection was used as a conservative substitute data point, and t-tests were not performed for these treatments.

3.2 Results and Discussion

3.2.1 Inoculation and Microbial Background Populations

Inoculation resulted in mean *L. monocytogenes* populations of 5.3, 6.6, and 7.1 log cfu/g for apples, cucumbers, and celery, respectively (**Error! Reference source not found.**). No suspect *Listeria* colonies were isolated from uninoculated products. Uninoculated cucumbers and celery yielded mesophilic bacteria populations of 6.3 and 3.1 log cfu/g, respectively. For apples sampled following wax removal, background microflora was less than the limit of detection in five of six samples; one apple yielded 2.0 log cfu/g APC. Due to the high inoculum levels, background microbes were assumed to not impact *L. monocytogenes* enumeration in the samples after treatment.

Table 1: Initial populations of *L. monocytogenes* on inoculated products

Product	Population (95% CI) (log cfu/g)
Apple	5.3 (4.7, 5.8)
Cucumber	6.6 (6.4, 6.8)
Celery	7.1 (6.9, 7.4)

At temperatures greater than 70°C, imperfections in the apple's epicuticular wax layer are sealed by the melting of the wax, which may prevent pathogen uptake *(93)*. However, because apples were visually inspected for external damage prior to inoculation, and cores (the main

pathogen entry point) were not sampled, it was assumed that this did not alter the overall inoculation and treatment results.

3.2.2 Apples

Populations of *L. monocytogenes* were significantly lower ($P < 0.05$) in all treated apples than the positive controls, as determined by individual t-tests. ANOVA followed by the Tukey test indicated that blanch yielded significantly greater ($P < 0.05$) reductions (mean 4.2 log cfu/g) than sanitizer soak, tap water rinse, tap water soak, and rinse+peel (Table 2). Sanitizer soak, tap water rinse, and tap water soak did not differ significantly in efficacy.

Table 2: Reduction of *L. monocytogenes* on apples, cucumbers, and celery following different kitchen-scale treatments

Product*	Treatment	*L. monocytogenes* reduction (log cfu/g) Mean (95% CI)**
Apple a	Sanitizer soak	1.2 (0.8, 1.6) C
	Tap water rinse	1.3 (0.3, 2.2) C
	Tap water soak	1.0 (0.5, 1.5) C
	Blanch	4.2 (3.6, 4.8) A
	Rinse+peel	2.4 (1.3, 4.8) BC
	Blanch+peel	3.5 (2.1, 5.0) AB
Cucumber a	Sanitizer soak	1.5 (1.2, 1.8) BC
	Tap water rinse	1.4 (1.2, 1.5) BC
	Tap water soak	0.6 (0.4, 0.8) C
	Blanch	5.1 (3.7, 6.4) A
	Rinse+peel	2.4 (1.4, 3.4) B
	Blanch+peel	5.9 (5.4, 6.4) A
Celery b	Santizer soak	0.6 (-0.2, 1.4) A
	Tap water rinse	0.5 (-0.2, 1.3) A
	Tap water soak	0.6 (0.2, 1.0) A
	Blanch	1.2 (0.3, 2.1) A

***** Within this column, products followed by a common lower-case letter did not have significantly different ($\alpha = 0.05$) mean reductions for all treatments; peeling treatments were excluded for unbiased comparison across products. Mean reductions were 2.8, 2.3, and 1.2 log cfu/g for apples, cucumbers, and celery, respectively.

** Within this column, mean reductions followed by the same letter were not significantly different ($\alpha = 0.05$) within the same product category.

Due to the structural differences between apples, the specialized apple slicer and corer did not always precisely core the apple. In preliminary work, apple cores tested separately from

apple slices had a significantly lower mean reduction (0.9 and 1.6 log cfu/g for cores and slices, respectively) of *L. monocytogenes* across all treatments. Other studies reported similar results *(23, 24, 98, 156)*. Because this portion of the apple is inedible, and therefore would not contribute to the risk of acquiring listeriosis from eating apples slices separated from the core of an intact apple, it was not included in further analysis. Nevertheless, the inability of the apple corer to precisely remove the entire core from sampled slices likely contributed to the variable results, and therefore to the resulting risk in actual food preparation and consumption scenarios.

It was hypothesized that, compared to standard handheld vegetable peeler, the apple peeler would be more effective at reducing *L. monocytogenes* populations, as the cutting tool is not continuously reintroduced into the contaminated surface. Both the standard handheld and apple peelers reduced *L. monocytogenes* significantly ($P < 0.05$) from the positive control (Table 3). A two-sample t-test indicated that there was no significant difference in the efficacies of the two tested peeling methods ($P = 0.83$), suggesting little food safety benefit from using one peeler over the other.

Table 3: Reduction of *L. monocytogenes* on apples following peeling with standard handheld or crank apple peelers

Peeler	*L. monocytogenes* Reduction (log cfu/g) Mean (95% CI)
Standard handheld	1.6 (0.7, 2.6)
Apple	1.8 (1.1, 2.5)

3.2.3 Cucumbers

Similar to apples, all cucumber treatments significantly lowered ($P < 0.05$) *L. monocytogenes* populations compared to the untreated controls, with sanitizer soak, tap water rinse, and tap water soak exhibiting similar efficacy. As expected, blanch+peel was the most

effective treatment for cucumbers (mean reduction 5.9 log cfu/g) but was not significantly different from blanching alone (mean reduction 5.1 log cfu/g) (Table 2).

3.2.4 Celery

Tap water soak and blanch significantly decreased ($P < 0.05$) *L. monocytogenes* compared to the positive control; however, tap water rinse and sanitizer soak yielded *P*-values of 0.23 and 0.13, respectively, meaning that these treatments resulted in no significant reduction in population. None of the treatments were significantly different from each other, but the maximum reduction was achieved with surface blanching (mean reduction 1.2 log cfu/g). This limited efficacy is likely due to inoculum internalization during dip inoculation, with *Listeria* migrating through the porous structure and end cuts of the celery during processing *(52, 95, 158, 177).*

Results of the celery inoculum uptake experiment confirmed that *L. monocytogenes* can migrate in celery (Figure 2). The mean *L. monocytogenes* population in the inoculated 1 mm end

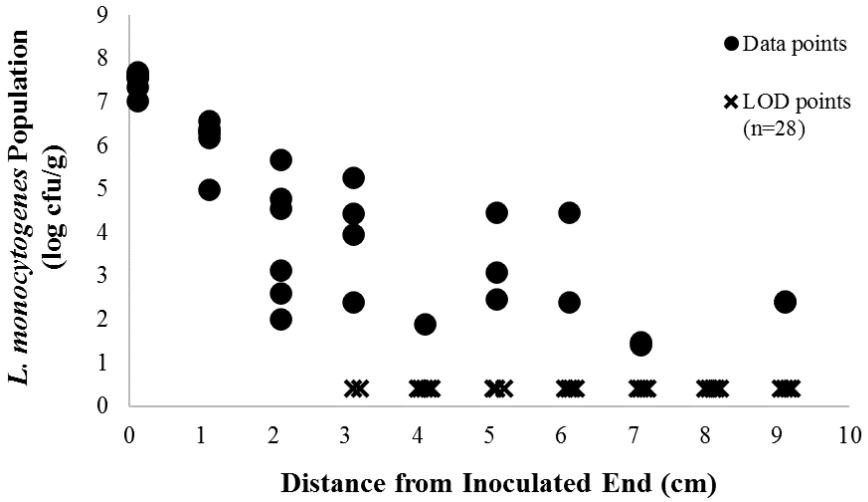

Figure 2: *L. monocytogenes* population and distance from inoculated celery end in inoculum uptake experiment.

piece was 7.5 log cfu/g. Although the numbers of *L. monocytogenes* decreased with distance

from the point of inoculation (Figure 2), *L. monocytogenes* was still detected as far as 9 cm from the cut end. These results support the internalization of *Listeria* into celery during submersion, with these cells remaining viable after the surface treatments *(175, 191)*.

3.2.5 Overall Treatment Comparisons

Overall, the surface decontamination treatments were most effective for cucumbers, and not significantly different for apples (Table 2). Efficacy for celery was minimal and significantly less ($P < 0.05$) than for both cucumbers and apples. As results from the inoculum uptake experiment indicated, this is assumed to be due to internalization of *Listeria* in celery.

Reductions due to sanitizer soak were relatively low, 0.6-1.5 log cfu/g, and significantly different from the control for apples and cucumbers, but not celery. Similarly, several past studies of various commercial produce sanitizers reported reductions of 0.5-2.3 log cfu/g, which were either not significantly different from or similar to reductions due to water treatments *(62, 98, 113, 115, 139, 176)*. However, Beuchat et al. *(16)* and Harris et al. *(81)* reported much higher reductions (> 4.8 log cfu/g). According to Lopez et al. *(113)* these differences could be due to the manner in which these treatments are applied in laboratory, commercial, and home settings. Given their low efficacy, these treatments may be insufficient as a risk reduction strategy for preparing fresh produce to be consumed by immunocompromised individuals.

Across all products, tap water rinse and soak yielded small reductions (0.5-1.4 log cfu/g), with no significant difference in efficacy between the two treatments. These results correspond with previous studies on leafy greens, carrots, broccoli, apples, tomatoes, and parsley *(53, 62, 90, 98, 131, 142)*, which reported reductions of 0.1-3.0 log cfu/g for scrubbings and soakings in sterile water or tap water. These results were slightly lower than those of Parnell et al. *(141)*, who

reported *Salmonella* reductions of > 4.6 log cfu/melon after scrubbing honeydew melons in water for 60 s, which was significantly more effective than soaking. However, their greater reduction can be attributed to the longer wash time, and variation in treatment efficacy due to the product's unique surface characteristics *(90, 98, 131, 141, 142)*. While recommended by the FDA *(183, 185)*, rinsing produce under running tap water is minimally effective and should not be used as the sole decontamination step when preparing fresh-cut produce for immunocompromised individuals.

Rinse+peel yielded *L. monocytogenes* reductions of ~2.4 log cfu/g for apples and cucumbers, with surviving populations ranging from 2.8 to 4.2 log cfu/g. Correspondingly, Erickson et al. *(53)* and Wade et al. *(190)* reported moderate reductions after peeling cucumbers and carrots (2.2 to 3.2 log cfu/g), with substantial numbers of survivors (2.6 to 2.7 log cfu/g). In an ANOVA comparing all apple and cucumber treatments, rinse+peel was significantly more effective ($P < 0.05$) than tap water rinse, tap water soak, and sanitizer soak, but significantly less effective ($P < 0.05$) than blanch and blanch+peel, indicating that the latter treatments may be most effective.

Surface blanching followed by peeling has not previously been reported as a pathogen reduction treatment for kitchen-scale fresh produce preparation. The results for apples and cucumbers indicated that blanch+peel may or may not reduce pathogens more effectively than blanching alone. However, in considering candidate treatments for preparing fresh produce for immunocompromised individuals, a conservative approach of combining blanching and peeling may be recommended. Results for apple and cucumber blanching alone were comparable to those reported by Ceylan et al. *(36)*, Bacgi and Temiz *(9)*, and Losikoff *(114)*, in which blanching vegetables in hot or boiling water for 30 s reduced *L. monocytogenes* populations by

3.3-5 log cfu/g. In contrast, Mazzotta *(121)* reported instantaneous 5-log cfu reductions when vegetables were blanched at temperatures greater than 82°C, and Monu et al. *(127)* modeled 6 log cfu/g reductions for spinach blanched at 90°C for 0.00002 s. However, they used flattened bags of 3-10 g homogenized vegetable samples that were presumably more susceptible to heat treatment than whole products, which could explain the perceived increased efficacy. Blanching was significantly less effective ($P < 0.05$) for celery than for apples and cucumbers, which again is likely due to internalization of the inoculum.

After blanching, the apple skin was slightly darker. However, all products appeared to maintain their texture, with blanching not affecting the ease of peeling apples or cucumbers. Briedt *(21)* and Fan *(57)* also reported that a 15 s blanch in boiling water did not affect the physical properties of cucumbers and cantaloupes, respectively. Mazzotta *(121)* reported similar results for broccoli; however, onions and peppers were more easily compromised. Vitamin C retention following blanching, which varies by product type, was not affected in cantaloupes *(57)*, but decreased 28% in peas *(157)*. Future studies assessing the impact of blanching on produce texture and nutrient depletion are needed to make informed decisions regarding the use of produce blanching for cancer patients on neutropenic diets.

Dip inoculation may not be representative of all contamination events, just the scenario in which the product becomes contaminated during washing. The same level of inoculum uptake may not occur in situations where the surface of the product becomes sporadically contaminated with pathogens. Due to the vulnerability of the target population to *L. monocytogenes*, using the present conservative approach is most prudent in developing data to be used in future risk models and updated recommendations for produce preparation.

3.2.6 Conclusions

Limited success was achieved with the proposed microbial reduction strategies for celery, given that the inoculum migrated through the celery from the cut end. For this reason, it is recommended that immunocompromised individuals not consume porous or pre-cut RTE produce, which has significant potential for pathogen internalization. Because sanitizer soak, tap water rinse, and tap water soak did not differ in efficacy, and were minimally effective in decreasing *L. monocytogenes,* these treatments are not recommended. For apples and cucumbers, blanch and blanch+peel were the most effective treatments, with mean *L. monocytogenes* reductions > 3.5 log cfu/g. Although these two treatments were not significantly different from one another, we recommend to conservatively blanch and peel products that are to be consumed by immunocompromised individuals. Further studies assessing product integrity, nutrient retention, and consumer appeal of such treated products will also help to inform risk models and improve food safety guidelines for kitchen/home-scale preparation of fresh fruits and vegetables for immunocompromised individuals.

CHAPTER 4: DEVELOPING A RISK MODEL FOR LISTERIOSIS IN CANCER PATIENTS WHO CONSUME READY-TO-EAT SALAD

While many listeriosis risk assessments have been performed for RTE foods, including salads, none are specific to cancer patients who have an increased risk of acquiring the disease. Additionally, the current typical risk management strategy for this population is exclusion of all raw produce, which has unknown efficacy and nutrient disadvantages to the patient. This study aimed to examine the risk posed to this vulnerable population from consuming produce and the effects of several kitchen-scale risk management strategies on that risk, thereby completing the second research objective of this thesis. The specific objectives necessary to meet this study's goal are: i) using the QMRA framework, develop a stochastic risk model that utilizes Monte Carlo simulation to output a probability distribution for risk associated with a model salad; and ii) incorporate the effects of salad-inclusive risk management strategies, such as kitchen-scale produce preparation techniques and proper storage, into the risk model to evaluate efficacy.

4.1 Materials and Methods

4.1.1 Risk Modeling Tool

Monte Carlo simulations of both the final exposure doses and the final outputs, the risk of listeriosis, were run in @Risk for Excel version 8.0 (Palisade, Ithaca, New York) with 10,000 iterations and a seed of 123. When sample size allowed ($n \geq 5$), parameter distributions were fit to data and ranked by the chi-square statistic; otherwise, a uniform distribution was assumed. In the case of a "tie" between distributions, the most plausible distribution was chosen (e.g., for growth rates, a bounded distribution). Finally, in @Risk, a sensitivity analysis was conducted using Spearman rank correlation coefficients, which were used to compare the magnitude of uncertainty and variability for each input parameter.

4.1.2 Dose-Response Modeling

An exponential dose-response model (Equation 1) was used to model *P(d)*, the probability of developing invasive listeriosis following consumption of a given dose, *d*. Parameter *k* represents the probability of an organism surviving to cause infection. This model was appropriate, as it assumes a random distribution of pathogen throughout the food (as one may expect in a mixed salad), and that one organism may survive to cause infection *(79)*. For microbial risk assessments, this model is generally preferred over models that have a minimum dose threshold *(187)*.

$$\boldsymbol{P(d) = 1 - e^{-kd}} \qquad \textbf{1}$$

Two approaches were used to create *k* distributions. The first approach transformed dose-dependent *k* values for generalized "susceptible" and "healthy" populations (FAO/WHO risk assessment, tables 2.18 and 2.21, respectively *(199)*), into *k* values specific to cancer patients, using relative susceptibilities (RS) (Equation 2) *(77, 130)*. Relative susceptibilities were based on listeriosis risk ratios for cancer patients compared to those under 65 with no immunocompromising conditions *(77)* and to all immunocompromised individuals *(130)*. Because hematological cancer patients had the highest RS in all studies, those values were used to devise a conservative model. These transformed data were pooled into one *k* distribution.

$$\boldsymbol{k_{cancer} = \frac{-ln\left[RS \times e^{\left(-k_{reference} \times dose\right)} - (RS - 1)\right]}{dose}} \qquad \textbf{2}$$

The second approach reconstructed the characterization of the *k* distribution by Pouillot et al. *(146)* for hematological cancer patients in @Risk. The final risk model was run separately with each *k* parameter approach so that the results could be compared.

4.1.3 Exposure Assessment

The simplified model salad product consisted of leafy greens (lettuce, spinach, mixed salad, arugula, kale, chicory, radicchio, endive, swiss chard, and watercress), tomatoes, and cucumbers. A thorough literature review was conducted to complete the exposure assessment for the model salad product. First, distributions for *L. monocytogenes* populations at the point of retail were created independently for each product. It can be assumed that the populations on retail samples reflect the changes in population that occur post-processing *(186)*.

Data from multiple leafy green studies (Table 4) were pooled (to better represent a variety of samples) to create a single distribution of *L. monocytogenes* concentration (log cfu/g). Requirements for data inclusion were that the study must report both prevalence and population and collect samples under retail conditions like those in the United States. MPN/g and cfu/g units were considered interchangeable due to similarity of study methods. For samples with undetectable levels of *L. monocytogenes*, concentration was recorded as the limit of detection in the corresponding study. If a range of concentrations was given, the geometric mean of that range was used. If only a maximum value was reported ("all samples were below 100 cfu/g"), the reported value was used. In studies where a portion of the positive samples were not enumerated, the geometric mean of the minimum and maximum concentrations from enumerated samples was used. The same approach was used for concentration of *L. monocytogenes* in cucumbers, although the distribution was fit to data from a single study *(80)*. These methods support a conservative risk model, in which results reflect the upper limit of exposure for this population.

Table 4: Summary of data used to create prevalence and concentration distribution for leafy greens

Product	Prevalence	Country	Reference
Leafy green vegetables	2/100 (2%)	Finland	*(133)*
Minimally processed salads	1/151 (0.7%)	Portugal	*(154)*
Packaged mixed salad	10/500 (2%)	Ireland	*(65)*
RTE salad and whole components	2/246 (0.8%)	Spain	*(1)*
RTE prepared salad vegetables	88/2,932 (3%)	United Kingdom	*(151)*
RTE leafy salad vegetables	0/35 (0%)	Croatia	*(101)*
RTE leafy salad vegetables	10/452 (2.2%)	Brazil	*(153)*
Minimally processed leafy vegetables	0/69 (0%)	Brazil	*(43)*
RTE and unprocessed leafy greens	15/6,115 (0.2%)	United States	*(201)*
Bagged leafy vegetable salads	22/2,966 (0.7%)	United States	*(74)*
Whole lettuce	0/151 (0%)	United Kingdom	*(112)*

Unfortunately, data on prevalence and concentration of *L. monocytogenes* in tomatoes in developed countries are lacking. This necessitated building a model in @Risk to simulate a tomato exposure scenario, using an approach previously applied by Todd *(179)*. For simplicity, the assumed contamination route was field irrigation water. Tomatoes then followed a simplified harvesting and processing pathway consisting of fruit growth in the field, a 200 ppm chlorinated wash, and transportation to retail stores in a delivery truck (Figure 3).

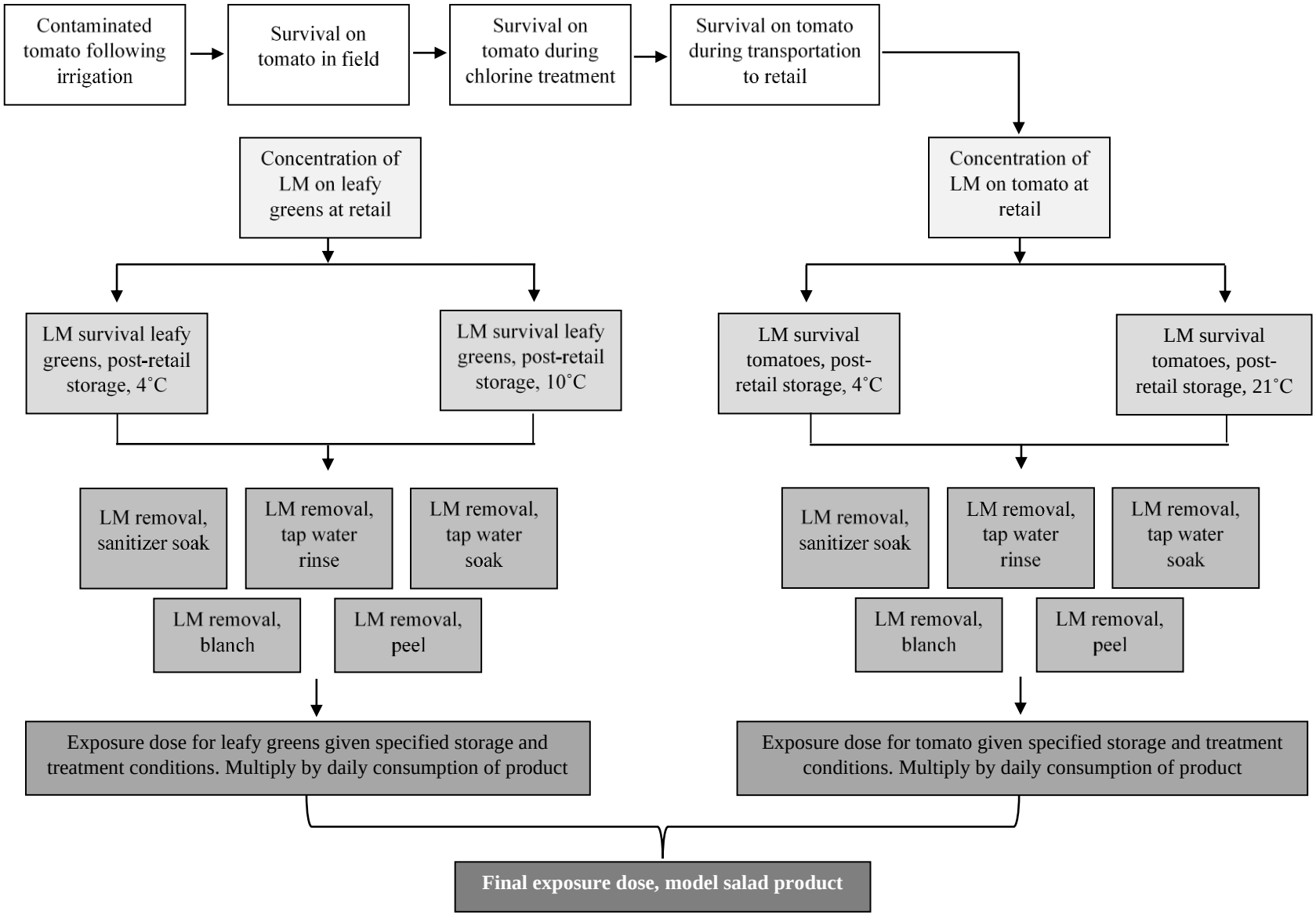

Figure 3: Components of exposure assessment

A distribution for *L. monocytogenes* prevalence and concentration in irrigation water was fit to data from Allard *(3)* and Watkins and Sleath *(192)*. Tomato irrigation and growing were based on Florida practices, as Florida is one of the two largest tomato producers in the United States *(200)*. Irrigation water applied per tomato plant was estimated by dividing drip irrigation water applied per acre of tomato plants *(170)* by the number of plants per acre *(66)*. It was then possible to calculate the *L. monocytogenes* concentration per contaminated tomato by multiplying the *L. monocytogenes* concentration by the water applied per plant, then dividing by 40 (the approximate number of tomatoes per tomato plant *(179)*). This implied that all irrigation water contacted the tomato fruit, which is improbable. However, the fraction of water that touches the tomato is a complex parameter depending on plant species, growth stage, and individual irrigation system design. Therefore, for the purpose of this risk assessment, it was conservatively assumed that all water contacted the tomato fruit.

Next, the changes in population of *L. monocytogenes* on tomatoes in the field, after a 200 ppm chlorine treatment during processing, and during truck transportation to retail were sequentially added to the exposure dose dose. Prior studies on *L. monocytogenes* survival on tomatoes were performed at 5°C *(144)* and 21°C *(15)*. According to expert opinion *(179)*, temperatures in the field and delivery truck can be approximated by uniform distributions between 10 and 40°C and 0 and 40°C, respectively. Because 21°C is close to the geometric mean of these distributions, survival at 21°C was assumed to be representative in the aforementioned cases. Based on growth study results *(15)*, it was assumed that *L. monocytogenes* populations on tomatoes in the field were constant after 8 days. Data from whole tomatoes that had and had not been treated with chlorine were used for *L. monocytogenes* survival during truck transport and in the growing field, respectively.

Survival distributions often included values that resulted in *L. monocytogenes* populations greater than maximum achievable populations (~10^9 cfu/g on various fresh produce *(86)*) or, if reduction due to the chlorine treatment was greater than *L. monocytogenes* on the tomato, below zero. Therefore, after each concentration calculation (in the field, after a chlorine treatment, and after transportation to retail), the distribution was truncated to exclude such values, using an IF function in @Risk. This function set values greater than 10^9 and less than 0 to 10^9 and 0, respectively.

Next, it was assumed that salad components were purchased from retail within one day of arrival, and exposure doses for each salad component were altered by various consumer risk management strategies. First, retail exposure doses were modified according to consumer storage practices. Refrigerated and elevated-temperature home storage conditions were considered (4-5°C and 10°C for leafy greens and cucumbers, and 4°C and 21°C for tomatoes, respectively). To calculate survival at the specified temperatures, populations of *L. monocytogenes* during storage (log cfu/g) were extracted from reference source data tables or graphs using DataThief III *(182)*, and change in population was divided by the number of days in the study. Population change per day was then multiplied by the number of days stored by consumers, the distribution for which was previously described in a USDA/FSIS risk assessment *(186)*. The original distribution used a range of values for the "most likely" parameter, but to reconstruct this distribution in @Risk, the mean of those values was used. Change in population during storage (log cfu) was added to the retail exposure dose.

L. monocytogenes reduction during each of the following kitchen-scale preparation techniques was then applied to each product: i) commercial sanitizer soak (1.5 to 2 min), which was Veggie Wash (2.0 oz/gal of water, Beaumont Products Inc., Kennesaw, Ga.) for tomatoes

and leafy greens and Ecolab sanitizer (Monogram Clean Force) for cucumbers; ii) tap water rinse (15 s); iii) tap water soak (1 to 5 min); iv) surface blanch (25 to 30 s); and v) peel. Because greens cannot be blanched without compromising integrity, or peeled, surface blanch and peel were only applicable to cucumbers and tomatoes. In these cases, tap water rinse was assumed for greens. Because few kitchen-scale foodborne pathogen removal studies exist for *L. monocytogenes*, others using *L. innocua* and/or *Salmonella* were included to fill distributions for some treatments, assuming the efficacy of the treatment would not vary substantially from the target scenario. Due to limited data for tomatoes specifically, distributions for tomato blanching and peeling included data from peas, potatoes, and cucumbers, and carrots and cucumbers, respectively. Products and references for each risk management parameter are listed inTable 5. Akin to the initial tomato concentration distribution, exposure dose distributions were truncated at each calculation to exclude impossible values.

Table 5: Summary of literature review for risk management parameters

Parameter	Pathogen	Product	Units	Reference
Storage time	*L. monocytogenes*	Fresh vegetables	Days	*186*
Growth 4-5°C	*L. monocytogenes*	Kale	Log cfu/g/day	*118, 119*
		Lettuce		*14, 28, 82, 94, 132, 171*
		Shredded chicory		*132*
		Packaged salad		*60, 82*
		Spinach		*54, 119, 135, 136*
		Tomato		*144*
		Cucumber		*10, 89*
Growth, 10°C	*L. monocytogenes*	Lettuce	Log cfu/g/day	*14, 25, 45, 82, 100, 110, 119*
		Packaged salad		*60, 82*
		Spinach		*119, 136*
		Cucumber		*48, 89, 110*
Growth, 21°C	*L. monocytogenes*	Tomato	Log cfu/g/day	*15*
Sanitizer soak	*L. monocytogenes*	Lettuce	Log cfu/g	*176*
		Spinach		*176*
		Tomato		*62*
		Cucumber		*75, Ch. 3 data*
	L. innocua	Lettuce		*98*
		Tomato		*98*
	Salmonella	Lettuce		*176*
		Spinach		*176*
		Tomato		*62*
Tap water rinse	*L. monocytogenes*	Lettuce	Log cfu/g	*176*
		Spinach		*176*
		Tomato		*62*
		Cucumber		*75, Ch. 3 data*
	L. innocua	Lettuce		*98*
		Tomato		*98*
	Salmonella	Lettuce		*176*
		Spinach		*176*
		Tomato		*62*
Tap water soak	*L. monocytogenes*	Lettuce	Log cfu/g	*131*
		Tomato		*47, 138*
		Cucumber		*75, Ch. 3 data*
Surface blanch	*L. monocytogenes*	Pea	Log cfu/g	*36*
		Potato		*36*
		Cucumber		*75, Ch. 3 data*
Peel	*Salmonella*	Carrot		*53*
	L. monocytogenes	Cucumber		*75, Ch. 3 data*
General population *k*, approach 1	*L. monocytogenes*	-	-	*199*
RS approach 1	*L. monocytogenes*	-	-	*77, 130*
Hematological cancer patient *k*, approach 2	*L. monocytogenes*	-	-	*146*

Distributions for consumption (g/day) of leafy greens and tomatoes were fit to data on consumption of these foods by outpatient cancer patients not placed on neutropenic diets *(87)*. It was assumed that cucumber consumption was the same as tomatoes. The post-treatment *L. monocytogenes* doses for each salad product (cfu/g) were then multiplied by corresponding consumption (g/day), resulting in a daily individual *L. monocytogenes* dose for each product. The daily product exposure doses were summed to yield the final daily exposure dose for a mixed salad. This value was used in further risk calculations.

4.1.4 Risk Characterization

Daily risk was calculated using Equation 1. Due to the previously discussed immunocompromising effects of chemotherapy, it can be assumed that the duration of the exposure period is one chemotherapy cycle, which can last from 3-4 weeks *(129)*. This time period had a uniform distribution, and daily risk was compounded to calculate risk per chemotherapy cycle (Equation 3).

$$\boldsymbol{P(d)_{chemo\ cycle} = 1 - \left(1 - P(d)_{daily}\right)^{days\ in\ chemo\ cycle}} \quad \textbf{3}$$

4.2 Results

4.2.1 Input Distributions

All distributions used to simulate the concentration of *L. monocytogenes* on tomatoes at retail are summarized in

Table 6. The *L. monocytogenes* concentration in irrigation water was modeled by a gamma distribution with $\alpha = 0.19133$ and $\beta = 169.53$. The amount of irrigation water applied to tomato crops, tomato plants per acre, weight of a tomato, number of days in the field after contamination, and time spent in the retail delivery truck were all modeled with uniform distributions. *L. monocytogenes* growth on tomatoes in the field and delivery truck, and survival

during a 200 ppm chlorine wash, were modeled with uniform and triangular distributions, respectively.

Table 6: Parameters used to model the concentration of *L. monocytogenes* on tomatoes at retail (cfu/g)

Parameter	n	Value/Distribution	Units	Source
Starting concentration of *L. monocytogenes* in irrigation water	25	Gamma: α=0.19133, β=169.53, shift=0.04	cfu/L	*61, 192*
Irrigation water applied to crop	29	Uniform: min=2271.246, max=15141.64	L/acre	*170*
Tomato plants per acre	1	Uniform: min=3630, max=5800	Plants/acre	*66*
Tomatoes per plant	1	40	Tomatoes/plant	*179*
Weight of tomato	1	Uniform: min=120, max=140	g/tomato	*15*
Days in field after contamination	1	Uniform: min=1, max=50	Days	*159*
L. monocytogenes growth on tomatoes, field, days 0-8	4	Uniform: min=0.2, max=0.2375	Log cfu/g/day	*15*
L. monocytogenes growth on tomatoes, field, days 8+	1	0	Log cfu/g/day	*15*
L. monocytogenes reduction on tomatoes, 200 ppm chlorine	5	Triangular: min=2.2273, likeliest=3.55, max=3.55	Log cfu/g	*16, 17*
L. monocytogenes transfer to uninoculated products, 200 ppm chlorine	1	0	% (0-1)	*19, 102*
Time in truck (transportation to retail)	1	Uniform: min=0, max=7	Days	*179*
L. monocytogenes growth on tomatoes, delivery truck	4	Uniform: min=0.175, max=0.2	Log cfu/g/day	*15*

The remaining distributions used in the exposure assessment are summarized in Table 7. Distributions for *k* parameter approaches 1 and 2 were inverse Gaussian and lognormal, respectively. The distribution for *k* using approach 1 was inverse Gaussian with $\mu=3.6559 \times 10^{-11}$, $\lambda=4.48437 \times 10^{-12}$, and shift=$-3.9456 \times 10^{-13}$. The mean and median of the distribution were 3.616×10^{-11} and 7.758×10^{-12}, respectively. Adopted from Pouillot et al., *(146)* the second approach *k* distribution was parameterized using the 50, 99, and 99.9 percentiles, 9.51×10^{-12},

5.44×10^{-8}, and 9.33×10^{-7}. The resulting mean and median were 9.605×10^{-9} and 9.510×10^{-12}, respectively.

Table 7: Distributions used in risk calculations

Parameter	n	Product	Distribution
L. monocytogenes concentration (cfu/g)	13,617	Leafy greens	Lognormal: μ=8.0691, σ=487.94, shift=0.0099782
	-	Tomatoes	See Table 3 for all input distributions
	100	Cucumbers	Exponential: β=0.5976, shift=0.034024
Consumption (g/day)	100	Leafy greens	Pearson5: α=2.1043 β=51.226, shift=-8.2991
	100	Tomatoes, cucumbers	Pearson5: α=1.0432 β=5.0161 shift=-1.5298
k, approach 1	8	-	Inverse Gaussian: μ=3.6559E-11 λ=4.84373E-12, shift=-3.94562E-13
k, approach 2	-	-	Lognormal: 50%=9.51E-12, 99%=5.44E-8, 99.9%=9.33E-7
Days in risk period (chemotherapy cycle)	1		Uniform: min=21, max=28
Post-retail storage time (days)	1		Pert: min=0.5, likeliest=3.5, max=10
L. monocytogenes growth (log cfu/g/day)	19	Leafy greens, 4-5°C	Extreme value: a=0.039401, b=0.065095
	28	Leafy greens, 10°C	Pert: min=-0.43015, likeliest= 0.32794, max=0.38253
	1	Tomatoes, 5°C	Static value=0.08
	4	Tomatoes, 21°C	Uniform: min=0.175, max=0.2
	3	Cucumbers, 4-5°C	Uniform: min=0.18, max=0.198333
	4	Cucumbers, 10°C	Uniform: min=0.041429, max=0.858
L. monocytogenes reduction, sanitizer soak (log cfu/g)	6	Leafy greens	Exponential: β=0.52167, shift=0.38306
	4	Tomatoes	Uniform: min=1.15, max=2.98
	6	Cucumbers	Pert: min=0.92434, likeliest=1.3828, max=2.4758
L. monocytogenes reduction, tap water rinse (log cfu/g)	6	Leafy greens	Laplace: μ=1.41, σ=0.26399
	4	Tomatoes	Uniform: min=1.82, max=2.44
	6	Cucumbers	Triangle: min=1.1569, likeliest=1.1569, max=1.75039
L. monocytogenes reduction, tap water soak (log cfu/g)	5	Leafy greens	Pert: min=0.5, likeliest=0.5, max=2.9438
	3	Tomatoes	Uniform: min=0.69, max=2.65
	6	Cucumbers	Triangle: min=0.17726, likeliest=0.75956, max=0.75956
L. monocytogenes reduction, surface blanch (log cfu/g)	14	Tomatoes	Extreme value: a=5.8666, b=1.9165
	6	Cucumbers	Logistic: α=5.16025, β=0.70488
L. monocytogenes reduction, peel (log cfu/g)	7	Tomatoes	Triangle: min=0.55093, likeliest=3.1402, max=3.1402
	6	Cucumbers	Beta general: α1=0.17621, α2=0.1547, min=1.0862, max=3.1402

L. monocytogenes concentration on leafy greens was modeled with a log-normal distribution with a mean of 8.07 and a standard deviation of 487.94. Concentration on cucumbers was described by an exponential distribution with a β of 0.5976, and shift of 0.034024. Consumption data for both leafy greens and tomatoes were fit to Pearson 5 distributions. Post-retail storage time was fit to a Pert distribution with minimum and maximum of 0.5 and 10 days, respectively, and a most likely value of 3.5 days. Growth in leafy greens at 4-5°C and 10°C was modeled by the extreme value distribution and the Pert distribution, respectively. Uniform distributions were used to model growth on cucumbers at both 4-5 °C (0.18 to 0.19833 log cfu/g/day) and 10 °C (0.041429 to 0.858 log cfu/g/day). Growth on tomatoes at 5°C was represented by a static value of 0.08 log cfu/g/day, and at 21°C, the uniform distribution ranged from 0.175 to 0.2 log cfu/g/day.

The effects of the sanitizer soak, tap water rinse, and tap water soak in greens were modeled by exponential, Laplace, and Pert distributions, respectively. In tomatoes, reductions due to sanitizer soak, tap water rinse, and tap water soak were fit to uniform distributions due to lack of data. For the same treatments in cucumbers, reduction distributions were Pert, triangle, and triangle, respectively. Across all products, mean reductions due to these treatments ranged between 0.57 and 2.13 log cfu/g. Blanching data were fit to extreme value and logistic distributions for tomatoes and cucumbers, respectively. Blanching was by far the most effective treatment, with mean reductions of 6.97 and 5.16 log cfu/g for tomatoes and cucumbers, respectively. The effects of peeling for tomatoes and cucumbers were fit to triangle and beta general distributions, respectively, and mean reductions ranged between 2.18 and 2.28 log cfu/g. Median daily exposure doses can be seen in Table 19.

4.2.2 Risk Distributions

Summarized distributions of risk per chemotherapy cycle for all treatments, storage conditions, and *k* approaches are shown in

. Across all scenarios, median risk ranged from approximately 7×10^{-11} to 1×10^{-7} (about 1 in 14 trillion to 1 in 10 million).

Table 8: Risk per chemotherapy cycle for each risk management strategy and *k* approach

Treatment	Storage Temp	*k* Approach	5%	50%	95%
Control	Refrigerated	1	2.6E-10	1.3E-08	9.5E-07
		2	4.2E-11	1.5E-08	1.6E-05
	High	1	8.9E-10	1.0E-07	6.8E-05
		2	1.5E-10	1.3E-07	5.3E-04
Sanitizer soak	Refrigerated	1	1.3E-11	7.4E-10	9.7E-08
		2	2.1E-12	8.7E-10	1.2E-06
	High	1	4.1E-11	5.8E-09	3.1E-06
		2	7.8E-12	7.2E-09	2.6E-05
Tap water rinse	Refrigerated	1	9.5E-12	5.2E-10	4.0E-08
		2	1.5E-12	5.6E-10	6.4E-07
	High	1	3.0E-11	3.9E-09	3.0E-06
		2	5.5-12	5.2E-09	2.2E-05
Tap water soak	Refrigerated	1	4.5E-11	2.5E-09	1.8E-07
		2	7.4E-12	2.8E-09	3.0E-06
	High	1	1.5E-10	2.0E-08	1.7E-05
		2	2.5E-11	2.6E-08	1.2E-04
Surface blanch tomato and cucumber, rinse greens	Refrigerated	1	4.2E-13	5.7E-11	2.1E-08
		2	7.9E-14	7.3E-11	1.9E-07
	High	1	1.2E-12	2.7E-10	1.4E-07
		2	2.7E-13	3.3E-10	1.2E-06
Peel tomato and cucumber, rinse greens	Refrigerated	1	3.2E-12	2.6E-10	3.2E-08
		2	5.1E-13	3.0E-10	4.8E-07
	High	1	1.1E-11	1.7E-09	1.1E-06
		2	2.1E-12	2.2E-09	8.9E-06

The approach used to model *k* had minimal effect on the predicted risk of listeriosis (Figure 4). Within a specified treatment and storage temperature, distribution of *k* was

observably tighter about the mean for the second *k* approach. The second *k* approach resulted in median risk less than one order of magnitude higher and 95th percentile risk up to two orders of

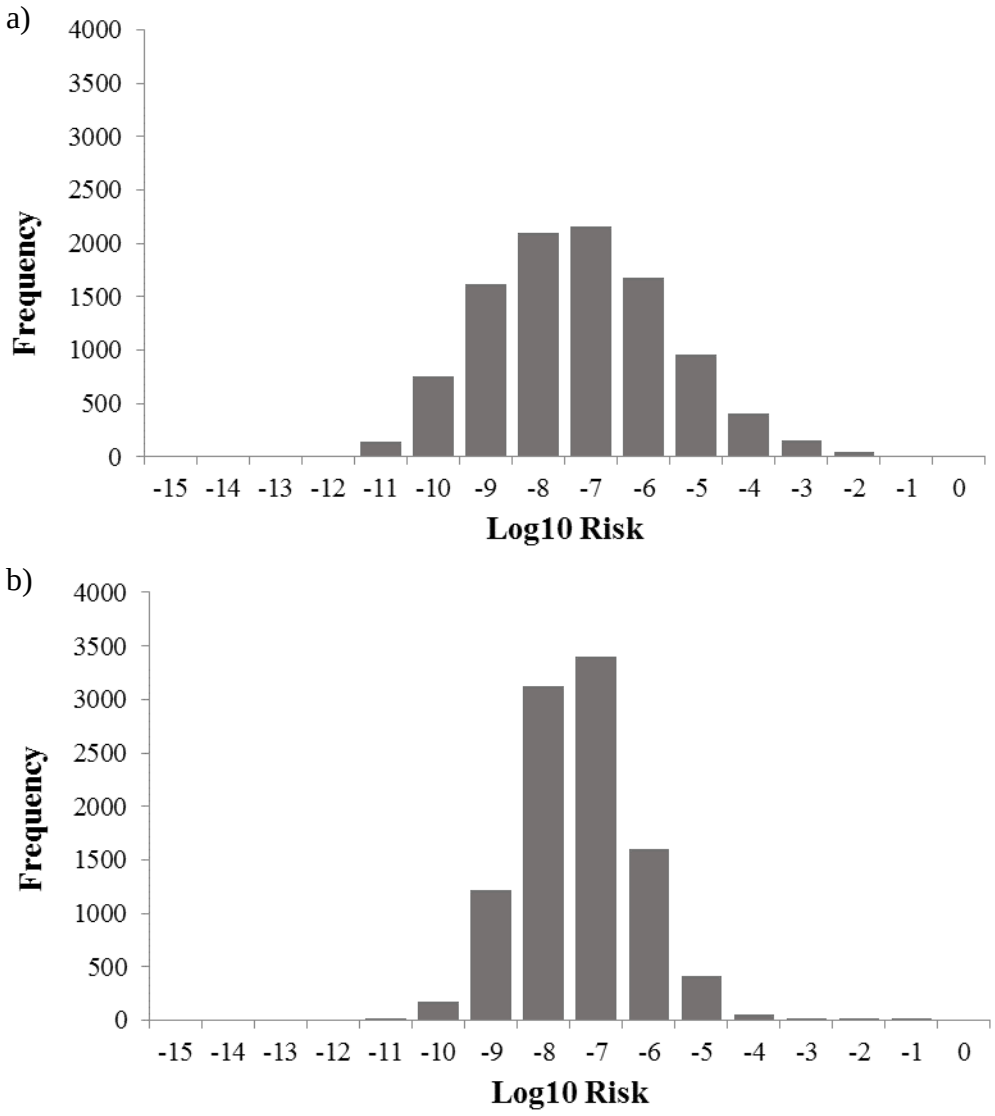

Figure 4: Histograms of risk distribution for refrigerated control salads, calculated using k approaches 1 (a) and 2 (b)

magnitude higher than that calculated using the first approach. Because of this marginal increase, risk distributions created using the second *k* approach were further visualized in boxplots (Figure 5). When the *k* approach and treatment were kept constant, temperature abuse of the salad

resulted in a median risk approximately two orders of magnitude higher than for refrigerated salad.

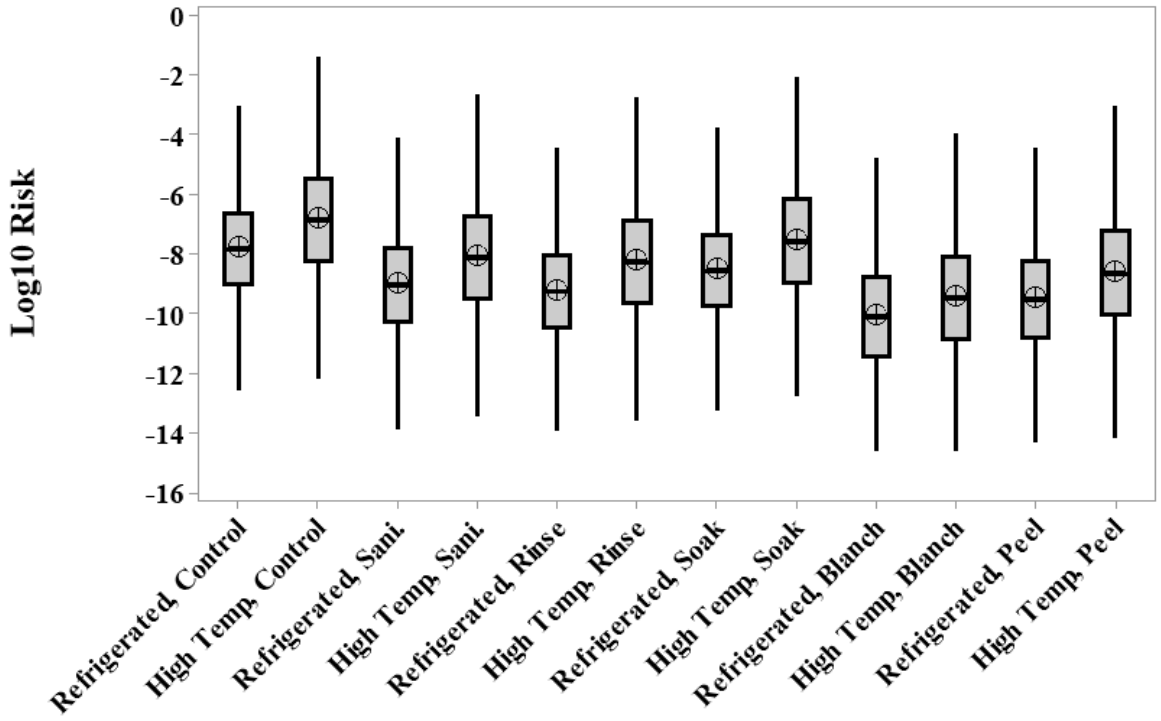

Figure 5: Box and whisker plots of risk distributions for each storage and treatment scenario, calculated using *k* approach 2.

When the salad was properly refrigerated, control scenarios resulted in the greatest median risk, which was 1.493×10^{-8} (approximately 1 in 67,000,000) and was one to two orders of magnitude greater than risk from salads that were subjected to sanitizer soak, tap water rinse, and tap water soak. Peeling refrigerated salad ingredients decreased median risk by about two orders of magnitude, to a maximum of 3.013×10^{-10}. For refrigerated salads in which tomatoes and cucumbers were blanched and greens were rinsed, median risk dramatically decreased to 7.347×10^{-11} (about 1 in 14 trillion). This scenario had the lowest risk of those tested.

4.2.3 Sensitivity Analysis

Sensitivity analyses revealed that consistently, for both *k* parameter approaches, the parameters with the top Spearman rank correlation coefficients varied depending on whether

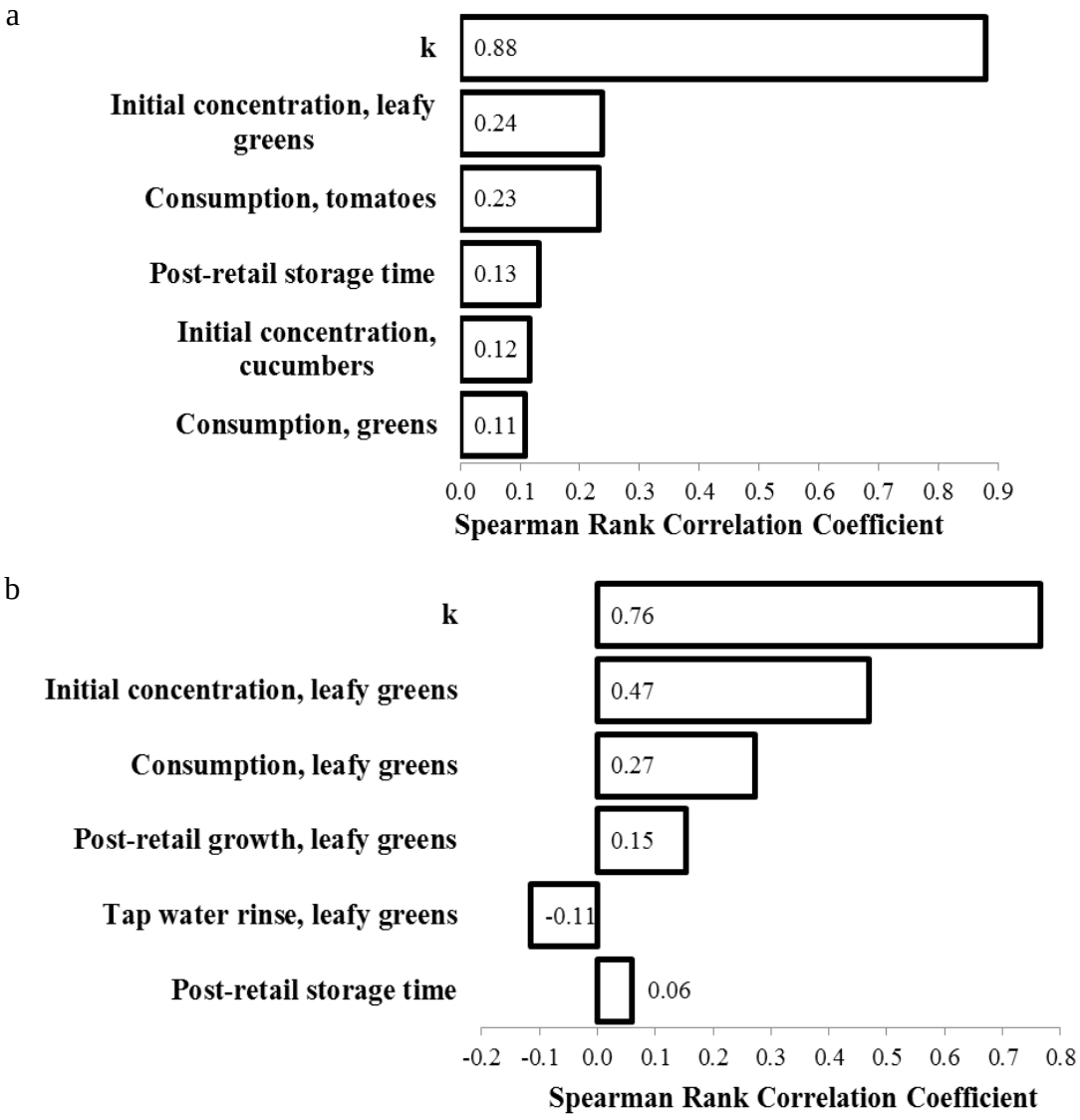

Figure 6: Spearman rank correlation coefficients for refrigerated control (a) and blanched (b) salads, calculated using *k* approach 2

ingredients were blanched (Figure 6). In such cases, the parameters with the highest correlation coefficients were *k* (0.47 to 0.76), initial contamination on greens (0.43 to 0.66), greens

consumption (0.24 to 0.37), *L. monocytogenes* growth on greens (0.15-0.27), and *L. monocytogenes* removal during rinsing (-0.11 to -0.15). For all other treatments and both *k* parameter approaches, the most impactful coefficients were *k* (0.47 to 0.88), initial contamination on greens (0.16 to 0.47), post-retail storage time (0.10 to 0.46), tomato consumption (0.17 to 0.37), initial cucumber contamination (0.07 to 0.27), and greens consumption (0.07 to 0.23). When these salads were stored at elevated temperatures, post-retail *L. monocytogenes* growth on cucumbers was also a highly influential parameter (0.24 to 0.46).

4.3 Discussion

The *k* parameter depends on both host and pathogen factors, as well as the more complex host-pathogen interaction. Distributions for *k* in this study were highly variable, but less so when the second *k* approach was used. This is because the characterization of the second *k* distribution was derived specifically for cancer patients, based on a wide array of epidemiological data *(146),* and could better describe the host-pathogen interaction. Still, the sensitivity analysis revealed that *k* had the greatest influence on risk. Because the exponential dose-response model solely relies on *k*, the distribution of which is highly variable, it is likely that a more complex dose-response model is needed to account for interactions between *L. monocytogenes* and cancer patients.

Values for *k* in this study were comparable to previously reported *k*-values. Buchanan et al. *(22)* and Lindqvist and Westöö *(111)* reported *k* values for immunocompromised populations of 1.179×10^{-10} and 5.6×10^{-10}, respectively. Chen et al. *(37)* studied the effect of *L. monocytogenes* genotypic subtype on parameter *k* and reported average *k* values for the general at-risk population of 1.32×10^{-8} to 5.01×10^{-11}, depending on genetic lineage. In the present study, average *k* values ranged from 9.61×10^{-9} to 3.62×10^{-11}. The similar *k* range between these

studies indicates that the current risk model reflects the wide range of *L. monocytogenes* genotypic subtypes present in foods. It was expected that, because of the increased listeriosis risk faced by cancer patients *(77, 130)*, *k* values for cancer patients would be higher than those for the general at-risk population. However, Pouillot et al. *(146)* found that mean *k* values for different immunocompromising conditions were within the same range described here. Some of the *k* values found in this and previous studies imply that a dose larger than 10^9 cfu/g would be required to produce substantial risk. As the FAO/WHO noted in their risk assessment *(199)*, this is hypothetical, and one should conclude that most of the population will not develop invasive listeriosis, despite the high dose. They also speculated that this contributes to the sporadic nature of listeriosis.

An average of 245 listeriosis cases per year occurred in France from 2001 and 2008, with 84 concurrent cancer and listeriosis cases *(77)*, indicating that about 34% of listeriosis cases were in cancer patients. A total of 660 cases of invasive listeriosis were reported in the United States in 2019 *(32)*, with about 34% (224) involving cancer patients based on the French estimate. Pinner et al. *(143)* tested food samples from listeriosis patients' refrigerators and found that 32% of the positive samples were vegetables. Therefore, for convenience it can be assumed that 71 of the 224 concurrent listeriosis cases were due to vegetables. It is estimated that 650,000 cancer patients are treated with chemotherapy in the U.S. per year *(34)*. When the median daily risk for refrigerated, untreated salad (6.086×10^{-10} for *k* approach 2) was compounded for a year (using Equation 3) and multiplied by the 650,000 patients undergoing chemotherapy, the result was 0.14 cases of listeriosis. While it is unlikely that all 71 expected cases from vegetables can be attributed to raw salad products, the estimated 0.14 cases still appears low. This could be due to simplification of the model, particularly in the exposure assessment. For instance, only one

exposure route (irrigation water) was considered for tomatoes for ease of calculations, even though it is possible for tomatoes to become contaminated through soil, plastic mulch, during harvest, and during postharvest handling *(179).* Additionally, contamination and growth can occur during packing and processing *(6, 147),* but this was not considered. This model also did not account for dynamic growth rates, which would have increased predicted growth during early storage. Considering the conservative assumptions in the exposure assessment (use of limit of detection instead of zero in concentration distributions and assuming all irrigation water contacts the tomatoes) this underestimation is even more profound, as it should represent the high end of risk. Clearly, a more robust model that incorporates adequate growth and exposure data is needed.

Information on the reduction of *L. monocytogenes* on produce by blanching is limited with data from only two studies included in this analysis. Reductions of *L. monocytogenes* on tomatoes after 30 s of blanching were described using Ceylan et al.'s *(36)* data for peas (about 7.5 to 8.3 log cfu/g) and potatoes (about 7.9 to 8.8 log cfu/g), which is only feasible in cases of extreme contamination. These results led to relatively low exposure doses, which were reflected in the low risk calculations. Therefore, while blanching was the most effective treatment in reducing risk, its perceived efficacy for tomatoes likely was influenced by biased data. More complete data are needed to better assess the listeriosis risk posed by blanched produce.

Nonetheless, risk calculated in the present study is consistent with past risk assessments and gives insight into the severity of impacts that immunosuppression during cancer treatment might have on the risk of foodborne illness. The 2003 FDA/FSIS assessment *(186)* calculated the risk per serving of refrigerated, untreated vegetables for perinates (4.8×10^{-10}), the elderly (8.2×10^{-12}), and intermediate-aged individuals (8.4×10^{-13}). Perinatal risk was analogous to

refrigerated, untreated salad risk in the present study (5.465×10^{-10} to 6.086×10^{-10}), reinforcing that adults with cancer have a significantly greater listeriosis risk than those with other immunocompromising conditions.

Ding et al. *(49)* presented a daily median risk of 2.47×10^{-6} for high-risk groups who consumed refrigerated lettuce that had been soaked in tap water. They noted that the model was oversimplified and overestimated the actual number of listeriosis cases in Korea. The corresponding risk from the present study was much lower, ranging from 1.032×10^{-10} to 1.135×10^{-10}. The discrepancy can be attributed to their model using *L. monocytogenes* contamination data from lettuce at the farm without accounting for further processing, during which the product would typically undergo a chlorine treatment, substantially reducing the pathogen concentration *(16, 17, 139)*. The current study used real or calculated *L. monocytogenes* concentration data from retail, so this step was incorporated.

Carrasco et al. *(29)* conducted a risk assessment on RTE salads in Spain and determined the mean risk per serving to range from 2.40×10^{-2} to 2.60×10^{-2} for high-risk populations. Their calculated risk is much greater than the mean daily risk for refrigerated, untreated salad in this study (2.456×10^{-7} to 4.451×10^{-6}); however, a different dose-response equation (and therefore risk distributions) was used. It is typically inappropriate to represent risk using a mean because risk distributions are often highly skewed, which heavily biases the mean. This bias influences the risk and can lead to overly conservative risk management strategies, such as the neutropenic diet. Also, Carrasco et al. *(29)* recognized that because their conservative growth model did not account for the slower growth under retail conditions, their model overpredicted the number of listeriosis cases by about three orders of magnitude (when compared to epidemiological data), resulting in an inflated daily risk.

When salad ingredients were refrigerated and rinsed, which is the current FDA recommendation *(183)*, median computed risk was remarkably low (about 5×10^{-10}, or 1 in 2 trillion). In nosocomial listeriosis outbreaks in which the food vehicle was identified, investigations revealed temperature abuse of the food and/or lack of hospital and patient-specific food safety guidelines *(40, 160)*. These results suggest that dietary inclusion of produce might be safe when appropriate food safety guidelines are strictly followed. The risks of chemotherapy-related morbidities, such as venous thromboembolism *(97)* and osteonecrosis *(7)*, which can be disabling and/or lethal, are far greater (2.2×10^{-2} and 1.1×10^{-2}, respectively) and are accepted by patients at the onset of treatment. However, the benefit of chemotherapy treatment is undoubtedly greater than the nutritional and wellness benefits from consuming raw produce. It would be imprudent to make a risk management decision without first quantifying these benefits in decision analysis. Thus, while the risk of listeriosis from salad consumption in this study was small, further work is needed to determine if the risk is acceptable for the target population.

Sensitivity analysis revealed that the model variables with the greatest impact on the result changed depending on the use of blanching as a treatment, because as previously noted, predicted *L. monocytogenes* reductions after blanching tomatoes and cucumbers generally removed almost all *L. monocytogenes* cells. In all scenarios, *k* was a highly influential parameter, which was expected, as it varies considerably with individual physiology, pathogen strain, and the host-pathogen interaction. The initial concentration of *L. monocytogenes* in leafy greens was also consistently a key parameter. For all products, median initial concentrations were similar, but because the distribution in leafy greens included more extreme values, the *Listeria* concentration was occasionally much higher than for other products, which was carried through the model. Because this distribution was fit to the most extensive, thorough, dataset used in the

model, the large effect on the result can be ascribed to inherent variability. In salads containing blanched tomatoes and cucumbers, leafy green consumption and post-retail growth were prominent parameters. This is reasonable because leafy greens were the only unblanched product, and therefore likely the main contributor of *L. monocytogenes*. Both these distributions included data from multiple (>5) studies, so the influence is due to innate variability.

Because greens were a major contributor in such cases, the risk model was rerun for salads composed of solely tomatoes and cucumbers. Median risk for a refrigerated, blanched, salad decreased from 3.3×10^{-10} to 4.9×10^{-14}. This difference was less (approximately one order of magnitude) for all other treatments. However, this suggests that for maximum risk reduction, it may beneficial to exclude products that cannot be blanched. Risk and exposure dose distributions for the tomato and cucumber salad are shown in Appendix D.

For products that were not blanched, consumption of both leafy greens and tomatoes were influential parameters. This is plausible, as individual diets are variable, particularly during cancer treatment when food aversions are common. However, because consumption data were from self-reporting surveys, patient estimates may lack precision and contribute to uncertainty. Because the survey was conducted amongst outpatient patients not following neutropenic diets, the results are likely applicable to similar patients treated without diet restrictions. In hospitals that enforce the neutropenic diet, salad ingredient consumption would be lower and likely less variable. Another key variable was consumer storage time, which was based on data from the FDA/FSIS 2003 risk assessment *(186)*. Collecting more data for this distribution would reduce uncertainty, but some inherent variability would always remain, as this is another parameter that varies from person to person.

The current model does not account for the positive effects that consuming produce has on the immune system *(5, 26, 30, 44, 72, 76, 107, 116, 124, 125, 167)*, which would theoretically reduce infection susceptibility. While these effects are widely known, they have not been quantified in a way that translates to risk analysis. Quantitatively evaluating these effects could provide further justification for produce inclusion in the diets of cancer patients and be utilized in decision analysis necessary to make informed risk management decisions. Furthermore, food safety risk communication for cancer patients is currently inadequate; 34% of cancer patients know they face an increased risk of foodborne illness *(55)*, and the FDA's "Food Safety for People with Cancer" *(183)* does not address food safety guidelines much beyond reinforcement or amplification of standard recommendations for healthy individuals. Therefore, an important future step for this work would involve the development of patient-centered informational material that accurately communicates the risk reported in this study.

4.4 Conclusions

The model presented in this study is the first to consider the unique, increased listeriosis risk faced by cancer patients, which was shown to be similar to that of perinates. Simplification of the exposure assessment led the model to slightly underestimate cases when compared to an appraisal of epidemiological data but results generally agreed with the few past studies on listeriosis risk from salads. The strong influence of the *k* parameter on risk combined with its high variability may indicate that a more advanced model would more accurately estimate risk.

The median listeriosis risk resulting from consuming refrigerated, untreated salad was far below other chemotherapy-related risks cancer patients routinely accept. This risk is even lower when salad components are blanched, and lower still when products that cannot be blanched are excluded from the salad, although the efficacy of blanching should be reevaluated once more

data become available. Consequently, this study supports the body of literature that questions the infection-reduction effectiveness of modified diets that exclude raw produce, if appropriate and sufficient pathogen control and reduction strategies are employed. However, quantitative decision analysis is needed to make valid risk management decisions. Future work will include adding more exposure scenarios in the tomato exposure analysis, using decision analysis to compare the benefits and risk of consuming raw produce, and creating effective risk communication materials for both patients and their caretakers.

CHAPTER 5: CONCLUSIONS

This thesis presented the results from two novel studies: the effectiveness of kitchen-scale treatments in decreasing *L. monocytogenes* on fresh produce, and the development of a listeriosis risk model for cancer patients who consume these products in prepared salads. Cancer patients are a vulnerable population currently not adequately served by available food safety information. This thesis found that rinsing, the current FDA recommendation for home produce preparation, is minimally effective in reducing *L. monocytogenes* populations and subsequent listeriosis risk for various types of produce. Other techniques seldom tested until the current study, such as blanching and/or peeling, were more effective in reducing *L. monocytogenes* and risk. However, more data are needed to validate these findings.

L. monocytogenes was particularly difficult to remove from celery due to internalization, as may be the case with other porous products. Yet, a comparable porous product, leafy greens, was considered in the risk assessment, resulting in low overall risk. However, because they could not be blanched, leafy greens were the main contributor to risk of *L. monocytogenes* in salads with blanched tomatoes and cucumbers. Once they were removed, risk decreased substantially. Decision analysis is needed to determine whether such a strategy would be beneficial for cancer patients.

The risk model predicted a daily median listeriosis risk congruent with that previously reported for perinates. This is suggestive of the unique, increased risk that cancer patients face, and confirms the need for specialized risk analyses and food safety interventions for this group. Present hospital-enforced food safety intervention methods, such as the neutropenic diet, are restrictive and not based on quantitative risk analysis. The second portion of the study found that median risks from refrigerated, untreated, and blanched salads were six to nine orders of

magnitude lower, respectively, than risks from debilitating and/or potentially lethal conditions resulting from chemotherapy, which are regularly accepted by cancer patients. This result suggests that excluding produce as a risk-reduction strategy should be reconsidered, and that shifting the focus to proper storage and hyper-hygienic preparation might result in sufficiently acceptable risk. This can be assessed in future decision analysis. This thesis considered one pathogen, *L. monocytogenes*, with particular evidence of concern. Other foodborne pathogens pose a risk to cancer patients who consume produce, and while they were not examined in the present study, results may be applicable to such situations.

CHAPTER 6: FUTURE WORK

The present study compared the efficacy of kitchen-scale microbial reduction strategies for apples, cucumbers, and celery. To better account for differences in produce morphology, future studies should include additional products, both porous and non-porous, as growing conditions and surface morphology play large roles in *L. monocytogenes* growth and removal. These results could be used to inform product-specific risk models and provide greater insight into safe dietary produce inclusion.

This thesis also did not consider declines in nutrient content and product integrity resulting from produce treatment, which affect the value of produce inclusion in terms of the immune-system and mental health benefits for patients. Investigating these food science and nutrition facets would help to help maximize the impact of this multidisciplinary project.

The exposure assessment conducted in this study was simplified and excluded several key routes of contamination. An improved version would consider contamination of tomatoes from soil, plastic mulch, during harvest, and in the packing house. It would also consider the various environments in which tomatoes are grown (green houses, fields, etc.), and how they affect risk. Modeling growth on all products with dynamic growth rates would support better understanding of how transportation times and consumer storage affect *L. monocytogenes* concentration and risk, lending to more factual recommendations.

Finally, it is crucial to use decision analysis methods to quantify the nutrition and wellness benefits associated with consuming raw produce. Doing so will facilitate the development of accurate risk management strategies and help determine acceptable risk for cancer patients.

The ultimate goal of this research, which will be carried out in subsequent doctoral work, is the development of improved training and risk communication materials for hospital staff, caretakers, and patients, and quantitative assessment of the risk-reduction benefits of these interventions. This will involve retesting the efficacy of produce treatments when performed by these groups and working with risk communication experts to create effective communication materials. Such materials could influence the widespread use of the neutropenic diet and hopefully improve patient diets and overall health outcomes.

APPENDICES

Appendix A: Produce Preparation Experiment Raw Data

Table 9: Apple sample and dilution weights

Treatment	Replication	Subsample	Weight (g)	Weight + Dilution (g)
Control	1	1	38.41	193.01
		2	30.04	148.92
	2	1	34.64	172.95
		2	28.79	143.18
	3	1	32.96	165.81
		2	29.67	148.16
Sanitizer soak	1	1	35.37	175.82
		2	25.84	129.97
	2	1	28.98	143.00
		2	42.80	213.43
	3	1	22.88	124.56
		2	32.94	164.93
Tap water rinse	1	1	28.91	144.61
		2	38.81	193.57
	2	1	42.80	213.43
		2	23.90	119.96
	3	1	28.67	140.97
		2	24.49	122.87
Tap water soak	1	1	33.90	167.90
		2	28.68	136.28
	2	1	40.58	200.75
		2	29.71	148.47
	3	1	35.40	177.12
		2	26.77	133.95
Blanch	1	1	34.18	170.05
		2	37.36	186.06
	2	1	33.03	164.84
		2	32.04	160.18
	3	1	34.95	173.80
		2	35.61	178.12
Rinse+peel	1	1	25.14	142.08
		2	18.49	91.90
	2	1	17.77	89.11
		2	24.50	122.52
	3	1	20.02	99.83
		2	33.26	167.00
Blanch+peel	1	1	19.20	95.92
		2	19.25	95.77
	2	1	22.52	112.55
		2	25.87	130.19
	3	1	14.89	75.45
		2	24.01	120.18

Table 10: Plate counts for apple treatments*

Treatment	Replication	Subsample	Number of Serial Dilutions**				
			+1	0	-1	-2	-3
Control	1	1		TNTC	TNTC	35, 47	2, 6
		2		TNTC	TNTC	86, 100	7, 12
	2	1		TNTC	TNTC	46, 62	6, 7
		2		TNTC	46, 65	8, 9	0, 2
	3	1		TNTC	TNTC	23, 26	38, 7
		2		TNTC	155, 160	15, 15	1, 2
Sanitizer soak	1	1	TNTC	TNTC	89, 91	5, 10	
		2	TNTC	TNTC	32, 40	5, 7	
	2	1	TNTC	37, 28	1, 3	0, 2	
		2	TNTC	TNTC	35, 39	0, 2	
	3	1	TNTC	TNTC	25, 31	0, 3	
		2	TNTC	140, 160	13, 24	0, 3	
Tap water rinse	1	1	TNTC	TNTC	52, 77	2, 7	
		2	TNTC	TNTC	53, 55	2, 10	
	2	1	TNTC	TNTC	39, 60	2, 6	
		2	TNTC	23, 39	2, 0	1, 0	
	3	1	54, 81	12, 14	2, 0	0, 0	
		2	TNTC	TNTC	140, 160	14, 20	
Tap water soak	1	1	TNTC	TNTC	191, 193	15, 20	
		2	TNTC	TNTC	23, 47	4, 2	
	2	1	TNTC	143, 198	18, 21	0, 0	
		2	TNTC	TNTC	36, 35	2, 6	
	3	1	TNTC	64, 76	4, 5	0, 0	
		2	TNTC	TNTC	63, 93	3, 4	
Blanch	1	1	9, 15	2, 4	0, 0		
		2	2, 4	0, 0	0, 0		
	2	1	9, 4	0, 1	0, 0		
		2	2, 1	0, 0	0, 0		
	3	1	0, 0	0, 0	0, 0		
		2	0, 2	0, 0	0, 0		
Rinse+peel	1	1	4, 11	1, 1	0, 0	0, 0	
		2	38, 36	3, 6	0, 0	0, 0	
	2	1	23, 50	2, 8	4, 2	0, 0	
		2	74, 51	5, 9	0, 1	0, 0	
	3	1	TNTC	TNTC	36, 39	2, 3	
		2	TNTC	219, 250	21, 26	1, 2	
Blanch+peel	1	1	7, 9	0, 0	0, 0	0, 0	
		2	1, 0	0, 0	0, 0	0, 0	
	2	1	82, 77	6, 9	0, 0	0, 0	
		2	9, 13	0, 0	0, 0	0, 0	
	3	1	TNTC	96, 109	11, 12	3, 1	
		2	0, 0	0, 0	0, 0	0, 0	

*Two samples were plated per dilution
**TNTC = Too numerous to count

Table 11: Cucumber sample and dilution weights

Treatment	Replication	Subsample	Weight (g)	Weight + Dilution (g)
Control	1	1	14.00	70.39
		2	14.54	72.39
	2	1	15.41	76.70
		2	16.52	87.32
	3	1	12.33	61.19
		2	10.76	54.00
Sanitizer soak	1	1	17.82	88.59
		2	11.32	56.20
	2	1	10.15	50.24
		2	14.22	71.36
	3	1	14.64	73.17
		2	15.36	76.78
Tap water rinse	1	1	14.51	72.58
		2	15.06	75.92
	2	1	18.32	93.10
		2	14.51	72.25
	3	1	14.57	76.75
		2	15.50	80.73
Tap water soak	1	1	15.35	76.54
		2	12.68	64.12
	2	1	11.57	57.40
		2	15.43	76.52
	3	1	15.43	97.27
		2	19.53	100.62
Blanch	1	1	12.65	63.77
		2	14.92	74.51
	2	1	14.94	74.52
		2	11.90	59.49
	3	1	14.26	70.89
		2	16.70	82.41
Rinse+peel	1	1	10.39	51.75
		2	9.76	48.69
	2	1	10.36	51.48
		2	11.46	57.08
	3	1	9.32	46.10
		2	12.80	65.10
Blanch+peel	1	1	9.28	46.38
		2	10.23	50.28
	2	1	13.71	68.62
		2	10.37	51.48
	3	1	10.05	50.18
		2	10.72	52.99

Table 12: Plate counts for cucumber treatments*

Treatment	Replication	Subsample	Number of Serial Dilutions**				
			+1	0	-1	-2	-3
Control	1	1		TNTC	TNCT	83, 85	6, 6
		2		TNTC	TNTC	105, 114	8, 14
	2	1		TNTC	TNTC	112, 142	8, 12
		2		TNTC	TNCT	110, 122	8, 10
	3	1		TNTC	TNTC	36, 70	5, 8
		2		TNTC	TNTC	40, 44	2, 2
Sanitizer soak	1	1		TNTC	TNTC	39, 37	4, 4
		2		TNTC	90, 103	8, 11	0, 1
	2	1		TNTC	TNTC	45, 38	2, 3
		2		TNTC	TNTC	31, 25	0, 2
	3	1		TNTC	TNTC	40, 44	2, 6
		2		TNTC	183, 175	21, 22	3, 3
Tap water rinse	1	1		TNTC	TNTC	37, 51	4, 8
		2		TNTC	TNTC	49, 61	3, 5
	2	1		TNTC	TNTC	25, 31	1, 5
		2		TNTC	TNTC	57, 65	1, 2
	3	1		TNTC	143, 196	10, 21	1, 2
		2		TNTC	TNTC	30, 33	2, 3
Tap water soak	1	1		TNTC	TNTC	168, 179	19, 24
		2		TNTC	TNTC	TNTC	33, 29
	2	1		TNTC	TNTC	TNTC	52, 73
		2		TNTC	TNTC	TNTC	50, 42
	3	1		TNTC	TNTC	65, 78	7, 8
		2		TNTC	TNTC	73, 86	4, 11
Blanch	1	1	38, 51	3, 5	1, 0		
		2	0, 0	0, 0	0, 0		
	2	1	5, 7	1, 1	0, 0		
		2	1, 0	0, 0	0, 0		
	3	1	3, 4	0, 0	0, 0		
		2	TNTC	59, 80	3, 5		
Rinse+peel	1	1	TNTC	TNTC	TNTC	78, 80	
		2	TNTC	80, 91	11, 15	1, 0	
	2	1	TNTC	166, 163	18, 23	0, 1	
		2	TNTC	177, 168	18, 18	1, 0	
	3	1	TNTC	TNTC	226, 195	17, 22	
		2	TNTC	31, 36	2, 0	1, 3	
Blanch+peel	1	1	0, 0	0, 0	0, 0		
		2	0, 0	0, 0	0, 0		
	2	1	3, 4	1, 0	0, 0		
		2	1	0, 0	0, 0		
	3	1	4, 5	1, 3	0, 0		
		2	0	0, 0	0, 0		

*Two samples were plated per dilution
**TNTC = Too numerous to count

Table 13: Celery sample and dilution weights

Treatment	Replication	Subsample	Weight (g)	Weight + Dilution (g)
Control	1	1	13.30	66.91
		2	8.29	42.18
	2	1	19.39	97.07
		2	11.39	57.63
	3	1	12.74	63.19
		2	15.29	77.47
Sanitizer soak	1	1	11.12	54.97
		2	18.84	95.31
	2	1	12.56	62.75
		2	14.33	71.75
	3	1	19.40	102.39
		2	17.77	88.00
Tap water rinse	1	1	14.68	54.97
		2	16.05	80.54
	2	1	23.74	117.17
		2	21.52	107.98
	3	1	10.22	51.24
		2	13.19	65.58
Tap water soak	1	1	13.97	69.90
		2	9.66	49.29
	2	1	15.18	76.16
		2	14.41	74.16
	3	1	9.76	48.99
		2	23.85	119.26
Blanch	1	1	16.01	79.98
		2	16.91	85.17
	2	1	23.14	115.92
		2	16.62	81.25
	3	1	16.03	80.47
		2	15.38	76.24

Table 14: Plate counts for celery treatments*

Treatment	Replication	Subsample	Number of Serial Dilutions**				
			-1	-2	-3	-4	-5
Control	1	1	TNTC	TNTC	TNTC	79, 97	
		2	TNTC	TNTC	273, 292	28, 21	
	2	1		TNTC	161, 171	15, 20	2, 3
		2		TNTC	257, 221	28, 20	4, 1
	3	1		TNTC	173, 178	11, 14	0, 1
		2		TNTC	236, 263	16, 23	1, 2
Sanitizer soak	1	1	TNTC	173, 190	18, 19	3, 1	
		2	TNTC	TNTC	TNTC	55, 50	
	2	1		227, 230	18, 34	3, 3	0, 0
		2		TNTC	51, 60	6, 9	0, 0
	3	1		108, 121	13, 14	1, 0	0, 0
		2		TNTC	TNTC	67, 80	8, 10
Tap water rinse	1	1	TNTC	TNTC	TNTC	127, 152	
		2	TNTC	TNTC	TNTC	47, 55	
	2	1		126, 127	13, 8	0, 0	0, 0
		2		196, 210	12, 27	1, 0	1, 0
	3	1		TNTC	127, 143	11, 17	1, 0
		2		115, 143	11, 15	0, 4	0, 0
Tap water soak	1	1	TNTC	TNTC	65, 69	6, 7	
		2	TNTC	TNTC	70, 74	5, 6	
	2	1		TNTC	52, 72	7, 7	2, 1
		2		TNTC	29, 38	4, 5	0, 0
	3	1		TNTC	TNTC	30, 39	3, 2
		2		TNTC	27, 29	5, 1	0, 0
Blanch	1	1	TNTC	TNTC	220, 242		
		2	TNTC	TNTC	278, 296		
	2	1	TNTC	164, 168	11, 17	1, 4	
		2	TNTC	84, 115	9, 9	0, 0	
	3	1	125, 99	6, 15	2, 0	0, 0	
		2	226, 241	17, 22	1, 2	0, 0	

*Two samples were plated per dilution
**TNTC = Too numerous to count

Table 15: Celery inoculum internalization experiment sample weights and dilutions

Distance from inoculated end (cm)	Replication	Subsample	Weight (g)	Weight + Dilution (g)
0.1	1	1	0.92	5.30
		2	1.01	6.79
	2	1	0.40	2.76
		2	0.58	3.02
	3	1	0.61	3.11
		2	0.39	3.1
1.1	1	1	1.27	6.25
		2	1.99	6.79
	2	1	1.58	8.25
		2	0.93	4.60
	3	1	1.58	7.94
		2	1.35	6.78
2.1	1	1	1.53	7.67
		2	2.36	11.88
	2	1	1.50	8.45
		2	1.25	6.45
	3	1	1.78	8.88
		2	1.45	7.24
3.1	1	1	1.54	8.99
		2	2.69	13.55
	2	1	1.46	7.29
		2	1.28	6.35
	3	1	1.77	8.97
		2	1.80	9.00
4.1	1	1	1.58	8.00
		2	1.11	6.20
	2	1	1.51	7.83
		2	2.11	10.68
	3	1	1.86	10.00
		2	1.63	8.18
5.1	1	1	1.55	8.05
		2	2.48	13.45
	2	1	1.41	8.20
		2	1.01	5.85
	3	1	1.85	9.48
		2	1.77	8.99
6.1	1	1	1.74	9.13
		2	2.55	13.70
	2	1	1.31	6.67
		2	1.07	5.52
	3	1	1.71	8.53
		2	1.85	9.39

Table 15 (cont'd)

Distance from inoculated end (cm)	Replication	Subsample	Weight (g)	Weight + Dilution (g)
7.1	1	1	1.12	5.62
		2	2.41	12.55
	2	1	1.31	7.34
		2	1.23	7.61
	3	1	1.92	9.87
		2	1.91	9.67
8.1	1	1	1.20	6.13
		2	2.37	12.10
	2	1	1.43	7.69
		2	1.15	5.46
	3	1	1.78	8.96
		2	1.51	7.93
9.1	1	1	1.41	7.08
		2	2.46	12.63
	2	1	1.42	7.13
		2	0.96	5.16
	3	1	1.51	7.50
		2	2.42	11.99

Table 16: Plate counts for celery inoculum internalization experiment*

Distance from Inoculated End (cm)	Replication	Subsample	Number of Serial Dilutions**				
			-0	-1	-2	-3	-4
0.1	1	1	TNTC	TNTC	TNTC	179, 176	22, 18
		2	TNTC	TNTC	TNTC	TNTC	32, 34
	2	1	TNTC	TNTC	TNTC	TNTC	44, 68
		2	TNTC	TNTC	TNTC	TNTC	88, 88
	3	1	TNTC	TNTC	TNTC	TNTC	61, 80
		2	TNTC	TNTC	TNTC	TNTC	55, 69
1.1	1	1	TNTC	170, 167	11, 25	1, 6	
		2	TNTC	TNTC	TNTC	44, 46	
	2	1	TNTC	TNTC	TNTC	37, 42	
		2	TNTC	TNTC	TNTC	50, 52	
	3	1	TNTC	TNTC	TNTC	40, 44	
		2	TNTC	TNTC	TNTC	69, 78	
2.1	1	1	2, 2	1, 0	0, 0	0, 0	
		2	TNTC	70, 71	5, 9	1, 0	
	2	1	22, 26	3, 4	0, 0	0,0	
		2	11, 5	0, 2	0, 0	0, 0	
	3	1	TNTC	115, 132	14, 18	1, 1	
		2	TNTC	TNTC	87, 102	6, 14	
3.1	1	1	168, 148	13, 19	2, 0	0, 0	
		2	TNTC	51, 61	2, 6	1, 1	
	2	1	0, 0	0, 0	0, 0	0, 0	
		2	4, 6	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	TNTC	TNTC	32, 42	5, 7	
4.1	1	1	2, 1	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
5.1	1	1	22, 24	3, 1	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 0	0, 0	0, 1	
		2	0, 0	0, 1	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
6.1	1	1	TNTC	46, 63	9, 9	1, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 0	0, 0	0, 0	
		2	0,0	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 1	0, 0	0, 0	

Table 16 (cont'd)

Distance from Inoculated End (cm)	Replication	Subsample	Number of Serial Dilutions**				
			-0	-1	-2	-3	-4
7.1	1	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 1	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 1	0, 0	0, 0	0, 0	
8.1	1	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
9.1	1	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	2	1	0, 0	0, 1	0, 0	0, 0	
		2	0, 0	0, 0	0, 0	0, 0	
	3	1	0, 0	0, 0	0, 0	0, 0	
		2	0, 0	0, 1	0, 0	0, 0	

*Two samples were plated per dilution
**TNTC = Too numerous to count

Table 17: Apple peeling experiment sample weights and dilutions

Peeler	Replication	Subsample	Weight (g)	Weight + Dilution (g)
Standard	1	1	24.11	120.55
handheld		2	23.95	118.24
	2	1	23.76	119.11
		2	21.3	106.6
	3	1	20.6	101.57
		2	22.83	113.8
Apple	1	1	14.38	72.13
		2	11.96	59.8
	2	1	18.24	91.32
		2	11.08	55.42
	3	1	21.38	106.89
		2	13.78	68.90
Positive	1	1	28.08	141.43
control		2	29.06	145.35
	2	1	27.67	138.20
		2	30.62	153.39
	3	1	36.19	179.99
		2	32.60	163.10

Table 18: Apple peeling experiment plate counts*

Peeler	Replication	Subsample	Number of Serial Dilutions**				
			+1	-0	-1	-2	-3
Standard	1	1	TNTC	103, 117	8, 10	0, 0	
handheld		2	38, 41	3, 7	0, 1	0, 0	
	2	1	TNTC	TNTC	55, 70	10, 2	
		2	TNTC	TNTC	48, 51	7, 8	
	3	1	20, 28	2, 5	1, 0	0, 0	
		2	65, 43	7, 13	0, 2	0, 0	
Apple	1	1	83, 56	6, 9	1, 1	0, 0	
		2	33, 35	1, 3	0, 0	0, 0	
	2	1	TNTC	77, 81	7, 11	1, 2	
		2	TNTC	TNTC	51, 65	4, 6	
	3	1	67, 72	7, 13	1, 2	0, 0	
		2	TNTC	38, 41	4, 6	0, 0	
Positive	1	1		TNTC	109, 114	14, 15	2, 1
control		2		TNTC	195, 136	18, 36	2, 5
	2	1		TNTC	89, 67	8, 9	2, 2
		2		TNTC	214, 249	20, 16	0, 0
	3	1		TNTC	22, 27	2, 11	1, 2
		2		TNTC	TNTC	113, 98	13, 16

*Two samples were plated per dilution
**TNTC = Too numerous to count

Appendix B: *L. monocytogenes* Exposure Dose Summary

Table 19: Daily exposure dose (cfu/g) for *L. monocytogenes* in fresh salad

Treatment	Storage Temp	5%	50%	95%
Control	Refrigerated	3.3	6.3E01	1.5E03
	High	9.4	4.E02	1.5E05
Sanitizer soak	Refrigerated	1.6E-01	3.4	6.0E01
	High	4.7E-01	2.5E01	7.5E03
Tap water rinse	Refrigerated	1.2E-01	2.3	6.5E01
	High	3.2E-01	3.4E01	6.8E03
Tap water soak	Refrigerated	5.2E-01	1.2E01	2.7E02
	High	1.5	8.3E01	3.9E04
Flash blanch (rinse greens)	Refrigerated	5.2E-03	2.6E-01	5.1E01
	High	1.3E-01	1.2	3.3E02
Peel (rinse greens)	Refrigerated	3.6E-02	1.2	6.3E01
	High	1.2E-01	7.3	2.8E03

Appendix C: *L. monocytogenes* Exposure Dose Distributions

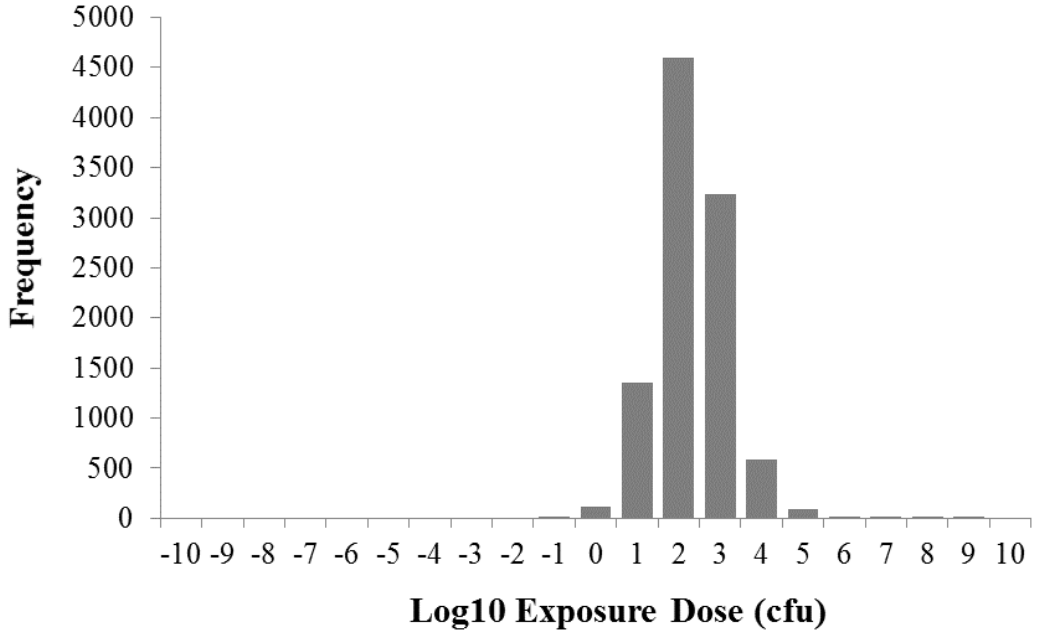

Figure 7: Exposure dose histogram for refrigerated, untreated salad

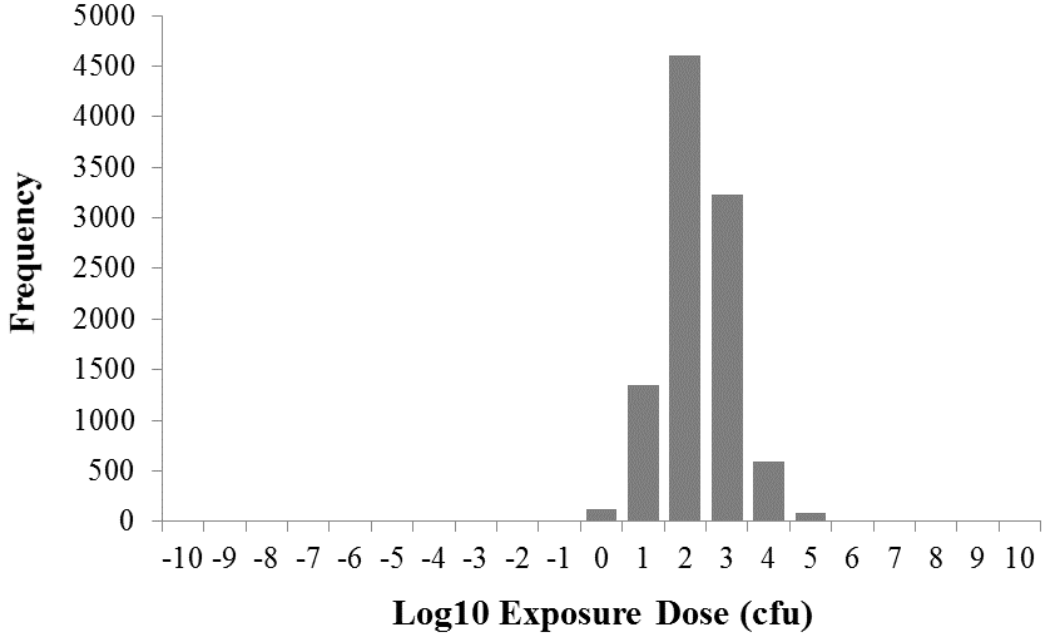

Figure 8: Exposure dose histogram for elevated temperature, untreated salad

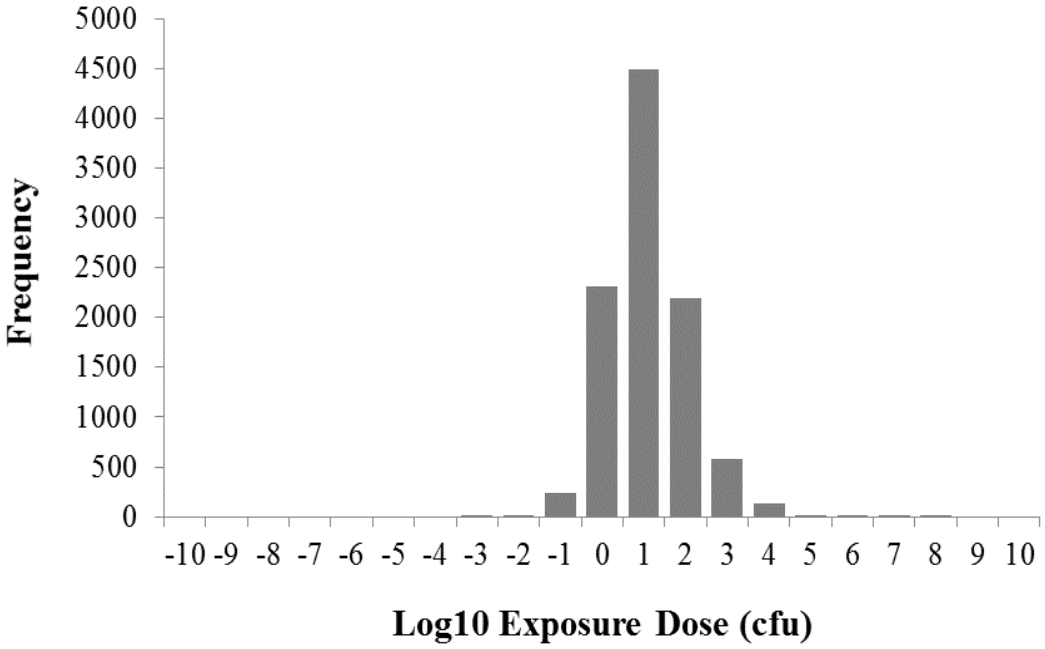

Figure 9: Exposure dose histogram for refrigerated salad treated with sanitizer soak

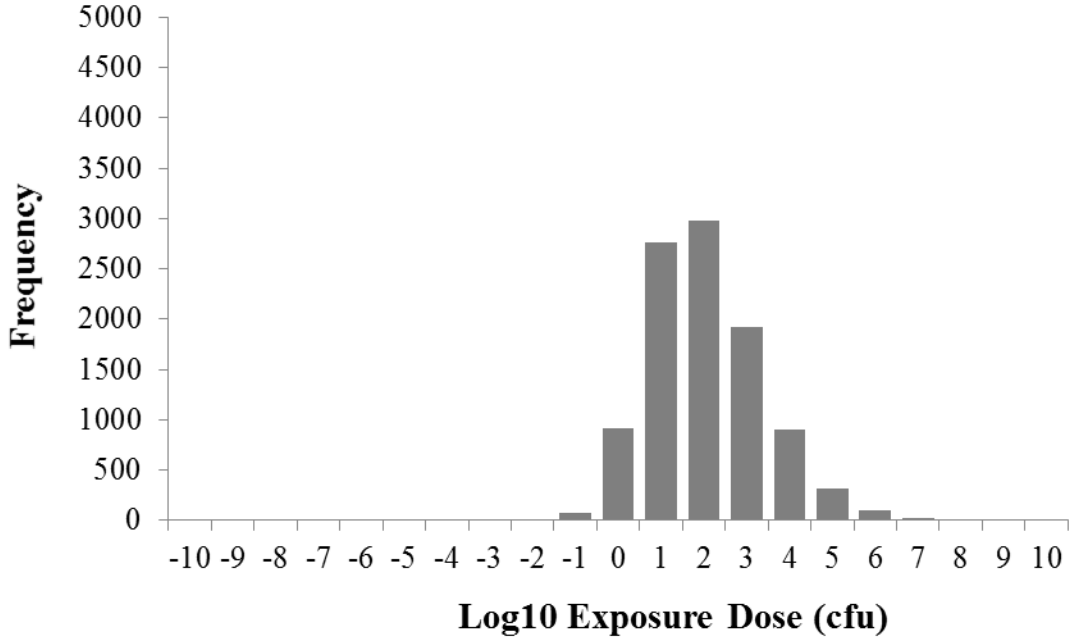

Figure 10: Exposure dose histogram for elevated temperature salad treated with sanitizer soak

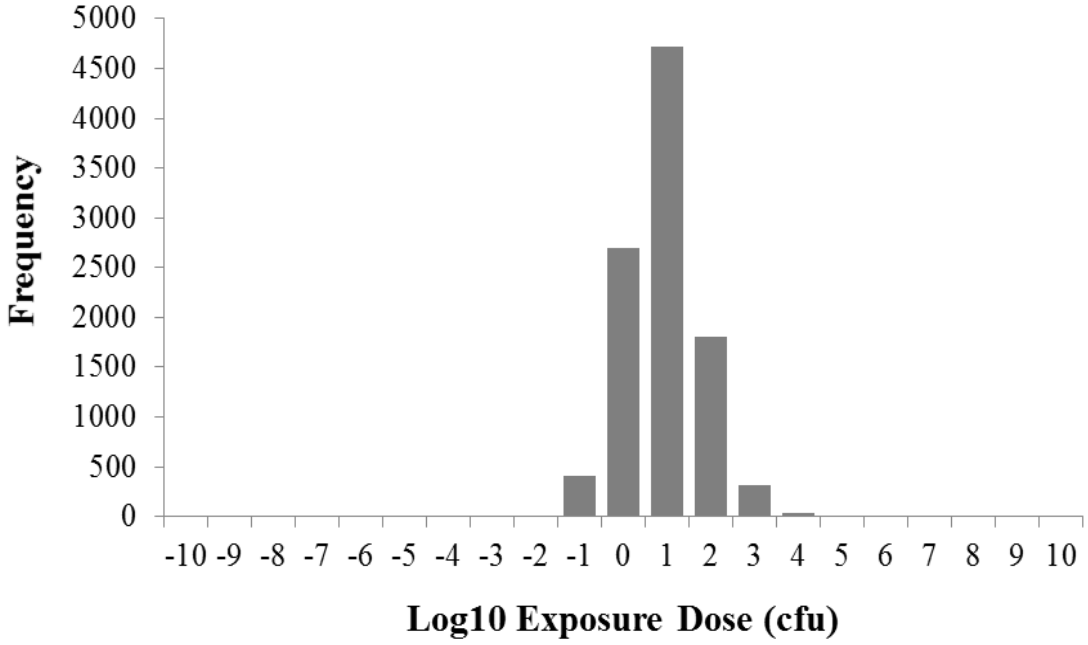

Figure 11: Exposure dose histogram for refrigerated salad treated with tap water rinse

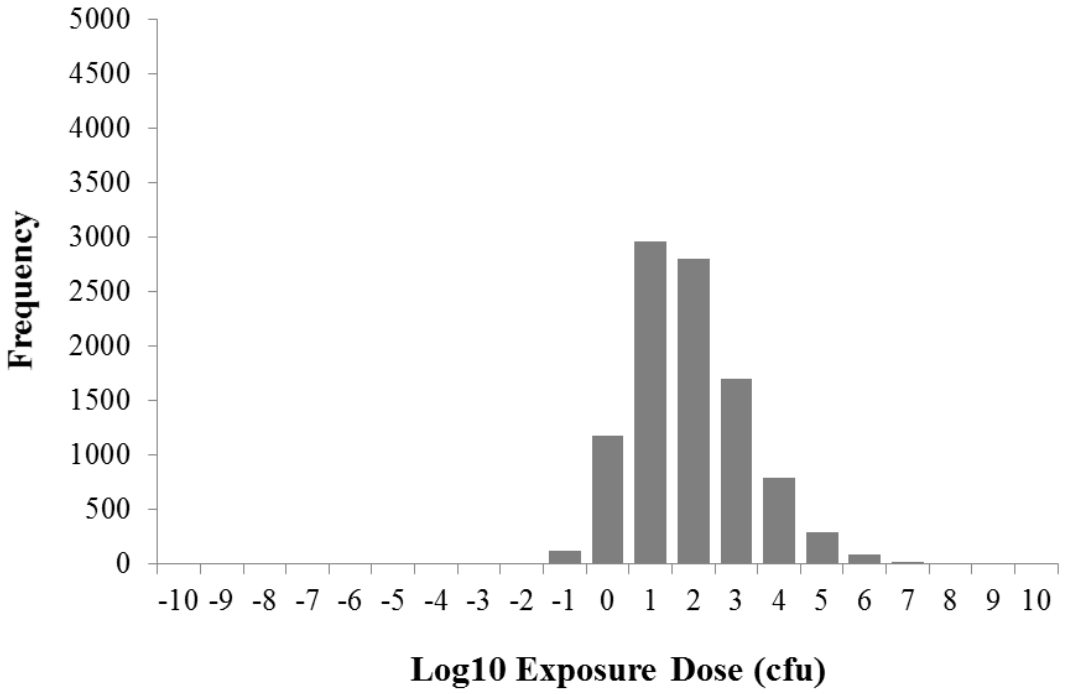

Figure 12: Exposure dose histogram for elevated temperature salad treated with tap water rinse

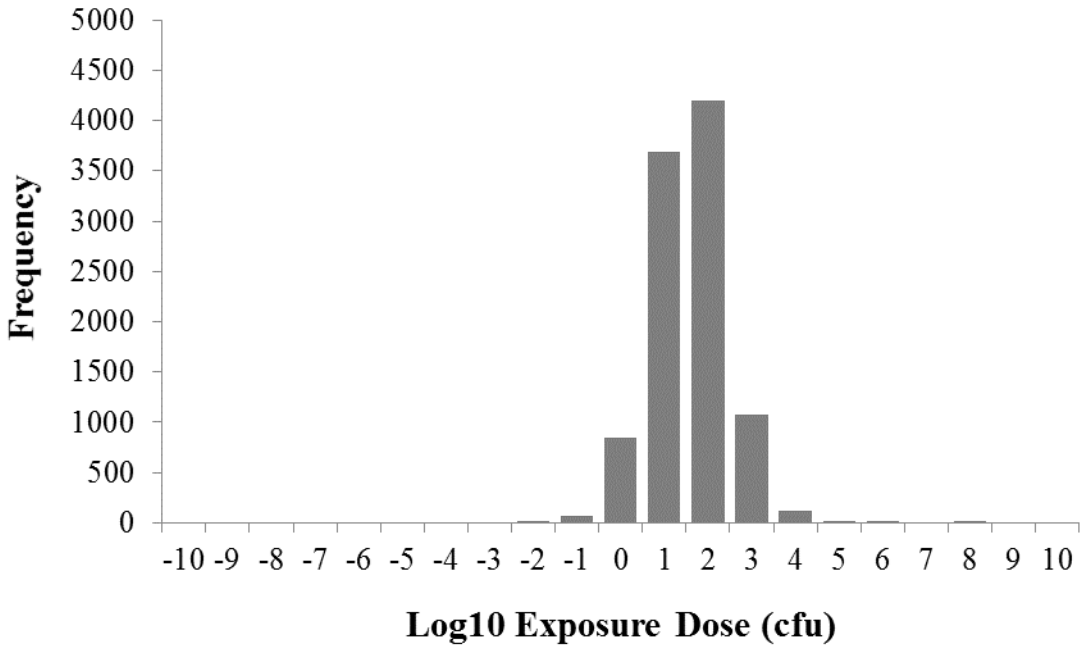

Figure 13: Exposure dose histogram for refrigerated salad treated with tap water soak

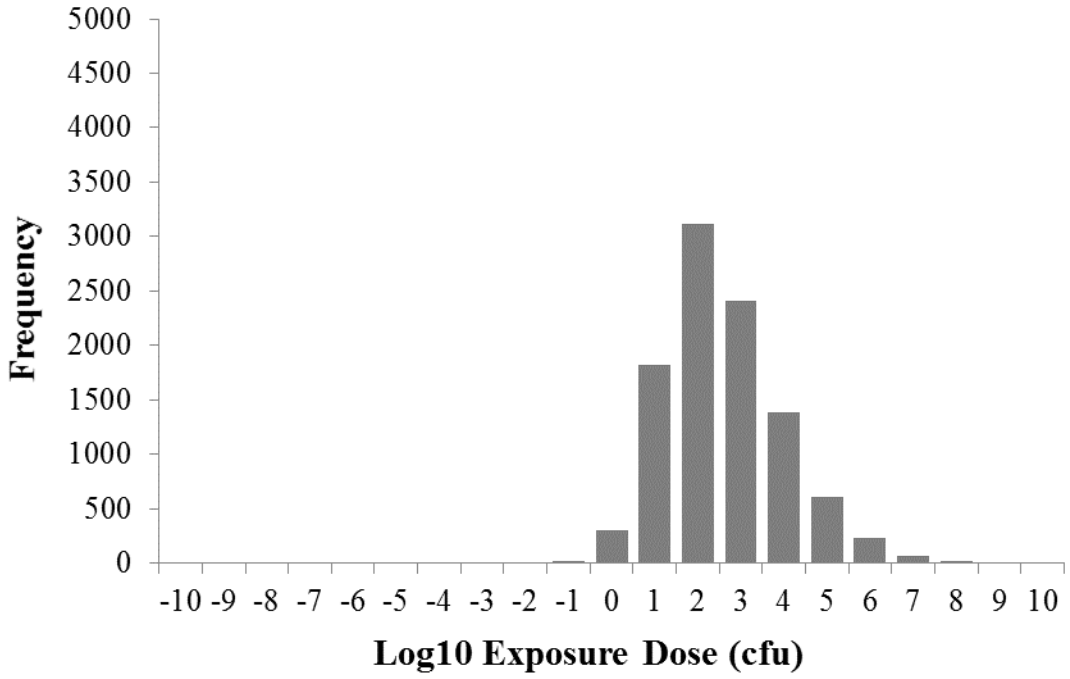

Figure 14: Exposure dose histogram for elevated temperature salad treated with tap water soak

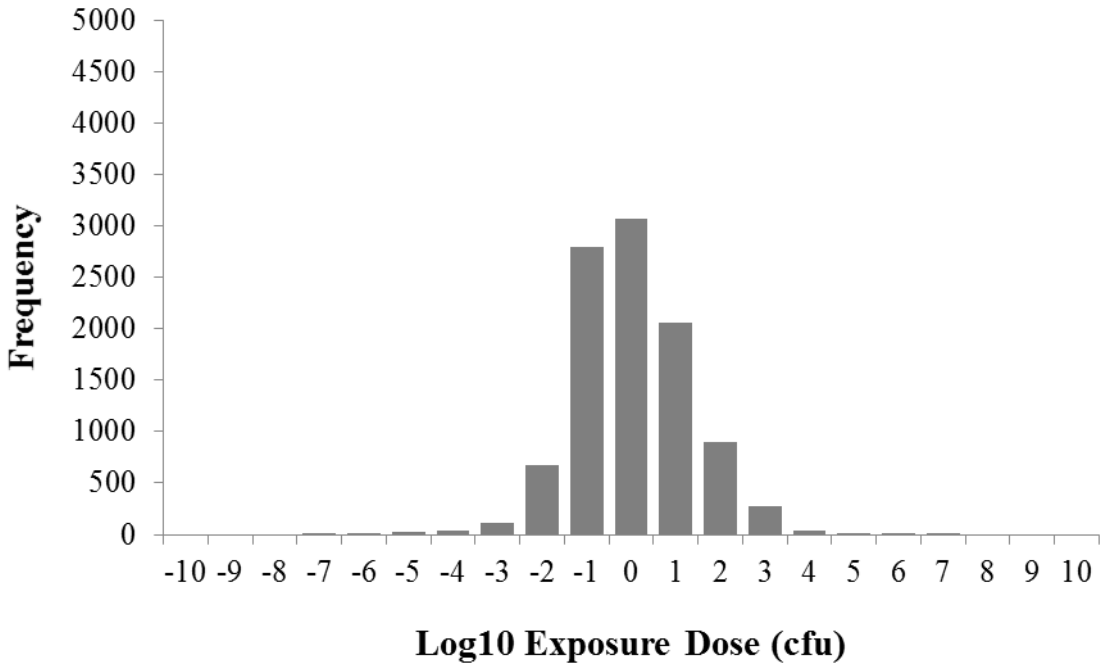

Figure 15: Exposure dose histogram for refrigerated salad treated by blanching

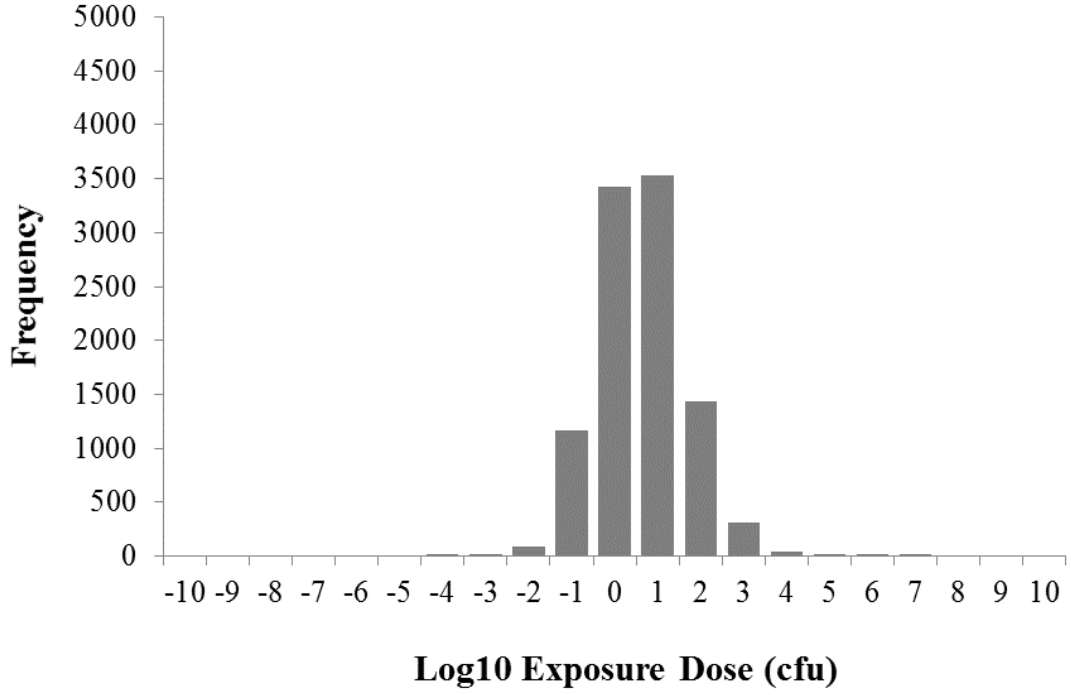

Figure 16: Exposure dose histogram for elevated temperature salad treated by blanching

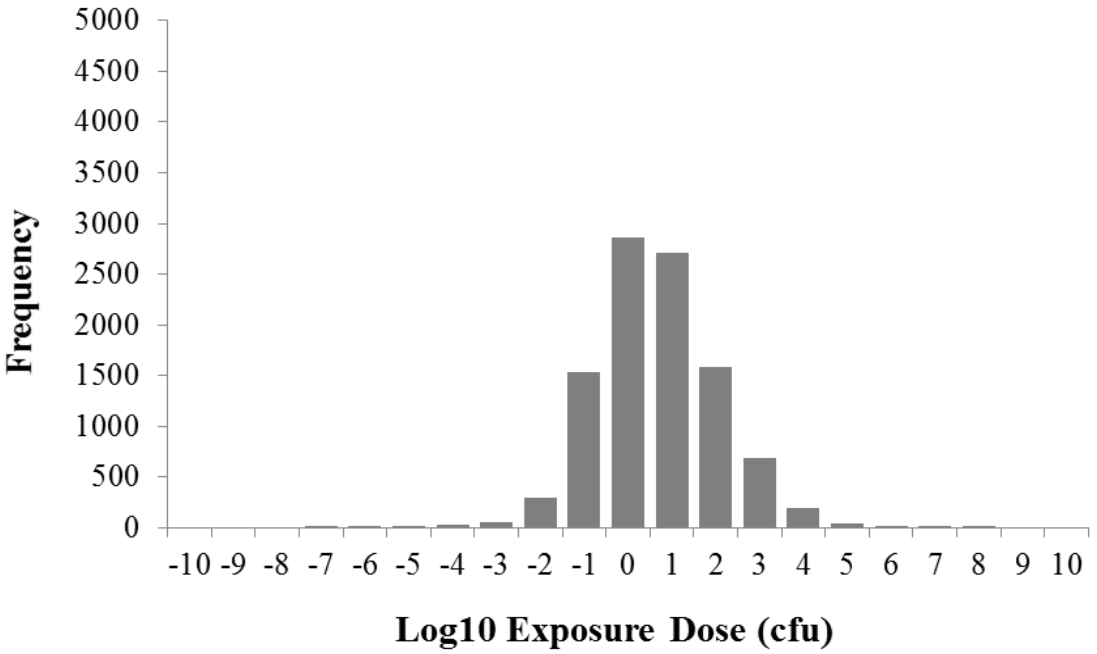

Figure 17: Exposure dose histogram for refrigerated salad treated by peeling

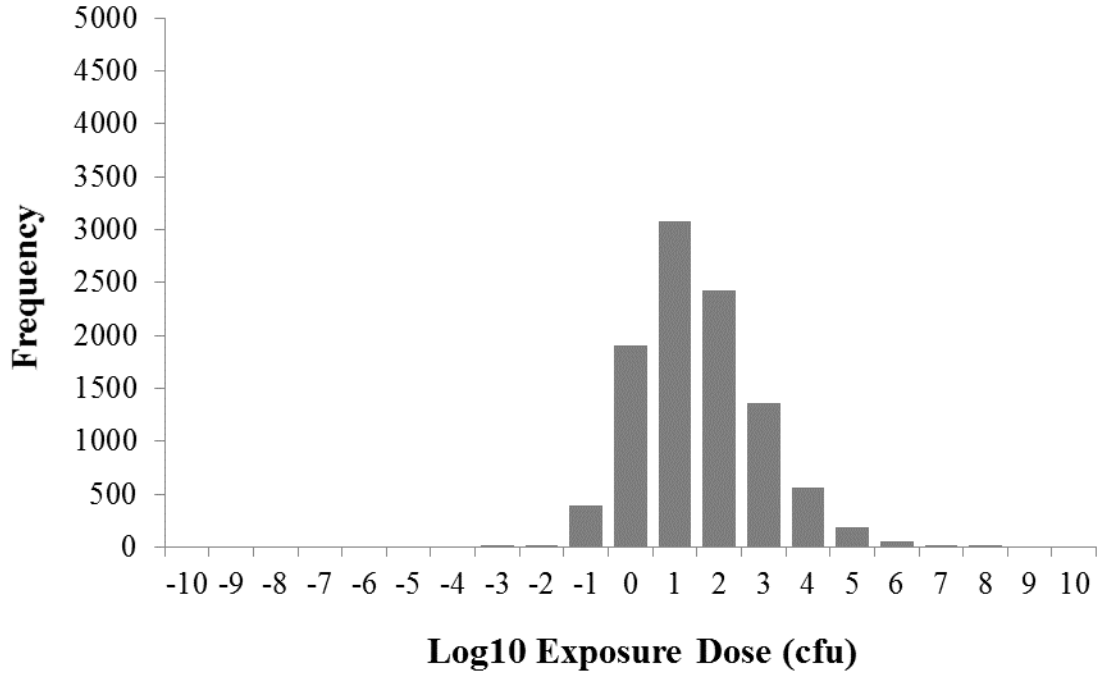

Figure 18: Exposure dose histogram for elevated temperature salad treated by peeling

Appendix D: *L. monocytogenes* Exposure Dose and Risk Distributions, Salad Without Lettuce

Table 20: Daily exposure dose (cfu) for *L. monocytogenes* in cucumber and tomato salad

Treatment	Storage Temp	5%	50%	95%
Control	Refrigerated	6.9E-01	2.8E01	3.8E02
	High	2.1	1.7E02	1.3E05
Sanitizer soak	Refrigerated	1.7E-02	7.8E-01	1.4E01
	High	4.6E-02	4.9	4.4E03
Tap water rinse	Refrigerated	2.3E-03	9.9E-01	1.6E01
	High	6.0E-02	6.2	5.4E03
Tap water soak	Refrigerated	1.3E-01	6.0	9.8E01
	High	3.4E-01	3.8E01	3.5E04
Flash blanch (rinse greens)	Refrigerated	7.9E-07	2.1E-04	3.9E-02
	High	2.7E-06	1.6E-03	3.5
Peel (rinse greens)	Refrigerated	2.5E-03	2.0E-01	1.2E01
	High	8.5E-03	1.5	1.7E03

Table 21: Risk per chemotherapy cycle for each risk management strategy and k approach, cucumber and tomato salad

Treatment	Storage Temp	*k* Approach	5%	50%	95%
Control	Refrigerated	1	6.4E-11	5.5E-09	2.9E-07
		2	1.3E-11	5.5E-09	5.0E-06
	High	1	2.2E-10	4.1E-08	5.2E-05
		2	4.6E-11	5.3E-08	3.2E-04
Sanitizer soak	Refrigerated	1	1.7E-12	1.6E-10	9.8E-09
		2	3.3E-13	1.6E-10	1.6E-07
	High	1	5.4E-12	1.2E-09	1.6E-06
		2	1.1E-12	1.6E-09	1.1E-05
Tap water rinse	Refrigerated	1	2.1E-12	1.9E-10	1.2E-08
		2	3.9E-13	2.0E-10	1.9E-07
	High	1	6.9E-12	1.5E-09	2.3E-06
		2	1.4E-12	2.0E-09	1.4E-05
Tap water soak	Refrigerated	1	1.2E-11	1.2E-09	7.0E-08
		2	2.4E-12	1.2E-09	1.2E-06
	High	1	3.9E-11	9.0E-09	1.4E-05
		2	8.0E-12	1.2E-08	8.2E-05
Surface blanch tomato and cucumber, rinse greens	Refrigerated	1	0.0	4.9E-14	1.5E-10
		2	0.0	4.5E-14	1.6E-11
	High	1	0.0	3.6E-13	1.1E-09
		2	0.0	4.4E-13	7.7E-09
Peel tomato and cucumber, rinse greens	Refrigerated	1	2.4E-13	4.3E-11	6.1E-09
		2	5.0E-14	4.9E-11	7.9E-08
	High	1	9.2E-13	3.4E-10	5.9E-07
		2	2.0E-13	4.4E-10	3.7E-06

REFERENCES

REFERENCES

1. Abadias, M., J. Usall, M. Anguera, C. Solson, and I. Vinas. 2008. Microbiological quality of fresh, minimally-processed fruit and vegetables, and sprouts from retail establishments. *Int. J. Food Microbiol*. 123:121-129.

2. Alexandre, E. M. C., T. R. S. Brandao, and C. L. M. Silva. 2012. Assessment of the impact of hydrogen peroxide solutions on microbial loads and quality factors of red bell peppers, strawberries and watercress. *Food Control*. 27:362-368.

3. Allard, S. M., M. T. Callahan, A. Bui, A. M. C. Ferelli, J. Chopyk, S. Chattopadhyay, E. F. Mongodin, S. A. Micallef, and A. R. Sapkota. 2019. Creek to table: Tracking fecal indicator bacteria, bacterial pathogens, and total bacterial communities from irrigation water to kale and radish crops. *Sci. Total Enviro*. 666:461-471.

4. Allerberger, F., and M. Wagner. 2010. Listeriosis: a resurgent foodborne infection. *Clin. Microbiol. Infect*. 16:16-23.

5. Anderson, R., I. Hay, H. A. Vanwyk, and A. Theron. 1983. Ascorbic acid in bronchial asthma. *S. Afr. Med. J*. 63:649-652.

6. Angelo, K. M., A. R. Conrad, A. Saupe, H. Dragoo, N. West, A. Sorenson, A. Barnes, M. Doyle, J. Beal, K. A. Jackson, S. Stroika, C. Tarr, Z. Kucerova, S. Lance, L. H. Gould, M. Wise, and B. R. Jackson. 2017. Multistate outbreak of *Listeria monocytogenes* infections linked to whole apples used in commercially produced, prepackaged caramel apples: United States, 2014–2015. *Epidemiol. Infect*. 145:848-856.

7. Arico, M., M. F. P. Boccalatte, D. Silvestri, E. Barisone, C. Messina, R. Chiesa, N. Santoro, P. Tamaro, A. Lippi, D. Gallisai, G. Basso, G. De Rossi, and Aieop. 2003. Osteonecrosis: an emerging complication of intensive chemotherapy for childhood acute lymphoblastic leukemia. *Haematologica*. 88:747-753.

8. Avritscher, E. B. C., C. D. Cooksley, and L. S. Elting. 2004. Scope and epidemiology of cancer therapy-induced oral and gastrointestinal mucositis. *Semin. Oncol. Nurs*. 20:3-10.

9. Bagci, U., and A. Temiz. 2011. Microbiological quality of fresh-squeezed orange juice and efficacy of fruit surface decontamination methods in microbiological quality. *J. Food Prot*. 74:1238-1244.

10. Bardsley, C. A., L. N. Truitt, R. C. Pfuntner, M. D. Danyluk, S. L. Rideout, and L. K. Strawn. 2019. Growth and survival of *Listeria monocytogenes* and *Salmonella* on whole and sliced cucumbers. *J. Food Prot*. 82:301-309.

11. Bavishi, C., and H. L. DuPont. 2011. Systematic review: the use of proton pump inhibitors and increased susceptibility to enteric infection. *Aliment. Pharmacol. Ther*. 34:1269-1281.

12. Bemrah, N., M. Sanaa, M. H. Cassin, M. W. Griffiths, and O. Cerf. 1998. Quantitative risk assessment of human listeriosis from consumption of soft cheese made from raw milk. *Prev. Vet. Med*. 37:129-145.

13. Beuchat, L. R. 1996. *Listeria monocytogenes*: incidence on vegetables. *Food Control*. 7:223-228.

14. Beuchat, L. R., and R. E. Brackett. 1990. Survival and growth of *Listeria monocytogenes* on lettuce as influenced by shredding, chlorine treatment, modified atmosphere packaging and temperature. *J. Food Sci*. 55:755-758.

15. Beuchat, L. R., and R. E. Brackett. 1991. Behavior of *Listeria monocytogenes* inoculated into raw tomatoes and processed tomato products. *Appl. Environ. Microbiol*. 57:1367-1371.

16. Beuchat, L. R., L. J. Harris, T. E. Ward, and T. M. Kajs. 2001. Development of a proposed standard method for assessing the efficacy of fresh produce sanitizers. *J. Food Prot*. 64:1103-1109.

17. Beuchat, L. R., B. V. Nail, B. B. Adler, and M. R. S. Clavero. 1998. Efficacy of spray application of chlorinated water in killing pathogenic bacteria on raw apples, tomatoes, and lettuce. *J. Food Prot*. 61:1305-1311.

18. Bhilwadikar, T., S. Pounraj, S. Manivannan, N. K. Rastogi, and P. S. Negi. 2019. Decontamination of microorganisms and pesticides from fresh fruits and vegetables: A comprehensive review from common household processes to modern techniques. *Compr. Rev. Food Sci. Food Saf.* 18:1003-1038.

19. Brackett, R. E. 1987. Antimicrobial effect of chlorine on *Listeria monocytogenes. J. Food Prot*. 50:999-1003.

20. Braun, L. E., H. D. Chen, and H. Frangoul. 2014. Significant inconsistency among pediatric oncologists in the use of the neutropenic diet. *Pediatr. Blood Cancer*. 61:1806-1810.

21. Breidt, F., J. S. Hayes, and H. P. Fleming. 2000. Reduction of microflora of whole pickling cucumbers by blanching. *J. Food Sci*. 65:1354-1358.

22. Buchanan, R. L., W. G. Damert, R. C. Whiting, and M. vanSchothorst. 1997. Use of epidemiologic and food survey data to estimate a purposefully conservative dose-response relationship for *Listeria monocytogenes* levels and incidence of listeriosis. *J. Food Prot*. 60:918-922.

23. Buchanan, R. L., S. G. Edelson, R. L. Miller, and G. M. Sapers. 1999. Contamination of intact apples after immersion in an aqueous environment containing *Escherichia coli* O157 : H7. *J. Food Prot*. 62:444-450.

24. Burnett, S. L., and L. R. Beuchat. 2002. Differentiation of viable and dead *Escherichia coli* O157 : H7 cells on and in apple structures and tissues following chlorine treatment. *J. Food Prot*. 65:251-259.

25. Carlin, F., and C. Nguyenthe. 1994. Fate of *Listeria monocytogenes* on four types of minimally processed green salads. *Lett. Appl. Microbiol*. 18:222-226.

26. Carr, A. C., and S. Maggini. 2017. Vitamin C and immune function. *Nutrients*. 9:E1211.

27. Carr, S. E., and V. Halliday. 2015. Investigating the use of the neutropenic diet: a survey of U.K. dietitians. *J. Hum. Nutr. Diet*. 28:510-515.

28. Carrasco, E., F. Perez-Rodriguez, A. Valero, R. M. Garcia-Gimeno, and G. Zurera. 2008. Growth of *Listeria monocytogenes* on shredded, ready-to-eat iceberg lettuce. *Food Control*. 19:487-494.

29. Carrasco, E., F. Perez-Rodriguez, A. Valero, R. M. Garcia-Gimeno, and G. Zurera. 2010. Risk assessment and management of *Listeria monocytogenes* in ready-to-eat lettuce salads. *Compr. Rev. Food Sci. Food Saf*. 9:498-512.

30. Carter, L. W. 1993. Influences of nutrition and stress on people at risk for neutropenia: Nursing implications. *Oncol. Nurs. Forum*. 20:1241-1250.

31. Carter, L. W. 1994. Bacterial translocation: nursing implications in the care of patients with neutropenia. *Oncol. Nurs. Forum*. 21:857-65.

32. Centers for Disease Control and Prevention. 2015. National *Listeria* surveillance annual summary, 2014. Available at: https://www.cdc.gov/nationalsurveillance/pdfs/listeria-annual-summary-2014-508.pdf. Accessed 26 July, 2020.

33. Centers for Disease Control and Prevention. 2016. Multistate outbreak of listeriosis linked to packaged salads at Springfield, Ohio Dole processing facility (final update). Available at: https://www.cdc.gov/listeria/outbreaks/bagged-salads-01-16/index.html. Accessed 23 July, 2020.

34. Centers for Disease Control and Prevention. 2019. Information for health care providers. Available at: https://www.cdc.gov/cancer/preventinfections/providers.htm#:~:text=Each%20year%2C%20about%20650%2C000%20cancer,clinic%20in%20the%20United%20States. Accessed 5 June, 2020.

35. Centers for Disease Control and Prevention. 2019. *Listeria* (Listeriosis). Available at: https://www.cdc.gov/listeria/index.html. Accessed 26 July, 2020.

36. Ceylan, E., W. McMahon, and D. M. Garren. 2017. Thermal inactivation of *Listeria monocytogenes* and *Salmonella* during water and steam blanching of vegetables. *J. Food Prot*. 80:1550-1556.

37. Chen, Y. H., W. H. Ross, M. J. Gray, M. Wiedmann, R. C. Whiting, and V. N. Scott. 2006. Attributing risk to *Listeria monocytogenes* subgroups: Dose response in relation to genetic lineages. *J. Food Prot*. 69:335-344.

38. Choi, M. H., Y. J. Park, M. Kim, Y. H. Seo, Y. A. Kim, J. Y. Choi, D. Yong, S. H. Jeong, and K. Lee. 2018. Increasing incidence of listeriosis and infection-associated clinical outcomes. *Ann. Lab. Med*. 38:102-109.

39. Cobb, C. A., G. D. Curtis, D. S. Bansi, E. Slade, W. Mehal, R. G. Mitchell, and R. W. Chapman. 1996. Increased prevalence of *Listeria monocytogenes* in the faeces of patients receiving long-term H2-antagonists. *Eur. J. Gastroenterol. Hepatol*. 8:1071-1074.

40. Cokes, C., A. M. France, V. Reddy, H. Hanson, L. Lee, L. Kornstein, F. Stavinsky, and S. Balter. 2011. Serving high-risk foods in a high-risk setting: Survey of hospital food service practices after an outbreak of listeriosis in a hospital. *Infect. Control Hosp. Epidemiol*. 32:380-386.

41. Cotter, P. D., C. Stanton, R. P. Ross, and C. Hill. 2012. The impact of antibiotics on the gut microbiota as revealed by high throughput DNA sequencing. *Discov. Med*. 13:193-199.

42. Crawford, J., D. C. Dale, and G. H. Lyman. 2004. Chemotherapy-induced neutropenia: Risks, consequences, and new directions for its management. *Cancer*. 100:228-237.

43. de Oliveira, M. A., E. G. A. Ribeiro, A. M. M. Bergamini, and E. C. P. De Martinis. 2010. Quantification of *Listeria monocytogenes* in minimally processed leafy vegetables using a combined method based on enrichment and 16S rRNA real-time PCR. *Food Microbiol*. 27:19-23.

44. Deitch, E. A. 1994. Bacterial translocation: The influence of dietary variables. *Gut*. 35:S23-S27.

45. Delaquis, P., S. Stewart, S. Cazaux, and P. Toivonen. 2002. Survival and growth of *Listeria monocytogenes* and *Escherichia coli* O157:H7 in ready-to-eat iceberg lettuce washed in warm chlorinated water. *J. Food Prot*. 65:459-464.

46. DeMille, D., P. Deming, P. Lupinacci, and L. A. Jacobs. 2006. The effect of the neutropenic diet in the outpatient setting: A pilot study. *Oncol. Nurs. Forum*. 33:337-343.

47. Deza, M. A., M. Araujo, and M. J. Garrido. 2003. Inactivation of *Escherichia coli* O157:H7, *Salmonella enteritidis* and *Listeria monocytogenes* on the surface of tomatoes by neutral electrolyzed water. *Lett. Appl. Microbiol*. 37:482-487.

48. Dhokane, V. S., S. Hajare, R. Shashidhar, A. Sharma, and J. R. Bandekar. 2006. Radiation processing to ensure safety of minimally processed carrot (*Daucus carota*) and cucumber (*Cucumis sativus*): Optimization of dose for the elimination of *Salmonella Typhimurium* and *Listeria monocytogenes. J. Food Prot*. 69:444-448.

49. Ding, T., J. Iwahori, F. Kasuga, J. Wang, F. Forghani, M. S. Park, and D. H. Oh. 2013. Risk assessment for *Listeria monocytogenes* on lettuce from farm to table in Korea. *Food Control*. 30:190-199.

50. Elting, L. S., C. Cooksley, M. Chambers, S. B. Cantor, E. Manzullo, and E. B. Rubenstein. 2003. The burdens of cancer therapy. *Cancer*. 98:1531-1539.

51. Erickson, M. C. 2010. Microbial risks associated with cabbage, carrots, celery, onions, and deli salads made with these produce items. *Compr. Rev.Food Sci. Food Saf*. 9:602-619.

52. Erickson, M. C. 2012. Internalization of fresh produce by foodborne pathogens. *Annu. Rev. Food Sci. Technol*. 3:283-310.

53. Erickson, M. C., J. Liao, J. L. Cannon, and Y. R. Ortega. 2015. Role of brushes and peelers in removal of *Escherichia coli* O157:H7 and *Salmonella* from produce in domestic kitchens. *J. Food Prot.* 78:1624-1631.

54. Escalona, V. H., E. Aguayo, G. B. Martinez-Hernandez, and F. Artes. 2010. UV-C doses to reduce pathogen and spoilage bacterial growth in vitro and in baby spinach. *Postharvest Biol. Tec*. 56:223-231.

55. Ettefagh, L., H. Paden, I. Hatsu, K. Kane, M. Dogocu, A. Diaz, A. Bhatt, A. Beery, and S. Ilic. 2018. Factors associated with food safety behaviors in cancer patients seeking treatment. International Association for Food Protection Annual Meeting, Salt Lake City, Utah.

56. Faith, N., S. Kathariou, R. Sahaghian, J. Luchansky, and C. Czuprynski. 2009. Virulence for mice, resistance to synthetic gastric fluid, and biofilm formation of L*isteria monocytogenes* H7550, a serotype 4b strain isolated from frankfurters associated with the BilMar listeriosis outbreak. International Association for Food Protection Annual Meeting, Grapevine, Texas.

57. Fan, X., B. A. Annous, J. C. Beaulieu, and J. E. Sites. 2008. Effect of hot water surface pasteurization of whole fruit on shelf life and quality of fresh-cut cantaloupe. *J. Food Sci*. 73:M91-M98.

58. Farber, J. M., and P. I. Peterkin. 1991. *Listeria monocytogenes*, a foodborne pathogen. *Microbiol. Rev*. 55:476-511.

59. Farber, J. M., W. H. Ross, and J. Harwig. 1996. Health risk assessment of *Listeria monocytogenes* in Canada. *Int. J. Food Microbiol*. 30:145-156.

60. Farber, J. M., S. L. Wang, Y. Cai, and S. Zhang. 1998. Changes in populations of *Listeria monocytogenes* inoculated on packaged fresh-cut vegetables. *J. Food Prot*. 61:192-195.

61. Fenlon, D. R., J. Wilson, and W. Donachie. 1996. The incidence and level of *Listeria monocytogenes* contamination of food sources at primary production and initial processing. *J. Appl. Bacteriol*. 81:641-650.

62. Fishburn, J. D., Y. Tang, and J. F. Frank. 2012. Efficacy of various consumer-friendly produce washing technologies in reducing pathogens on fresh produce. *Food Prot. Trends*. 32:456-466.

63. Fox, N., and A. G. Freifeld. 2012. The neutropenic diet reviewed: Moving toward a safe food handling approach. *Oncology (Williston Park, N.Y.)*. 26:572-585.

64. Francis, G. A., and D. O'Beirne. 2002. Effects of vegetable type and antimicrobial dipping on survival and growth of *Listeria innocua* and *E-coli. Int. J. Food Sci. Technol*. 37:711-718.

65. Francis, G. A., and D. O'Beirne. 2006. Isolation and pulsed-field gel electrophoresis typing of *Listeria monocytogenes* from modified atmosphere packaged fresh-cut vegetables collected in Ireland. *J. Food Prot*. 69:2524-2528.

66. Freeman, J. H., E. J. McAvoy, N. S. Boyd, R. Kanissery, H. A. Smith, J. Desaeger, and J. W. Noling. 2019. Tomato production, p. 349-392. *In* Vegetable Production Handbook of Florida, 2019-2020.

67. Freifeld, A. G., E. J. Bow, K. A. Sepkowitz, M. J. Boeckh, J. I. Ito, C. A. Mullen, Raad, II, K. V. Rolston, J. A. H. Young, and J. R. Wingard. 2011. Clinical practice guideline for the use of antimicrobial agents in neutropenic patients with cancer: 2010 update by the Infectious Diseases Society of America. *Clin. Infect. Dis*. 52:E56-E93.

68. French, M. R., R. Levy-Milne, and D. Zibrik. 2001. A survey of the use of low microbial diets in pediatric bone marrow transplant programs. *J. Am. Diet. Assoc*. 101:1194-1198.

69. Gaul, L. K., N. H. Farag, T. Shim, M. A. Kingsley, B. J. Silk, and E. Hyytia-Trees. 2013. Hospital-acquired listeriosis outbreak caused by contaminated diced celery-Texas, 2010. *Clin. Infect. Dis*. 56:20-26.

70. Gillespie, I. A., J. McLauchlin, K. A. Grant, C. L. Little, V. Mithani, C. Penman, C. Lane, and M. Regan. 2006. Changing pattern of human listeriosis, England and Wales, 2001-2004. *Emerg. Infect. Dis*. 12:1361-1366.

71. Gillespie, I. A., J. McLauchlin, C. L. Little, C. Penman, P. Mook, K. Grant, and S. J. O'Brien. 2009. Disease presentation in relation to infection foci for non-pregnancy-aassociated human listeriosis in England and Wales, 2001 to 2007. *J. Clin. Microbiol*. 47:3301-3307.

72. Goldschmidt, M. C. 1991. Reduced bactericidal activity in neutrophils from scorbutic animals and the effect of ascorbic acid on these target bacteria in vivo and in vitro. *Am. J. Clin. Nutr*. 54:1214S-1220S.

73. Golnazarian, C. A., C. W. Donnelly, S. J. Pintauro, and D. B. Howard. 1989. Comparison of infectious dose of *Listeria monocytogenes* F5817 as determined for normal versus compromised C57B1/6J mice. *J. Food Prot*. 52:696-701.

74. Gombas, D. E., Y. H. Chen, R. S. Clavero, and V. N. Scott. 2003. Survey of *Listeria monocytogenes* in ready-to-eat foods. *J. Food Prot*. 66:559-569.

75. Gomez, C., B. P. Marks, and E. T. Ryser. 2020. Kitchen-scale treatments for reduction of *Listeria monocytogenes in* prepared produce for immunocompromised populations. Submitted to *J. Food Prot*.

76. Gomez-Conde, M. S., J. Garcia, S. Chamorro, P. Eiras, P. G. Rebollar, A. P. de Rozas, I. Badiola, C. de Blas, and R. Carabano. 2007. Neutral detergent-soluble fiber improves gut barrier function in twenty-five-day-old weaned rabbits. *J. Anim. Sci*. 85:3313-3321.

77. Goulet, V., M. Hebert, C. Hedberg, E. Laurent, V. Vaillant, H. De Valk, and J. C. Desenclos. 2012. Incidence of listeriosis and related mortality among groups at risk of acquiring listeriosis. *Clin. Infect. Dis*. 54:652-660.

78. Guevara, R. E., L. Mascola, and F. Sorvillo. 2009. Risk factors for mortality among patients with nonperinatal listeriosis in Los Angeles County, 1992-2004. *Clin. Infect. Dis*. 48:1507-1515.

79. Haas, C. N., J. B. Rose, and C. P. Gerba. 2014. Quantitative microbial risk assessment. Wiley-Blackwell, New York.

80. Hadjilouka, A., K. S. Mantzourani, A. Katsarou, M. Cavaiuolo, A. Ferrante, S. Paramithiotis, M. Mataragas, and E. H. Drosinos. 2015. Estimation of *Listeria monocytogenes* and *Escherichia coli* O157:H7 prevalence and levels in naturally contaminated rocket and cucumber samples by deterministic and stochastic approaches. *J. Food Prot*. 78:311-322.

81. Harris, L. J., L. R. Beuchat, T. M. Kajs, T. E. Ward, and C. H. Taylor. 2001. Efficacy and reproducibility of a produce wash in killing *Salmonella* on the surface of tomatoes assessed with a proposed standard method for produce sanitizers. *J. Food Prot*. 64:1477-82.

82. Harris, L. J., J. N. Farber, L. R. Beuchat, M. E. Parish, T. V. Suslow, E. H. Garrett, and F. F. Busta. 2003. Outbreaks associated with fresh produce: Incidence, growth, and survival of pathogens in fresh and fresh-cut produce. *Compr. Rev. Food Sci. Food Saf*. 2:78-141.

83. Heng, M. S., J. B. Gauro, A. Yaxley, and J. Thomas. Does a neutropenic diet reduce adverse outcomes in patients undergoing chemotherapy? *Eur. J. Cancer Care*. 29:e13155.

84. Herrador, Z., A. Gherasim, R. Lopez-Velez, and A. Benito. 2019. Listeriosis in Spain based on hospitalisation records, 1997 to 2015: Need for greater awareness. *Eurosurveillance*. 24:22-31.

85. Ho, J. L., K. N. Shands, G. Friedland, P. Eckind, and D. W. Fraser. 1986. An outbreak of type 4b Listeria monocytogenes infection involving patients from eight Boston hospitals. *JAMA Intern. Med*. 146:520-524.

86. Hoelzer, K., R. Pouillot, and S. Dennis. 2012. *Listeria monocytogenes* growth dynamics on produce: A review of the available data for predictive modeling. *Foodborne Pathog. Dis*. 9:661-673.

87. Ilic, S. 2014. Block 2014 food frequency questionairre. Personal communication, January 9, 2019. Contact email address: ilic.2@osu.edu.

88. Interagency Retail *Listeria monocytogenes* Risk Assessment Workgroup. 2013. Interagency risk assessment: *Listeria monocytogenes* in retail delicatessens, technical report. Available at: https://www.fsis.usda.gov/wps/wcm/connect/c0c6dfbc-ad83-47c1-bcb8-8db6583f762b/Lm-Retail-Technical-Report.pdf?MOD=AJPERES. Accessed 26 July, 2020.

89. Jacxsens, L., F. Devlieghere, and J. Debevere. 2002. Temperature dependence of shelf-life as affected by microbial proliferation and sensory quality of equilibrium modified atmosphere packaged fresh produce. *Postharvest Biol. Tec*. 26:59-73.

90. Jensen, D. A., L. M. Friedrich, L. J. Harris, M. D. Danyluk, and D. W. Schaffner. 2015. Cross contamination of *Escherichia coli* O157:H7 between lettuce and wash water during home-scale washing. *Food Microbiol*. 46:428-433.

91. Johnsen, B. O., E. Lingaas, D. Torfoss, E. H. Strom, and I. Nordoy. 2010. A large outbreak of *Listeria monocytogenes* infection with short incubation period in a tertiary care hospital. *J. Infect*. 61:465-470.

92. Joint Food and Agriculture Organization/World Health Organization Expert Consultation. 1995. Application of risk analysis to food standards issues. Available at: https://apps.who.int/iris/bitstream/handle/10665/58913/WHO_FNU_FOS_95.3.pdf?sequence=1&isAllowed=y. Accessed 26 July, 2020.

93. Kabelitz, T., B. Schmidt, W. B. Herppich, and K. Hassenberg. 2019. Effects of hot water dipping on apple heat transfer and post-harvest fruit quality. *LWT-Food Sci. Technol*. 108:416-420.

94. Kakiomenou, K., C. Tassou, and G. J. Nychas. 1998. Survival of *Salmonella enteritidis* and *Listeria monocytogenes* on salad vegetables. *World J. Microbiol. Biotechnol*. 14:383-387.

95. Kaminski, C. N., G. R. Davidson, and E. T. Ryser. 2014. *Listeria monocytogenes* transfer during mechanical dicing of celery and growth during subsequent storage. *J. Food Prot*. 77:765-771.

96. Kathariou, S., P. Metz, H. Hof, and W. Goebel. 1987. Tn916-induced mutations in the hemolysin determinant affecting virulence of *Listeria monocytogenes. J. Bacteriol*. 169:1291-7.

97. Khorana, A. A., N. M. Kuderer, E. Culakova, G. H. Lyman, and C. W. Francis. 2008. Development and validation of a predictive model for chemotherapy-associated thrombosis. *Blood*. 111:4902-4907.

98. Kilonzo-Nthenge, A., F. C. Chen, and S. L. Godwin. 2006. Efficacy of home washing methods in controlling surface microbial contamination on fresh produce. *J. Food Prot*. 69:330-334.

99. Kleinhendler, E., M. J. Cohen, A. E. Moses, O. Paltiel, J. Strahilevitz, and A. Cahan. 2018. Empiric antibiotic protocols for cancer patients with neutropenia: A single–center study of treatment efficacy and mortality in patients with bacteremia. *Int. J. Antimicrob. Agents*. 51:71-76.

100. Koseki, S., and S. Isobe. 2005. Growth of *Listeria monocytogenes* on iceberg lettuce and solid media. *Int. J. Food Microbiol*. 101:217-225.

101. Kovacevic, M., J. Burazin, H. Pavlovic, M. Kopjar, and V. Pilizota. 2013. Prevalence and level of *Listeria monocytogenes* and other *Listeria sp.* in ready-to-eat minimally processed and refrigerated vegetables. *World J. Microbiol. Biotechnol*. 29:707-712.

102. Lang, M. M., L. J. Harris, and L. R. Beuchat. 2004. Survival and recovery of *Escherichia coli* O157:H7, *Salmonella*, and *Listeria monocytogenes* on lettuce and parsley as affected by method of inoculation, time between inoculation and analysis, and treatment with chlorinated water. *J. Food Prot*. 67:1092-1103.

103. Lassiter, M., and S. M. Schneider. 2015. A pilot study comparing the neutropenic diet to a non-neutropenic diet in the allogeneic hematopoietic stem cell transplantation population. *Clin. J. Oncol. Nurs*. 19:273-8.

104. Lecuit, M., S. Dramsi, C. Gottardi, M. Fedor-Chaiken, B. Gumbiner, and P. Cossart. 1999. A single amino acid in E-cadherin responsible for host specificity towards the human pathogen *Listeria monocytogenes. Embo. J*. 18:3956-3963.

105. Lee, C. Y., H. C. Tsai, C. M. Kunin, S. S. Lee, K. S. Wu, and Y. S. Chen. 2014. Emergence of sporadic non-clustered cases of hospital-associated listeriosis among immunocompromised adults in southern Taiwan from 1992 to 2013: Effect of precipitating immunosuppressive agents. *BMC Infect. Dis*. 14:145-152

106. Leung, K., K. Hope, and V. Sheppeard. 2018. A review of listeriosis notifications and co-existing conditions in New South Wales, 2010-2015. *Commun. Dis. Intell*. 42:S2209-6051

107. Levy, R., O. Shriker, A. Porath, K. Riesenberg, and F. Schlaeffer. 1996. Vitamin C for the treatment of recurrent furunculosis in patients with impaired neutrophil functions. *J. Infect Dis*. 173:1502-1505.

108. Li-Cohen, A. E., and C. M. Bruhn. 2002. Safety of consumer handling of fresh produce from the time of purchase to the plate: a comprehensive consumer survey. *J. Food Prot*. 65:1287-1296.

109. Liao, C. H. 2007. Inhibition of foodborne pathogens by native microflora recovered from fresh peeled baby carrot and propagated in cultures. *J. Food Sci*. 72:M134-M139.

110. Likotrafiti, E., P. Smirniotis, A. Nastou, and J. Rhoades. 2013. Effect of relative humidity and storage temperature on the behavior of *Listeria monocytogene*s on fresh vegetables. *J. Food Safety*. 33:545-551.

111. Lindqvist, R., and A. Westoo. 2000. Quantitative risk assessment for *Listeria monocytogenes* in smoked or gravad salmon and rainbow trout in Sweden. *Int. J. Food Microbiol*. 58:181-196.

112. Little, C., D. Roberts, E. Youngs, and J. De Louvois. 1999. Microbiological quality of retail imported unprepared whole lettuces: A PHLS food working group study. *J. Food Prot*. 62:325-328.

113. Lopez, K., K. Phalen, C. I. Vahl, K. R. Roberts, and K. J. K. Getty. 2016. Investigation of handling practices for fresh produce and the efficacy of commercially available produce washes on removal of pathogens and natural microflora on whole cantaloupe surfaces. *Food Control*. 68:251-259.

114. Losikoff, M. B. 1990. The efficacy of a boiling water blanch on the inactivation of *Listeria monocytogenes* in diced celery. Master of Science in Food Science, University of Maryland.

115. Lukasik, J., M. L. Bradley, T. M. Scott, M. Dea, A. Koo, W. Y. Hsu, J. A. Bartz, and S. R. Farrah. 2003. Reduction of poliovirus 1, bacteriophages, *Salmonella* Montevideo, and *Escherichia coli* O157:H7 on strawberries by physical and disinfectant washes. *J. Food Prot*. 66:188-193.

116. Lynch, S. R., and J. D. Cook. 1980. Interaction of vitamin C and iron. *Ann. NY Acad. Sci*. 355:32-44.

117. Lyytikainen, O., T. Autio, R. Maijala, P. Ruutu, T. Honkanen-Buzalski, M. Miettinen, M. Hatakka, J. Mikkola, V. J. Anttila, T. Johansson, L. Rantala, T. Aalto, H. Korkeala, and A. Siitonen. 2000. An outbreak of *Listeria monocytogenes* serotype 3a infections from butter in Finland. *J. Infect. Dis*. 181:1838-1841.

118. Mansur, A. R., and D. H. Oh. 2015. Combined effects of thermosonication and slightly acidic electrolyzed water on the microbial quality and shelf life extension of fresh-cut kale during refrigeration storage. *Food Microbiol*. 51:154-162.

119. Marik, C. M., J. Zuchel, D. W. Schaffner, and L. K. Strawn. 2020. Growth and survival of *Listeria monocytogenes* on intact fruit and vegetable surfaces during postharvest handling: A systematic literature review. *J. Food Prot*. 83:108-128.

120. Martinsen, T. C., K. Bergh, and H. L. Waldum. 2005. Gastric juice: A barrier against infectious diseases. *Basic Clin. Pharmacol. Toxicol*. 96:94-102.

121. Mazzotta, A. S. 2001. Heat resistance of *Listeria monocytogenes* in vegetables: Evaluation of blanching processes. *J. Food Prot*. 64:385-387.

122. McCollum, J. T., A. B. Cronquist, B. J. Silk, K. A. Jackson, K. A. O'Connor, S. Cosgrove, J. P. Gossack, S. S. Parachini, N. S. Jain, P. Ettestad, M. Ibraheem, V. Cantu, M. Joshi, T. DuVernoy, N. W. Fogg, J. R. Gorny, K. M. Mogen, C. Spires, P. Teitell, L. A. Joseph, C. L. Tarr, M. Imanishi, K. P. Neil, R. V. Tauxe, and B. E. Mahon. 2013. Multistate outbreak of listeriosis associated with cantaloupe. *N. Engl. J. Med*. 369:944-953.

123. McGrath, P. 2002. Reflections on nutritional issues associated with cancer therapy. *Cancer Prac*. 10:94-101.

124. Mohammed, B. M., B. J. Fisher, D. Kraskauskas, D. Farkas, D. F. Brophy, A. A. Fowler, 3rd, and R. Natarajan. 2013. Vitamin C: A novel regulator of neutrophil extracellular trap formation. *Nutrients*. 5:3131-3151.

125. Moliterno, A. R., and J. L. Spivak. 1996. Anemia of cancer. *Hematol. Oncol. Clin. North Am.* 10:345-362.

126. Montassier, E., T. Gastinne, P. Vangay, G. A. Al-Ghalith, S. B. des Varannes, S. Massart, P. Moreau, G. Potel, M. F. de La Cochetiere, E. Batard, and D. Knights. 2015. Chemotherapy-driven dysbiosis in the intestinal microbiome. *Aliment. Pharmacol. Ther*. 42:515-528.

127. Monu, E. A., M. Valladares, D. H. D'Souza, and P. M. Davidson. 2015. Determination of the thermal inactivation kinetics of *Listeria monocytogenes*, *Salmonella enterica*, and *Escherichia coli* O157:H7 and non-O157 in buffer and a spinach homogenate. *J. Food Prot*. 78:1467-1471.

128. Moody, K., M. Meyer, C. A. Mancuso, M. Charlson, and L. Robbins. 2006. Exploring concerns of children with cancer. *Support. Care Cancer*. 14:960-966.

129. Moody, K. M., R. A. Baker, R. O. Santizo, I. Olmez, J. M. Spies, A. Buthmann, L. Granowetter, R. Y. Dulman, K. Ayyanar, J. B. Gill, and A. E. Carroll. 2018. A randomized trial of the effectiveness of the neutropenic diet versus food safety guidelines on infection rate in pediatric oncology patients. *Pediatr. Blood Cancer*. 65.

130. Mook, P., S. J. O'Brien, and I. A. Gillespie. 2011. Concurrent conditions and human listeriosis, England, 1999-2009. *Emerg. Infect. Dis*. 17:38-43.

131. Nastou, A., J. Rhoades, P. Smirniotis, I. Makri, M. Kontominas, and E. Likotrafiti. 2012. Efficacy of household washing treatments for the control of *Listeria monocytogenes* on salad vegetables. *Int. J.Food Microbiol*. 159:247-253.

132. Nguyen-the, C., and F. Carlin. 1994. The microbiology of minimally processed fresh fruits and vegetables. *Crit. Rev. Food Sci. Nutr*. 34:371-401.

133. Nousiainen, L. L., S. Joutsen, J. Lunden, M. L. Hanninen, and M. Fredriksson-Ahomaa. 2016. Bacterial quality and safety of packaged fresh leafy vegetables at the retail level in Finland. *Int. J. Food Microbiol*. 232:73-79.

134. Olaimat, A. N., and R. A. Holley. 2012. Factors influencing the microbial safety of fresh produce: A review. *Food Microbiol*. 32:1-19.

135. Omac, B., R. G. Moreira, and E. Castell-Perez. 2018. Quantifying growth of cold-adapted *Listeria monocytogenes* and *Listeria innocua* on fresh spinach leaves at refrigeration temperatures. *J. Food Eng*. 224:17-26.

136. Omac, B., R. G. Moreira, A. Castillo, and E. Castell-Perez. 2015. Growth of *Listeria monocytogenes* and *Listeria innocua* on fresh baby spinach leaves: Effect of storage temperature and natural microflora. *Postharvest Biol. Technol*. 100:41-51.

137. Painter, J. A., R. M. Hoekstra, T. Ayers, R. V. Tauxe, C. R. Braden, F. J. Angulo, and P. M. Griffin. 2013. Attribution of foodborne illnesses, hospitalizations, and deaths to food commodities by using outbreak data, United States, 1998-2008. *Emerg. Infect. Dis*. 19:407-415.

138. Park, E. J., E. Alexander, G. A. Taylor, R. Costa, and D. H. Kang. 2009. The decontaminative effects of acidic electrolyzed water for *Escherichia coli* O157:H7, *Salmonella typhimurium*, and *Listeria monocytogenes* on green onions and tomatoes with differing organic demands. *Food Microbiol*. 26:386-390.

139. Park, E. J., P. M. Gray, S. W. Oh, J. Kronenberg, and D. H. Kang. 2008. Efficacy of FIT produce wash and chlorine dioxide on pathogen control in fresh potatoes. *J. Food Sci*. 73:M278-M282.

140. Park, S. H., M. R. Choi, J. W. Park, K. H. Park, M. S. Chung, S. Ryu, and D. H. Kang. 2011. Use of organic acids to inactivate *Escherichia coli* O157:H7, *Salmonella typhimurium*, and *Listeria monocytogenes* on organic fresh apples and lettuce. *J. Food Sci*. 76:M293-M298.

141. Parnell, T. L., L. J. Harris, and T. V. Suslow. 2005. Reducing Salmonella on cantaloupes and honeydew melons using wash practices applicable to postharvest handling, foodservice, and consumer preparation. *Int. J. Food Microbiol*. 99:59-70.

142. Pezzuto, A., S. Belluco, C. Losasso, I. Patuzzi, P. Bordin, A. Piovesana, D. Comin, R. Mioni, and A. Ricci. 2016. Effectiveness of washing procedures in reducing *Salmonella*

enterica and *Listeria monocytogenes* on a raw leafy green vegetable (*Eruca vesicaria*). *Front. Microbiol*. 7.

143. Pinner, R. W., A. Schuchat, B. Swaminathan, P. S. Hayes, K. A. Deaver, R. E. Weaver, B. D. Plikaytis, M. Reeves, C. V. Broome, and J. D. Wenger. 1992. Role of foods in sporadic listeriosis II: Microbiologic and epidemiologic investigation. *JAMA*. 267:2046-2050.

144. Poimenidou, S. V., D. N. Chatzithoma, G. J. Nychas, and P. N. Skandamis. 2016. Adaptive response of *Listeria monocytogenes* to heat, salinity and low pH, after habituation on cherry tomatoes and lettuce leaves. *PloS One*. 11.

145. Pouillot, R., V. Goulet, M. L. Delignette-Muller, A. Mahe, and M. Cornu. 2009. Quantitative risk assessment of *Listeria monocytogenes* in French cold-smoked salmon: II. Risk characterization. *Risk Anal*. 29:806-819.

146. Pouillot, R., K. Hoelzer, Y. H. Chen, and S. B. Dennis. 2015. *Listeria monocytogenes* dose response revisited-Incorporating adjustments for variability in strain virulence and host susceptibility. *Risk Anal*. 35:90-108.

147. Prazak, A. M., E. A. Murano, I. Mercado, and G. R. Acuff. 2002. Prevalence of *Listeria monocytogenes* during production and postharvest processing of cabbage. *J. Food Prot*. 65:1728-1734.

148. Rahman, A., D. Munther, A. Fazil, B. Smith, and J. H. Wu. 2018. Advancing risk assessment: mechanistic dose-response modelling of *Listeria monocytogenes* infection in human populations. *R. Soc. Open Sci*. 5:180343.

149. Rodgers, S. L., J. N. Cash, M. Siddiq, and E. T. Ryser. 2004. A comparison of different chemical sanitizers for inactivating *Escherichia coli* O157 : H7 and *Listeria monocytogenes* in solution and on apples, lettuce, strawberries, and cantaloupe. *J. Food Prot*. 67:721-731.

150. Ross, T., S. Rasmussen, A. Fazil, G. Paoli, and J. Sumner. 2009. Quantitative risk assessment of Listeria monocyrogenes in ready-to-eat meats in Australia. *Int. J. Food Microbiol*. 131:128-137.

151. Sagoo, S. K., C. L. Little, and R. T. Mitchell. 2003. Microbiological quality of open ready-to-eat salad vegetables: Effectiveness of food hygiene training of management. *J. Food Prot*. 66:1581-1586.

152. Sanaa, M., L. Coroller, and O. Cerf. 2004. Risk assessment of listeriosis linked to the consumption of two soft cheeses made from raw milk: Camembert of Normandy and Brie of Meaux. *Risk Anal*. 24:389-399.

153. Sant'Ana, A. S., M. C. Igarashi, M. Landgraf, M. T. Destro, and B. Franco. 2012. Prevalence, populations and pheno- and genotypic characteristics of *Listeria*

monocytogenes isolated from ready-to-eat vegetables marketed in Sao Paulo, Brazil. *Int. J. Food Microbiol*. 155:1-9.

154. Santos, M. I., A. Cavaco, J. Gouveia, M. R. Novais, P. J. Nogueira, L. Pedroso, and M. Ferreira. 2012. Evaluation of minimally processed salads commercialized in Portugal. *Food Control*. 23:275-281.

155. Sapers, G. M. 2001. Efficacy of washing and sanitizing methods for disinfection of fresh fruit and vegetable products. *Food Technol.Biotechnol*. 39:305-311.

156. Sapers, G. M., R. L. Miller, M. Jantschke, and A. M. Mattrazzo. 2000. Factors limiting the efficacy of hydrogen peroxide washes for decontamination of apples containing *Escherichia coli. J. Food Sci*. 65:529-532.

157. Selman, J. D., and E. J. Rolfe. 1982. Effects of water blanching on pea seeds, changes in vitamin C content. *J. Food Technol*. 17:219-234.

158. Seo, K. H., and J. F. Frank. 1999. Attachment of *Escherichia coli* O157:H7 to lettuce leaf surface and bacterial viability in response to chlorine treatment as demonstrated by using confocal scanning laser microscopy. *J. Food Prot*. 62:3-9.

159. Shamshiri, R. R., J. W. Jones, K. R. Thorp, D. Ahmad, H. C. Man, and S. Taheri. 2018. Review of optimum temperature, humidity, and vapour pressure deficit for microclimate evaluation and control in greenhouse cultivation of tomato: a review. *Int. Agrophys*. 32:287-302.

160. Shetty, A., J. McLauchlin, K. Grant, D. O'Brien, T. Howard, and E. M. Davies. 2009. Outbreak of *Listeria monocytogenes* in an oncology unit associated with sandwiches consumed in hospital. *J. Hosp. Infect*. 72:332-336.

161. Siegman-Igra, Y., R. Levin, M. Weinberger, Y. Golan, D. Schwartz, Z. Samra, H. Konigsberger, A. Yinnon, G. Rahav, N. Keller, N. Bisharat, J. Karpuch, R. Finkelstein, M. Alkan, Z. Landau, J. Novikov, D. Hassin, C. Rudnicki, R. Kitzes, S. Ovadia, Z. Shimoni, R. Lang, and T. Shohat. 2002. *Listeria monocytogenes* infection in Israel and review of cases worldwide. *Emerg. Infect. Dis*. 8:305-310.

162. Silk, B. J., K. A. Date, K. A. Jackson, R. Pouillot, K. G. Holt, L. M. Graves, K. L. Ong, S. Hurd, R. Meyer, R. Marcus, B. Shiferaw, D. M. Norton, C. Medus, S. M. Zansky, A. B. Cronquist, O. L. Henao, T. F. Jones, D. J. Vugia, M. M. Farley, and B. E. Mahon. 2012. Invasive listeriosis in the foodborne diseases active surveillance network (FoodNet), 2004-2009: Further targeted prevention needed for higher-risk groups. *Clin. Infect. Dis*. 54:S396-S404.

163. Silk, B. J., M. H. McCoy, M. Iwamoto, and P. M. Griffin. 2014. Foodborne listeriosis acquired in hospitals. *Clin. Infect. Dis*. 59:532-540.

164. Smith, L. H., and S. G. Besser. 2000. Dietary restrictions for patients with neutropenia: A survey of institutional practices. *Oncol. Nurs. Forum*. 27:515-520.

165. Smith, M. A., K. Takeuchi, G. Anderson, G. O. Ware, H. M. McClure, R. B. Raybourne, N. Mytle, and M. P. Doyle. 2008. Dose-response model for *Listeria monocytogenes*-induced stillbirths in nonhuman primates. *Infect. Immun*. 76:726-731.

166. Sonbol, M. B., B. Firwana, M. Diab, A. Zarzour, and T. E. Witzig. 2015. The effect of a neutropenic diet on infection and mortality rates in cancer patients: A meta-analysis. *Nutr. Cancer*. 67:1230-1238.

167. Spaeth, G., R. D. Berg, R. D. Specian, and E. A. Deitch. 1990. Food without fiber promotes bacterial translocation from the gut. *Surgery*. 108:240-247.

168. Spickler, A. R. 2005. Listeriosis. Available at: http://www.cfsph.iastate.edu/Factsheets/pdfs/listeriosis.pdf. Accessed 18 May, 2020.

169. Ssemanda, J. N., H. Joosten, M. C. Bagabe, M. H. Zwietering, and M. W. Reij. 2018. Reduction of microbial counts during kitchen scale washing and sanitization of salad vegetables. *Food Control*. 85:495-503.

170. Stanley, C. D., and G. A. Clark. 2004. Water requirements for drip-irrigated tomato production in Southwest Florida. Available at: http://irrigationtoolbox.com/ReferenceDocuments/Extension/Florida/SS43200.pdf. Accessed 18 May, 2020.

171. Steinbruegge, E. G., R. B. Maxcy, and M. B. Liewen. 1988. Fate of *Listeria monocytogenes* on ready to serve lettuce. *J. Food Prot*. 51:596-599.

172. Stephan, R., D. Althaus, S. Kiefer, A. Lehner, C. Hatz, C. Schmutz, M. Jost, N. Gerber, A. Baumgartner, H. Hächler, and M. Mäusezahl-Feuz. 2015. Foodborne transmission of *Listeria monocytogenes* via ready-to-eat salad: aAnationwide outbreak in Switzerland, 2013–2014. *Food Control*. 57:14-17.

173. Swaminathan, B., and P. Gerner-Smidt. 2007. The epidemiology of human listeriosis. *Microbes and Infection*. 9:1236-1243.

174. Szabo, E. A., K. J. Scurrah, and J. M. Burrows. 2000. Survey for psychrotrophic bacterial pathogens in minimally processed lettuce. *Lett. Appl.Microbiol*. 30:456-460.

175. Takeuchi, K., and J. F. Frank. 2000. Penetration of *Escherichia coli* O157:H7 into lettuce tissues as affected by inoculum size and temperature and the effect of chlorine treatment on cell viability. *J. Food Prot*. 63:434-440.

176. Tang, Y. 2010. Reduction of *Salmonella*, *Escherichia coli* O157:H7, and *Listeria monocytogenes* on the surface of leafy greens and green onions using innovative home washing technologies. Master of Science, University of Georgia.

177. Taormina, P. J., L. R. Beuchat, M. C. Erickson, L. Ma, G. D. Zhang, and M. P. Doyle. 2009. Transfer of *Escherichia coli* O157:H7 to iceberg lettuce via simulated field coring. *J. Food Prot*. 72:465-472.

178. Tennant, S. M., E. L. Hartland, T. Phumoonna, D. Lyras, J. I. Rood, R. M. Robins-Browne, and I. R. van Driel. 2008. Influence of gastric acid on susceptibility to infection with ingested bacterial pathogens. *Infect. Immun*. 76:639-645.

179. Todd, J. 2017. Quantification of select factors influencing the risk of contamination by *Salmonella* on tomatoes from farm to fork. Doctor of Philosophy, Rutgers University.

180. Trifilio, S., I. Helenowski, M. Giel, B. Gobel, J. Pi, D. Greenberg, and J. Mehta. 2012. Questioning the role of a neutropenic diet following hematopoetic stem cell transplantation. *Biol. Blood Marrow Transplant*. 18:1385-1390.

181. Tromp, S. O., H. Rijgersberg, and E. Franz. 2010. Quantitative microbial risk assessment for *Escherichia coli* O157:H7, *Salmonella enterica*, and *Listeria monocytogenes* in leafy green vegetables consumed at salad bars, based on modeling supply chain logistics. *J. Food Prot*. 73:1830-1840.

182. Tummers, B. 2006. DataThief III. Available at: https://datathief.org/. Accessed 26 July, 2020.

183. U. S. Department of Agriculture, and Food and Drug Administration. 2011. Food Safety for People with Cancer. Available at: https://www.fda.gov/food/people-risk-foodborne-illness/food-safety-people-cancer. Accessed 26 July, 2020.

184. U. S. Department of Agriculture, FSIS. 2010. FSIS comparative risk assessment for *Listeria monocytogenes* in ready-to-eat meat and poultry deli meats. Available at: https://www.fsis.usda.gov/shared/PDF/Comparative_RA_Lm_Report_May2010.pdf. Accessed 26 July, 2020.

185. U. S. Food and Drug Administration. 2018. Selecting and serving produce safely. Available at: https://www.fda.gov/food/buy-store-serve-safe-food/selecting-and-serving-produce-safely#:~:text=Buy%20Right,refrigerated%20or%20surrounded%20by%20ice. Accessed 26 July, 2020.

186. U. S. Food and Drug Administration, FSIS. 2003. Quantitative assessment of relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods. Available at: https://www.fda.gov/food/cfsan-risk-safety-assessments/quantitative-assessment-relative-risk-public-health-foodborne-listeria-monocytogenes-among-selected. Accessed July 26, 2020.

187. U.S. Environmental Protection Agency, and U.S. Department of Agriculture Food Safety and Inspection Service. 2012. Microbial risk assessment guideline: Pathogenic microorganisms with focus on food and water.Available at: https://www.epa.gov/risk/microbial-risk-assessment-guideline-pathogenic-microorganisms-focus-food-and-water. Accessed 26 July, 2020.

188. van der Velden, W., A. H. E. Herbers, M. G. Netea, and N. M. A. Blijlevens. 2014. Mucosal barrier injury, fever and infection in neutropenic patients with cancer: introducing the paradigm febrile mucositis. *B. J. Haematol*. 167:441-452.

189. van Tiel, F. H., M. M. Harbers, P. H. W. Terporten, R. T. C. van Boxtel, A. G. Kessels, G. Voss, and H. C. Schouten. 2007. Normal hospital and low-bacterial diet in patients with cytopenia after intensive chemotherapy for hematological malignancy: a study of safety. *Ann. Oncol*. 18:1080-1084.

190. Wade, I., C. Santiago, K. Avila, S. Perez, A. Sant'Ana, and D. Schaffner. 2012. Transfer of E*nterobacter aerogenes* from surface to flesh during peeling of raw carrots and cucumbers (P2-43). International Association for Food Protection Annual Meeting, Providence, Rhode Island.

191. Warriner, K., F. Ibrahim, M. Dickinson, C. Wright, and W. M. Waites. 2003. Internalization of human pathogens within growing salad vegetables. *Biotechnol. Gen. Eng. Rev*. 20:117-136.

192. Watkins, J., and K. P. Sleath. 1981. Isolation and enumeration of *Listeria monocytogenes* from sewage, sewage-sludge, and river water. *J. Appl. Bacteriol*. 50:1-9.

193. Weis, J., and H. P. Seeliger. 1975. Incidence of *Listeria monocytogenes* in nature. *Appl. Microbiol*. 30:29-32.

194. Wells, C. L., M. A. Maddaus, and R. L. Simmons. 1988. Proposed mechanisms for the translocation of intestinal bacteria. *Rev. Infect. Dis*. 10:958-979.

195. Williams, D., J. Castleman, C. C. Lee, B. Mote, and M. A. Smith. 2009. Risk of fetal mortality after exposure to *Listeria monocytogenes* based on dose-response data from pregnant guinea pigs and primates. *Risk Anal*. 29:1495-1505.

196. Williams, D., E. A. Irvin, R. A. Chmielewski, J. F. Frank, and M. A. Smith. 2007. Dose-response of *Listeria monocytogenes* after oral exposure in pregnant guinea pigs. *J. Food Prot*. 70:1122-1128.

197. Winter, C. H., S. O. Brockmann, S. R. Sonnentag, T. Schaupp, R. Prager, H. Hof, B. Becker, T. Stegmanns, H. U. Roloff, G. Vollrath, A. E. Kuhm, B. B. Mezger, G. K. Schmolz, G. B. Klittich, G. Pfaff, and I. Piechotowski. 2009. Prolonged hospital and community-based listeriosis outbreak caused by ready-to-eat scalded sausages. *J. Hosp. Infect*. 73:121-128.

198. World Health Organization (WHO). 2018. Listeriosis. Available at: https://www.who.int/news-room/fact-sheets/detail/listeriosis. Accessed 26 July, 2020. World Health Organization (WHO), and Food and Agriculture Organization (FAO). 2004. Risk assessment of Listeria monocytogenes in ready-to-eat foods: Technical report. Available at: http://www.fao.org/3/y5394e/y5394e00.htm. Accessed 26 July, 2020.

199. Wu, F., Z. F. Guan, and D. H. Suh. 2018. The effects of tomato suspension agreements on market price dynamics and farm revenue. *Appl. Econ. Perspect. Policy*. 40:316-332.

200. Zhang, G. D., Y. Chen, L. J. Hu, D. Melka, H. Wang, A. Laasri, E. W. Brown, E. Strain, M. Allard, V. K. Bunning, M. Parish, S. M. Musser, and T. S. Hammack. 2018. Survey of

foodborne pathogens, aerobic plate counts, total coliform counts, and *Escherichia coli* counts in leafy greens, sprouts, and melons marketed in the United States. *J. Food Prot*. 81:400-411.

201. Zhang, S., and J. M. Farber. 1996. The effects of various disinfectants against *Listeria monocytogenes* on fresh-cut vegetables. *Food Microbiol*. 13:311-321.

202. Zhu, Q., R. Gooneratne, and M. A. Hussain. 2017. *Listeria monocytogenes* in fresh produce: Outbreaks, prevalence and contamination levels. *Foods*. 6.