III. EXPOSURE ASSESSMENT

Exposure is a function of the quantity of a food consumed and the level of contamination in that food. While the contamination level in food at consumption is the important parameter in evaluating public health, most of the available contamination data pertain to foods sampled at retail stores. Hence, it was necessary to develop estimates of the frequency and amount of each serving of the contaminated foods likely to be consumed in the United States, as well as the *Listeria monocytogenes* levels in those foods. Limitations inherent in food consumption data and the paucity of contamination data for certain foods made certain assumptions necessary to develop the estimates. These limitations and assumptions are discussed later in this chapter.

The goal of this risk assessment was to provide information needed to focus risk management strategies among a variety of foods that could be potentially contaminated with *Listeria monocytogenes*, the purpose of the exposure assessment is to estimate the contamination and consumption of foods that have a potential for *Listeria monocytogenes* contamination. Therefore, this risk assessment modeled growth of *Listeria monocytogenes* in foods during post-retail storage and reduction of levels during home cooking or reheating of frankfurters. Growth was also modeled for some contamination data that were collected pre-retail to account for possible growth between manufacture and retail.

Foods that were included in the risk assessment were identified through a comprehensive review of the recall, microbiological and epidemiological literature. Each food was placed in one of 23 food categories. Using distributions of contamination and consumption data, estimates of exposure to *Listeria monocytogenes* in the various foods were derived. The components of the exposure assessment are provided in Figure III-1, and specific modeling details are provided in Appendix 3.

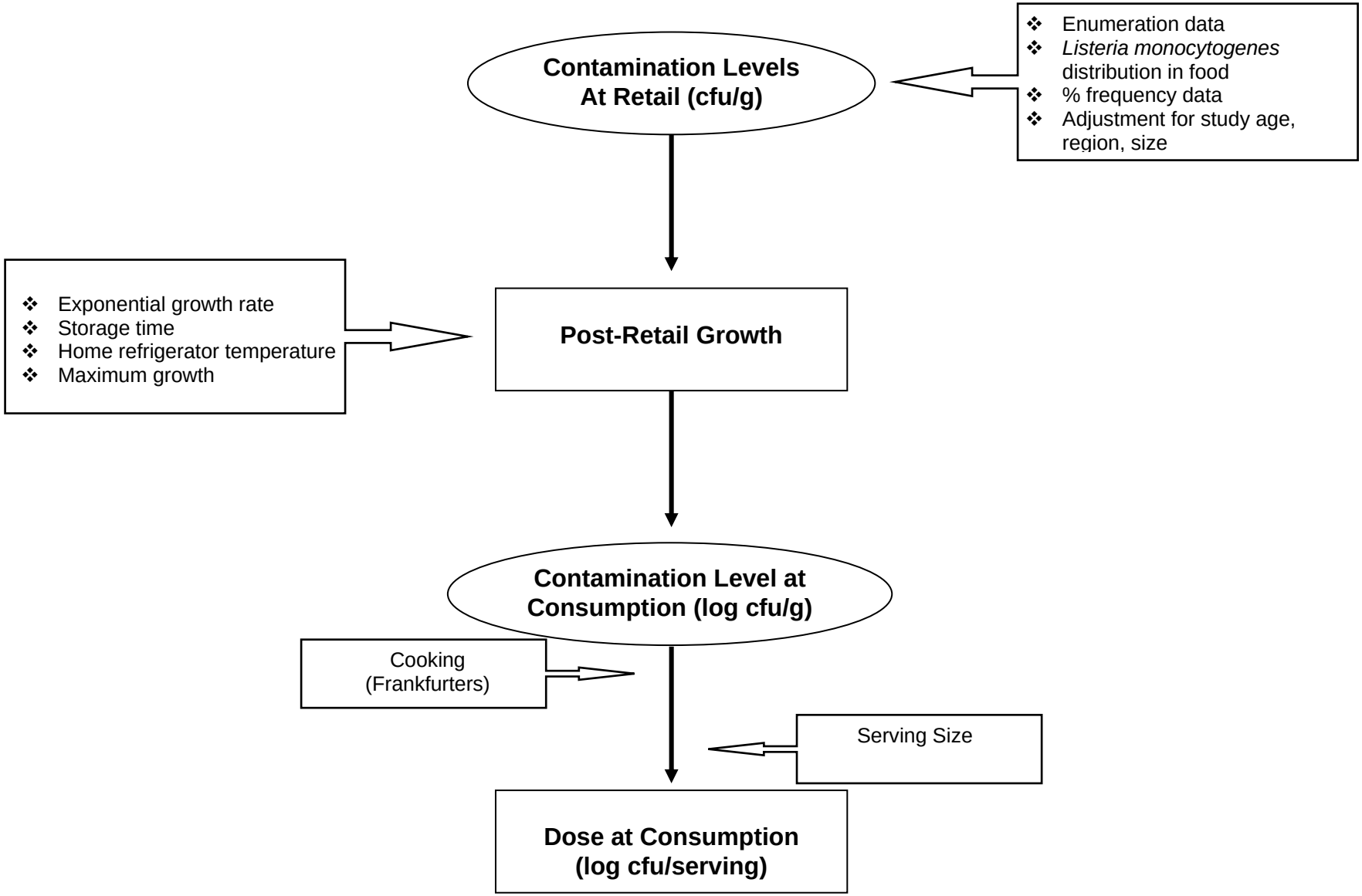


Figure III-1. Components of the Exposure Assessment Model

Food Category Identification

The first step in the exposure assessment was to consider appropriate foods to include in the risk assessment model. As the risk assessment progressed, foods and food categories were continually reevaluated and modifications were made based on new information, such as the results of growth models or new microbiological or epidemiological literature. Foods that have a significant potential for *Listeria monocytogenes* contamination were identified. They represent a subset of foods that comprise an individual's total diet. Foods that have not been linked to *Listeria monocytogenes* contamination were not included, for example, grain products (*e. g.,* bread, cookies, cakes), soft drinks, canned fruits, and cooked mixed dishes (*e. g.,* lasagna, soups). Furthermore, foods that have limited association with *Listeria monocytogenes* contamination (*e. g.,* cream-filled pastries) were not included because neither contamination level data nor appropriate data to serve as a substitute were available. It was also presumed that some foods that are cooked just prior to consumption (*e. g*., most meats and seafoods) present a very low likelihood of containing *Listeria monocytogenes* when consumed and were not included in this risk assessment. Eggs are an example of a food category that was not included in the risk assessment, but could be a vehicle for listeriosis. Although eggs have been implicated in one outbreak with two cases (Schwartz *et al*., 1988), *Listeria monocytogenes* has not been isolated from intact eggs and eggs products are typically cooked before consumption (Ryser and Marth, 1999).

A review of the literature was conducted to identify foods that have a significant potential for *Listeria monocytogenes* contamination. The review concentrated on the following:

- Outbreaks
- Sporadic cases, i.e. individual cases not reported as part of a documented outbreak
- Recalls and regulatory actions
- Literature related to prevalence and incidence of *Listeria monocytogenes* through analytical testing in North America (the United States and Canada)
- Literature on outbreaks, sporadic cases, and prevalence and incidence studies of *Listeria monocytogenes* in other countries

The next step in selecting foods for the risk assessment was a review of the available data on contamination and the ability of the food to support growth of *Listeria monocytogenes*. Food contamination data were compared with the available food consumption data to create food categories.

Foods that are ready-to-eat (RTE) were ultimately selected. Some RTE foods are raw and others receive some processing prior to sale. Still other RTE foods are fully cooked before sale but may be subjected to subsequent handling and storage, thereby increasing the possibility of recontamination.

The identified foods were further sorted into categories based upon food characteristics, use, and the potential for growth of *Listeria monocytogenes*. For example, Dry/Semi-dry Fermented Sausages were differentiated from other deli meats such as bologna, sliced turkey, and ham. The Cooked RTE Crustaceans food category contains peel-and-eat shrimp, steamed and boiled shrimp, and steamed crabs – foods that may be refrigerated and eaten chilled or allowed to cool after cooking, thus allowing for re-contamination and growth. The Vegetable food category includes many raw vegetables, as well as mixed vegetables such as bagged salads (without salad dressings). Similarly, the Fruits food category includes many raw and dried fruits and mixed fruits such as fruit salads (without salad dressings). In this updated risk assessment, the vegetable and fruit salads with salad dressings are included in the Deli-type Salad food category. While there is a single Deli-type Salad food category for reporting purposes, to model growth of *Listeria monocytogenes*, salads were segregated into growth and non-growth salads and considered the use of preservatives in salads made in bulk for distribution to retail stores.

In this updated risk assessment, the cheese categories have been reorganized into six categories based on moisture content. Another update to the categories included splitting the Miscellaneous Dairy Products into two categories. The Cultured Milk Products category includes the low pH dairy foods manufactured with lactic acid fermentation. Of this category, yogurt is the most frequently consumed food, followed by sour cream and buttermilk. The High Fat and Other Dairy Products category includes the remainder of the dairy products that generally support growth (including powdered products when reconstituted). Butter, cream and half and half are the most prominent foods in this category, but shakes and chocolate milk made with cocoa or syrup are also included. The frankfurter category has been divided into reheated and not

reheated frankfurters to distinguish the impact that reheating before consumption can have on the predicted risk. The number of unreheated frankfurters was represented by a triangular distribution with a minimum of 4%, most likely of 7%, and maximum of 10% of the total frankfurters consumed without reheating. These values were based on surveys conducted by USDA and American Meat Institute.

Table III-1 lists the 23 food categories that were used in this risk assessment. The food categories fall into five general groups: Seafood, Produce, Dairy, Meat, and Combination Foods. (See Appendix 5 for a detailed listing of the foods included in each food category.)

Table III-1. Food Categories Used in this Listeria monocytogenes Risk Assessment

SEAFOOD
Smoked Seafood (i.e., finfish and mollusks)
Raw Seafood (i.e., finfish and mollusks)
Preserved Fish (i.e., dried, pickled, and marinated finfish)
Cooked Ready-to-Eat Crustaceans (i.e., shrimp and crab)
PRODUCE
Vegetables (raw)
Fruits (raw and dried)
DAIRY
Fresh Soft Cheese (i.e., Queso Fresco, Queso de Creama, and Queso de Puna)
Soft Unripened Cheese, >50% moisture (i.e., cottage cheese, cream cheese, and ricotta)
Soft Ripened Cheese, >50% moisture (i.e., brie, camembert, feta, and mozzarella)
Semi-soft Cheese, 39-50% moisture (i.e., blue, brick, monterey, and muenster)
Hard Cheese, <39% moisture (i.e., cheddar, colby, and parmesan)
Processed Cheese (i.e., cheese foods, spreads, and slices)
Pasteurized Fluid Milk
Unpasteurized Fluid Milk
Ice Cream and Frozen Dairy Products
Cultured Milk Products (i.e., yogurt, sour cream and buttermilk)
High Fat and Other Dairy Products (i.e., butter, cream, other miscellaneous dairy products)
MEAT
Frankfurters (reheated)
Frankfurters (not reheated)
Dry/Semi-dry Fermented Sausages
Deli Meats (cooked, ready-to-eat)
Pâté and Meat Spreads
COMBINATION FOODS
Deli-type Salads (i.e., fruit, vegetable, meat, pasta, egg, or seafood salads with dressing)

Food Consumption Data

Data from two large-scale, nationwide food consumption surveys were used to provide estimates of exposure to *Listeria monocytogenes* via distributions of food consumption. The first survey is the Continuing Survey of Food Intakes by Individuals (CSFII 1994-96). This is the latest survey of consumers of all ages conducted by USDA's Agricultural Research Service (USDA/ARS, 1998a, 1998b). The survey consists of the following:

- Two 24-hour recalls of foods eaten during two nonconsecutive days (with the interview for the second day conducted 3 to 10 days after the interview for the first day, but not on the same day of the week).
- Sample weights for weighting the data so that they will more closely reflect consumption by the non-institutionalized United States population.
- A sample of 16,103 respondents, including:

Pregnant and/or lactating women	(n = 123)
Children under 4 years	(n = 2,284)
People 60 years and older	(n = 2,315)

- Over sampling of low income, young children, and the elderly (USDA ARS, 1998a).
- A Population Parameter of 261,897,280, appropriate for 1994-1996.

The second nationwide survey of food consumption is the Third National Health and Nutrition Examination Survey (NHANES III), which was conducted in 1988 to 1994 (US DHHS, 1998). NHANES was conducted by the National Center for Health Statistics in the Center for Disease Control and Prevention (CDC/NCHS), DHHS. The survey consists of the following:

- One 24-hour recall of foods eaten.
- Sample weights for weighting the data so that they will more closely reflect consumption by the non-institutionalized United States population.
- A sample of 30,818 respondents, including:

Pregnant and/or lactating women	(n = 399)
Children under 4 years	(n = 3,979)
People 60 years and older	(n = 3,919)

- Over sampling of young children, older persons, black persons, and Mexican Americans.
- A United States Population Parameter of 251,097,003, appropriate for 1988-1994.

Consumption data from the CSFII 94-96 survey were used for 21 of the 23 food categories. CSFII data were used preferentially because they are newer and account for up to two days of eating per respondent. Data for unpasteurized fluid milk and unreheated frankfurters were modeled based on CSFII data for pasteurized milk and all frankfurters consumed. NHANES III data were used for two food categories (Raw Seafood and Preserved Fish) for which there are fewer than 30 eating occasions (servings) in the CSFII survey.

The surveys contain consumption data for many foods and each food has an associated food code. Over 640 food codes for RTE foods were matched to one of the 23 food categories. The following information was extracted from the databases for each food category:

- Weighted descriptives (*e. g.,* mean amount eaten in grams, median amount eaten in grams, number of servings) that characterize all eating occasions in two nonconsecutive days of eating (one day for NHANES III).
- Distributions of the amount of food (in grams) eaten in all servings over two days (one day for NHANES III).
- Distributions of the amount of food (in grams) eaten in all servings, expressed as weighted percentiles.
- Weighted descriptives to describe the amount of the food (in grams) eaten per person per day, as well as the number of eaters.
- Per capita estimates of food eaten.

Several limitations of the food consumption surveys had an impact on their use for risk assessment purposes. For some foods, it was a challenge to determine consumption. Surveys listed some particular foods under several food codes, such as ham consumed alone or ham in a ham sandwich. The proportion of a particular food (such as ham) in a mixed ingredient product (such as a ham sandwich) was determined using a generic recipe provided by the survey. The gram amount of the food (ham) consumed was then calculated and added to the intake derived from other food codes for the specific food (ham). For this risk assessment, sandwiches were

broken down into individual ingredients. Specifically, for frankfurters, dry semi/dry fermented sausages, deli meats, pâté and meat spreads, and deli salads, the actual consumption of meat or deli salad product consumed alone, as well as the proportion used in sandwiches, was used. In the case of vegetable and fruit salads (in which fruits and vegetables were the major component) and deli-type salads (not included in a sandwich), however, the entire salad was used, rather than the component ingredients.

The consumption surveys do not collect information from consumers to determine whether the milk they drank was pasteurized or unpasteurized (raw). Although federal law requires milk in interstate commerce to be pasteurized, some states allow unpasteurized milk to be sold and consumed within the state. Results of a 1995 FDA/CDC survey of all 50 states, Puerto Rico, and the District of Columbia, showed that 28 states (54%) permit the sale of unpasteurized fluid milk. However, it is estimated that unpasteurized milk accounts for less than 1% of the total volume of milk sold in these states (Headrick *et al.*, 1998). Because consumption surveys did not list "drinking occasions" (servings) of unpasteurized fluid milk, the consumption of this food category was modeled by estimating it as 0.5% of the amount consumed per serving of pasteurized milk (54% x 1%). The consumption surveys did not provide any information on the storage and heating of frankfurters. Estimates for the fraction of frankfurters stored frozen before consumption and those eaten without reheating were obtained from other surveys.

Another limitation of food consumption surveys used is that some food categories have a small number of servings. Estimates based upon small sample sizes may be less statistically reliable than estimates based on larger sample sizes (USDA/ARS, 1998a). Although weighted food consumption data provide a better representation of the United States population, weighting small samples does not provide better reliability. In addition, the surveys do not provide corrections to account for underreporting and over reporting of the amount of a food eaten by consumers.

The food consumption surveys did not collect demographic information delineating consumers who are immunocompromised. Furthermore, the surveys did not measure consumption by the elderly who are living in nursing homes or other forms of assisted living outside of the home, nor did they contain a large enough sample of pregnant women to generalize consumption to all pregnant women. Thus, the available consumption data did not allow the determination of

comprehensive estimates of food consumption for each individual susceptible subpopulation. Consumption between the subpopulations was compared. Specifically, nonparametric statistical analyses were conducted to determine if there were significant differences between the distributions of the amount eaten in each serving (expressed as weighted percentiles) for the elderly and the intermediate-age population. Seventeen food categories had sufficient consumption data to permit these analyses. There were no statistically significant differences in consumption patterns for 14 of the examined 17 food categories. Thus, for the purpose of estimating the distribution of serving sizes, the food consumption data representing all eaters were used.

Note: Starting in 2002, CFSII and the dietary component of NHANES were merged into NHANES. The integrated survey will provide two 24-hour recalls of food consumption for 5,000 individuals a year and characterize "What We Eat in America."

Annual Number of Servings of Foods

In order to estimate the number of servings of the foods in each food category eaten in a year, some key data assumptions were necessary. First, it was assumed that the weighted number of servings for one (NHANES III) or two days (CSFII) of consumption of the foods in a specific food category could be extrapolated to the number of servings of those foods eaten by the population on an annual basis. Second, it was assumed that the weighted number of eaters of a food per day would represent the number of eaters of the food over 365 days. Obviously, there are some foods that individuals are more likely to eat each day (*e. g.,* vegetables, milk) and others that they eat frequently (*e. g*., fruits, deli meats) or occasionally (*e. g*., frankfurters, cottage cheese). Some foods are seasonal and are not available year round (*e. g*., some fruits and vegetables), and people may not be likely to purchase more costly items (*e. g*., shrimp, crabmeat) for regular consumption. Thus, it is important to note that when estimating the consumption of foods on an annual basis, all foods reported in food consumption surveys during a one- or two-day period are not likely to be eaten in the same frequency by the same people over an entire year. To estimate the number of annual servings for each food category, we divided the weighted number of serving consumed in two days by 2 (one-day basis) and then multiplied that value by 365 (annual basis). Table III-2 provides the annual number of servings of food consumed in the United States for each of the 23 food categories.

The annual number of servings associated with the pregnancy exposures resulting in neonatal deaths were estimated using the number of servings in the intermediate-aged group multiplied by the birth rate (1.74%) and a fractional exposure period. A triangular distribution with a minimum of 1 day, a most likely value of 7 days, and a maximum value of 30 days was used to represent the uncertainty in the exposure period. In order to estimate the number of servings in the neonatal group, the annual number of servings in the intermediate-age group was multiplied by the exposure period (triangle distribution) and divided by 365 days to estimate the number of per annum servings consumed by pregnant women. Because the perinatal exposure period is longer than neonatal (the total number of deaths includes prenatal, i.e., stillbirth, cases occurring in the last trimester), perinatal per serving death rates from listeriosis were estimated using an exposure period of 90 days (3/12 yr = 0.25) and a pregnancy rate (2.77%) rather than birth rate.

Serving Size Distributions

Empirical distributions were used to describe the serving sizes (grams of food eaten per serving) in the 23 food categories. These distributions are expressed as a series of population percentiles of the amount of food eaten per serving, weighted to reflect the consumption survey demographics. There were no uncertainties presented for these food categories because empirical distributions were used. The uncertainties associated with the serving size distributions would be relatively small, compared to other uncertainty distributions in this risk assessment for three reasons. First, even the smallest data sets used to characterize the serving size distributions are large relative to other parts of the *Listeria monocytogenes* risk model. Second, although the data may not be completely representative of the current behavior of the United States population, the data come from surveys that were explicitly designed for that purpose. Third, the variability (range) in serving sizes covers a smaller range (two logs) than many other parts of the model.

Table III-3 shows the 50^{th} (median), 75^{th}, 95^{th} and 99^{th} percentiles of the weighted distributions of serving size. For example, these percentiles for Smoked Seafood are 57, 75, 136 and 142 g/serving, respectively. This distribution indicates that half of the servings were less than 57 g and 95% of the servings were less than 136 g.

Table III-2. Estimates of the Total Number of Annual Servings of Foods Consumed in the United States by Population and Food Category

Food Category[a]	Intermediate-Age Population	Perinatal Population[b]	Elderly Population	Total Population[c]
SEAFOOD				
Smoked Seafood	1.6×10^8	1.1×10^6	4.1×10^7	2.0×10^8
Raw Seafood	1.8×10^8	1.3×10^6	5.7×10^5	1.8×10^8
Preserved Fish	8.3×10^7	5.7×10^5	2.2×10^7	1.1×10^8
Cooked Ready-to-Eat Crustaceans	4.7×10^8	3.3×10^6	8.1×10^7	5.5×10^8
PRODUCE				
Vegetables	6.8×10^{10}	4.7×10^8	1.7×10^{10}	8.5×10^{10}
Fruits	3.7×10^{10}	2.5×10^8	1.2×10^{10}	4.9×10^{10}
DAIRY				
Fresh Soft Cheese	6.9×10^7	4.8×10^5	1.3×10^6	7.1×10^7
Soft Unripened Cheese	3.4×10^9	2.3×10^7	1.0×10^9	4.4×10^9
Soft Ripened Cheese	1.7×10^9	1.2×10^7	1.8×10^8	1.9×10^9
Semi-soft Cheese	1.6×10^9	1.1×10^7	1.5×10^8	1.8×10^9
Hard Cheese	7.8×10^9	5.4×10^7	1.3×10^9	9.0×10^9
Processed Cheese	1.1×10^{10}	7.6×10^7	1.6×10^9	1.2×10^{10}
Pasteurized Fluid Milk[d]	7.2×10^{10}	5.0×10^8	1.5×10^{10}	8.7×10^{10}
Unpasteurized Fluid Milk[d]	3.6×10^8	2.5×10^6	7.5×10^7	4.4×10^8
Ice Cream and Frozen Dairy Products	1.2×10^{10}	8.2×10^7	3.1×10^9	1.5×10^{10}
Cultured Milk Products	6.1×10^9	4.2×10^7	1.2×10^9	7.2×10^9
High Fat and Other Dairy Products	1.6×10^{10}	1.1×10^8	4.3×10^9	2.1×10^{10}
MEAT				
Frankfurters, reheated[e]	5.5×10^9	3.8×10^7	5.8×10^8	6.1×10^9
Frankfurters, not reheated[e]	4.2×10^8	2.9×10^6	4.4×10^7	4.7×10^8
Dry/Semi-dry Fermented Sausages	1.5×10^9	1.1×10^7	2.5×10^8	1.8×10^9
Deli Meats	1.8×10^{10}	1.2×10^8	2.8×10^9	2.1×10^{10}
Pâté and Meat Spreads	9.7×10^7	6.7×10^5	2.1×10^7	1.2×10^8
COMBINATION FOODS				
Deli-type Salads	1.0×10^{10}	7.0×10^7	3.1×10^9	1.3×10^{10}

[a] Serving size data based on CSFII 94-96 extrapolated from two days of eating to an annual basis, except data for Raw Seafood and Preserved Fish from NHANES III were extrapolated from one day of eating. Servings denote variable amounts consumed and not a standard serving size that represents the amount customarily consumed per eating occasion.

[b] For the purposes of estimating rates of listeriosis per serving, the values for the perinatal group were calculated by adjusting the number of annual servings for the intermediate-aged group for the annual pregnancy rate: The annual pregnancy rate (2.77%) was multiplied by the number of servings for the intermediate-aged population and 0.25 (0.25 = 3/12, to estimate the number of pregnant women in the last 3 months of pregnancy).

[c] The annual number of servings for the total population was calculated by summing the values for the elderly and intermediate-aged populations. The perinatal group was not included because the servings for this population are a subset of the intermediate-aged group.

[d] Consumption of Pasteurized Fluid Milk is based on 99.5% of total milk consumption and consumption of Unpasteurized Fluid Milk is based on 0.5% of total fluid milk consumption.

[e] Consumption of not reheated frankfurters is a distribution based on an uncertainty range of 4 to 10% of the consumption of frankfurters. The value in the table is the mean of the distribution. The value for reheated frankfurters is the difference between the total frankfurters consumption and the value for not reheated frankfurters.

Table III-3. Percentiles of Serving Size Distributions for Each Food Category

Food Categories	Weighted Percentiles (grams per serving)[a]			
	50th	75th	95th	99th
Seafood				
Smoked Seafood	57	75	136	142
Raw Seafood	16	28	77	136
Preserved Fish	70	125	130	250
Cooked Ready-to-Eat Crustaceans	50	96	256	345
Produce				
Vegetables	28	55	123	220
Fruits	118	138	272	570
Dairy				
Fresh Soft Cheese	31	85	246	246
Soft Unripened Cheese	29	105	226	420
Soft Ripened Cheese	28	48	85	168
Semi-soft cheese	28	57	142	227
Hard Cheese	28	38	85	122
Processed Cheese	21	42	84	130
Pasteurized Fluid Milk	244	245	488	732
Unpasteurized Fluid Milk	244	245	488	732
Ice Cream and Frozen Dairy Products	132	186	330	454
Cultured Milk Products	114	227	245	490
High Fat and Other Dairy Products	13	30	312	510
Meats				
Frankfurters (reheated and not reheated)	57	114	171	285
Dry/Semi-dry Fermented Sausages	46	69	161	161
Deli Meats	56	75	113	196
Pâté and Meat Spreads	57	85	128	454
COMBINATION FOODS				
Deli-type Salads	97	177	301	464

[a] There are no uncertainties presented for these food categories because empirical distributions were used.

Note: Serving size denotes variable amount consumed and are not a standard serving size that represents the amount customarily consumed per eating occasion.

Food Contamination Data

Over the last fifteen years, numerous studies have been published that report on foods contaminated with *Listeria monocytogenes* in a variety of countries and locations. Contamination data included in this risk assessment were reported from the United States and other countries on six continents. Most of the studies were from the industrialized countries of North America and Western Europe. Many studies did not identify the sampling of imported foods or indicate whether imports were excluded from the study. Contaminant serotype information was not considered because the food contamination studies did not usually identify the serotypes.

Data sources included the published scientific literature, published and unpublished official government documents, and data obtained from the private sector. All data and references are available in the docket established for this risk assessment. Two types of data describing the levels of *Listeria monocytogenes* contamination in food were identified.

- Presence/absence (qualitative) data (i.e., the number of positive samples relative to the total sample collection).
- Enumeration (quantitative) data (i.e., the number of colony forming units (cfu) of *Listeria monocytogenes* that were measured from a sample). It is conventionally assumed that one cfu is equivalent to one organism.

Both qualitative and quantitative studies were used in the assessment (Table III-4; Appendix 7). Data from presence/absence studies (qualitative data) were converted to numerical data and included in the model by assigning the lowest possible contamination level that can be detected by the laboratory method. For a method that uses a 25-g sample, the lowest detectable level is 0.04 cfu/g of food. Consequently, the qualitative data could be used along with the quantitative data in the construction of the cumulative distribution curves of *Listeria monocytogenes* levels in food.

Because each food category usually includes many related types of foods, data were collected to represent all the foods in a designated food category. For example, the deli meats include, in part, ham, bologna, and sliced chicken. These deli meats have diverse microbial characteristics and there are relatively few existing studies for each of these foods. Hence, all data available on these products

were used with the assumption that the summation of the collected data represented the diverse compositional, geographic, seasonal, home vs. away-from-home, relative frequency of consumption, and other factors that affect the exposure from *Listeria monocytogenes* in these foods. Where methodologies or designations varied among multiple data sources, the original data were often regrouped or recalculated (particularly for the growth modeling work).

Table III-4. *Listeria monocytogenes* Contamination: Numbers of Qualitative and Quantitative Studies and Samples

Food Category	Number of Studies[a]				Number of Samples[b]	Percent of Positive Samples[c]
	Total	United States	Total Quant-itative	United States Quant-itative		
SEAFOOD						
Smoked Seafood	30	6	10	2	7,855	12.9
Raw Seafood	46	11	4	1	15,650	7.0
Preserved Fish	18	1	5	0	1,495	9.8
Cooked Ready-to-Eat Crustaceans	11	4	3	2	4,004	2.8
PRODUCE						
Vegetables	32	5	8	1	9,223	3.6
Fruits	4	2	0	0	254	11.8
DAIRY						
Fresh Soft Cheese	8	3	1	1	4,866	1.4
Soft Unripened Cheese	8	2	3	0	814	3.9
Soft Ripened Cheese	17	3	5	1	3,109	3.8
Semi-soft Cheese	11	3	3	1	2,615	3.1
Hard Cheese	12	2	2	0	973	1.4
Processed Cheese	4	1	1	0	325	0.9
Pasteurized Fluid Milk	30	3	3	1	12,407	0.4
Unpasteurized Fluid Milk	45	10	3	0	19,080	4.1
Ice Cream and Frozen Dairy Products	22	5	2	0	170,787	0.2
Cultured Milk Products	6	1	1	0	490	0.8
High Fat and Other Dairy Products	12	4	2	0	18,169	1.3
MEAT						
Frankfurters	9	6	2	2	3,763	4.8
Dry/Semi-dry Fermented Sausages	14	3	3	0	3,357	6.4
Deli Meats	19	4	3	1	33,824	1.9
Pâté and Meat Spreads	12	3	7	0	5,665	6.5
COMBINATION FOODS						
Deli-type Salads	16	6	5	1	17,915	3.8

[a] See Appendix 5 for the reference citation for each study.
[b] Total number of samples equals qualitative plus quantitative samples for each category.
[c] The percent of positive samples was calculated using the total positive samples in a food category. The value in the table is an unweighted percentage (i.e., does not reflect the weighting done to represent study reliability for predicting current *Listeria monocytogenes* levels in the United States).

Pairing consumption data with the appropriate contamination data was often imperfect. Dietary intake data were highly specific as to the type of food consumed (*e. g.*, smoked mussels). In contrast, the contamination data reported in the literature were often more generic (*e. g.*, samples may only be described as shellfish).

The analytical methods used in the food contamination studies to determine the presence of *Listeria monocytogenes* were generally well known and were approximately equal in sensitivity at about 1 cfu per 25 g sample (0.04 cfu/g). However, for enumeration methods of analysis, the sample size was usually less than 25 g and was not as sensitive (typically 20 to 50 cfu/g). Typically, the samples obtained for analysis were from non-composited samples of food. An exception, however, was unpasteurized fluid milk obtained from bulk tanks.

Contamination levels at consumption were modeled with the assumption that contamination distributions for a given food in the United States do not vary significantly from those in other countries, especially Western Europe and other developed countries. Similarly, it was assumed that all foods within a category have a similar pattern of contamination. Furthermore, all *Listeria monocytogenes* food isolates were accepted as having the potential to cause human illness. No differences in ability to grow or other characteristics between food and clinical isolates were assumed. As will be discussed later, the impact of these assumptions was considered in the uncertainty associated with relative risk determinations.

The available data on *Listeria monocytogenes* levels had some limitations that affected the distributions for levels of *Listeria monocytogenes* in foods. First, there are relatively few data points above the limit of detection (0.04 cfu/g). This is because the occurrence of detectable levels of *Listeria monocytogenes* in food is rare and because most surveys of the occurrence of *Listeria monocytogenes* in food did not quantify the levels in positive samples. Second, some of the data are not from the United States and this data may not always be representative of food and processing procedures in the United States. To create an estimate of the current United States distribution, the data sets were weighted by the number of samples in the data set, likelihood of the food in that country to be imported to the United States food supply, and the recency of the data. Third, there was a wide degree of variation between studies in the

occurrence of high levels of *Listeria monocytogenes*. The extent to which this variation reflects true variation in a particular food, is not known.

Many of the studies found in the published literature were conducted in the late 1980s and early 1990s. The extent that improved sanitation and other control measures implemented by the food industry have reduced the frequency and level of contamination since 1993 (when the earlier research was conducted) is difficult to determine from published literature. It was felt that some allowance should be made for the age of data and therefore, all data were used but the more recent data were given greater weight (details below). Because some food categories had little data, which would result in a biased estimate, the overall trend in contamination for all the food categories from before 1993 to after 1998 was obtained and applied to these data sets.

The length of time a food was held at retail before it was obtained for microbial sampling was not recorded in the survey studies. It was therefore necessary to assume that foods were sampled without bias and would represent the entire range of post-production and pre-sale conditions for that food.

Growth Data

Growth of *Listeria monocytogenes* in food is a function of the storage time, storage condition, and rate of growth in specific foods. The storage times were multiplied by the rate of growth to provide an estimate of the amount of *Listeria monocytogenes* growth occurring between retail purchase of the food and its consumption. The model includes consideration of the interaction of storage time and temperature and maximum growth that specific foods support.

Storage time

Some foods are consumed on the day of purchase whereas others remain in the home refrigerator for lengthy periods of time. This is a major source of variability in the estimate of growth and ultimately, in the numbers of *Listeria monocytogenes* consumed. Except for frankfurters and deli meats, no data were found on the storage of foods in the home; therefore, storage time, including variation and uncertainty, were estimated based on the expert judgment of the risk assessment team in consideration of recommendations developed by the Food Marketing

Institute (2002) and other individuals familiar with the production and use of the various foods. It is recognized that foods may be kept beyond the recommended storage times. This risk assessment modeled estimated consumer food practices, not necessarily the recommended practices. The values were developed by consensus of the risk assessment team and vetted by government subject matter experts and other scientific reviewers including those who submitted comments following the release of the draft risk assessment. The minimum, most likely and maximum storage times used to develop the distribution of storage times for the food categories are presented in Table III-5. These are skewed distributions with relatively few servings at the maximum storage time. For Smoked Seafood, as an example, over 90% of the servings are stored for less than 13 days.

Table III-5. Variation in Post-Retail Storage Times Assigned to the Food Categories

Food Categories	Storage time (days)[a]		
	Minimum	Most Likely	Maximum
SEAFOOD			
Smoked Seafood	0.5	3 to 5	15 to 30
Raw Seafood	0.5	1 to 2	10 to 20
Preserved Fish		[Not Applicable][b]	
Cooked Ready-to-Eat Crustaceans	0.5	1 to 2	10 to 20
PRODUCE			
Vegetables	0.5	3 to 4	8 to 12
Fruits	0.5	3 to 4	8 to 12
DAIRY			
Fresh Soft Cheese	0.5	1 to 5	15 to 30
Soft Unripened Cheese	0.5	6 to 10	15 to 45
Soft Ripened Cheese	0.5	6 to 10	15 to 45
Semi-Soft Cheese	0.5	6 to 10	15 to 45
Hard Cheese	0.5	6 to 10	90 to 180
Processed Cheese	0.5	6 to 10	45 to 90
Pasteurized Fluid Milk	0.5	3 to 5	10 to 15
Unpasteurized Fluid Milk	0.5	2 to 3	7 to 10
Ice Cream and Frozen Dairy Products		[Not Applicable][b]	
Cultured Milk Products	0.5	6 to 10	15 to 45
High Fat and Other Dairy Products	0.5	6 to 10	15 to 45
MEATS			
Frankfurters		[Not applicable][c]	
Dry/Semi-Dry Fermented Sausages	0.5	6 to 10	45 to 90
Deli Meats		[Not applicable][c]	
Pâté and Meat Spreads	0.5	6 to 10	15 to 45
COMBINATION FOODS			
Deli-type Salads	0.5	3 to 4	8 to 12

[a] For the food categories a BertPert distribution with these minimum, most likely and maximum parameters were used.
[b] Not applicable because this is a food category that does not support growth.
[c] Emperical data was used (see Table III-6).

Estimating duration of post-retail storage for Frankfurters and Deli Meats

Preliminary data from a study being conducted for FSIS by Georgetown University (Wachsmuth, 2000) provided information for frankfurters and deli meats used in the draft risk assessment. For frankfurters, 3 of 73 respondents gave 21 days storage and 3 gave 30 days as the maximum time. For deli meats, 2 of 81 respondents gave 21 days of storage, and 2 gave 30 days as the maximum time. FSIS also questioned people who called in to their telephone Meat and Poultry Hot Line about their frankfurter storage and cooking or reheating practices. Of 136 callers, one had kept frankfurters 90 days and one for 180 days (Wachsmuth, 2000).

In response to the need for more comprehensive information on consumer practices for frankfurters and deli meats, the American Meat Institute (AMI) commissioned a consumer survey that asked how long, on average, deli meats and frankfurters were stored before consumption (American Meat Institute, 2001). The responses are shown in Table III-6. These data were used to model storage times for frankfurters and deli meats as described in section "Modeling: Growth Between Retail and Consumption."

Table III-6. Refrigerated Storage Times for Frankfurters and Deli Meats in the Home

Average Storage Time	Distribution (Fraction) of Responders[a]	
	Pre-packaged deli meats and frankfurters	Custom sliced deli meats
1 to 3 days	0.32	0.39
4 to 7 days	0.37	0.36
8 to 10 days	0.06	0.03
11 to 14 days	0.04	0.01
15 to 21 days	0.01	0
22 to 30 days	0.01	0
31 to 60 days	0.01	0
61 days or more	0	0
Always freeze	0.03	0.01
Don't eat	0.13	0.17
Don't know/refused	0.02	0.02

[a]Source: American Meat Institute, 2001

Refrigeration Storage temperature

Data for home refrigerator temperatures were obtained from a 1999 survey conducted by Audits International (Audits International, 1999). Nine hundred thirty nine refrigerators in the United

States were included in the survey. Approximately 26% of the refrigerators exceeded 41°F (5°C) and 1.4% exceeded 50°F (10°C) (Table III-7). The refrigeration temperatures were modeled with a discrete distribution where temperature values were randomly sampled from the data provided by Audits International.

Table III-7. Frequency Distribution of Home Refrigerator Temperatures

Refrigerator Temperature (°F)	Frequency (%)
< 32	9
33 - 35	10
36 - 38	25
39 - 41	29
42 - 44	18
45 - 47	5
48 - 50	3
51 - 53	0.4
54 - 56	0.5
57 - 59	0.4
60 - 63	0.1

Total number of refrigerators in survey = 939 (Audits International, 1999)

Growth Rate

A summary of the growth rate data is presented in Table III-8 and a complete list of the literature data can be found in Appendix 8. Significant differences in composition and processes are present within many of the food categories. Within the Smoked Seafood food category, for example, there were hot and cold smoked fish, various salt levels, both aerobic and vacuum packaging, and different fish species. The modeling process used a cumulative table of the actual data points, not the means and standard deviations presented in Table III-8.

Table III-8. Mean Exponential *Listeria monocytogenes* Growth Rates and Total Number of Samples From Growth Rate Studies for Each Food Category

Food Categories	Growth Rate at 5 °C		Number of Samples[b]
	Mean (log_{10} cfu/g per day)[a]	Standard Deviation	
SEAFOOD			
Smoked Seafood	0.150	0.096	27
Raw Seafood	0.152	0.126	5
Preserved Fish		**No Growth**	
Cooked Ready-to-Eat Crustaceans	0.384	0.110	3
PRODUCE			
Vegetables	0.072	0.114	26
Fruits	0.046	0.047	5
DAIRY			
Fresh Soft Cheese	0.082	0.138	10
Soft Unripened Cheese	0.090	0.286	29
Soft Ripened Cheese	- 0.013[a]	0.133	17
Semi-soft cheese	- 0.043[a]	0.032	10
Hard Cheese	- 0.053[a]	0.065	11
Processed Cheese	- 0.045[a]	0.055	6
Pasteurized Fluid Milk[c]	0.257[c]	0.105	11
Unpasteurized Fluid Milk[c]	0.257[c]	0.105	11
Ice Cream and Frozen Dairy Products		**No Growth**	
Cultured Milk Products	- 0.168[a]	0.142	5
High Fat and Other Dairy Products	0.114	0.118	6
MEATS			
Frankfurters	0.131	0.051	5
Dry/Semi-dry Fermented Sausage	- 0.016[a]	0.016	4
Deli Meats	0.282	0.196	23
Pâté and Meat Spreads	0.252	0.154	2
COMBINATION FOODS			
Deli-type Salads (growth)	0.122	0.030	2
Deli-type Salads (non-growth)	-0.143	0.134	19

[a]Negative values indicate a decline in population for the mean growth rate.
[b] See Appendix 8 for more details about the studies.
[c]Pasteurized and unpasteurized milk were combined for analysis of exponential growth rate of fluid milk.

Modeling: *Listeria monocytogenes* Levels in Food at Retail

The majority of the data collected on the contamination of foods only determined whether or not a sample, typically 25 g, contains *Listeria monocytogenes*. Compared to the amount of qualitative data on the presence or absence of *Listeria monocytogenes* in foods, there is relatively little recent quantitative data available. This is due to the additional laboratory effort necessary

to enumerate samples, the low frequency of detecting positive samples, the need to test a large number of samples, and regulatory requirements that do not require enumerative data. Therefore, the approach taken was to develop a generic contamination model to describe the distribution of *Listeria monocytogenes* in food.

A three-step process was used to model levels of *Listeria monocytogenes* in food at retail.

Step 1: Characterize the distribution of *Listeria monocytogenes* across food categories using the contamination data reported in selected quantitative data sets (i.e., create a generic distribution).
Step 2: Characterize the uncertainty distribution for the frequency of detectable contamination for each food category using prevalence data adjusted to account for study size, age, and country of origin.
Step 3: Integrate the quantitative data from generic distributions (step 1) with the adjusted prevalence data, specific for each food category (step 2).

The general approach was to assume that the contaminated samples are detectable contaminations arising from a continuous log normal distribution of contamination. The minimum detectable level from presence/absence tests is typically 1 organism in 25 g or 0.04 organisms per gram. A low percentage of samples has contamination at or above this level and the remainder has non-detectable levels (i.e., <0.04 organisms/g). There may be no detectable *Listeria monocytogenes* in a specific sample (a 25.0 g package), but if 1000 packages from that lot are analyzed *Listeria monocytogenes* might be found. The average contamination could be one organism in 1000 packages (or a level of 0.00004 organisms per gram), far below the detectable level of 0.04 organisms/g. Therefore, what is observable with the presence/absence and quantitative tests is only the upper tail of the distribution. As shown in Figure III-2, the model fits a curve to the log cfu/g data and the mean and standard deviation are calculated. This curve represents a food category with approximately 10% of the samples positive for *Listeria monocytogenes*. It also shows that 3.1% of the samples have more than 100 cfu/g.

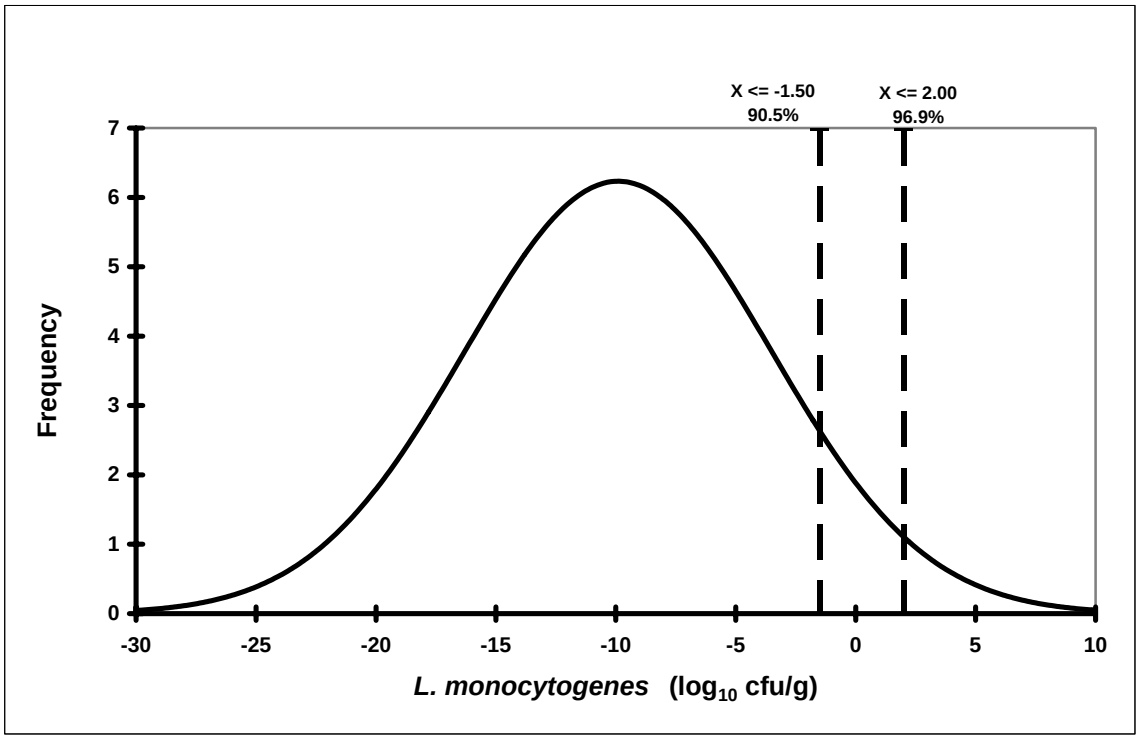


Figure III-2. Example of the Contamination Curve for a Typical Food Category Showing Frequencies of Detectable and Nondetectable Samples

Studies with enumerated samples were selected and fitted to a normal distribution. The standard deviations from each of these studies were used to estimate the uncertainty in the distribution. The presence/absence data for each food category were then used to create a frequency distribution of contamination at the 0.04 cfu/g level. A normal curve with the appropriate standard deviation was then fit to the presence/absence data by "sliding" the mean until the percentage of positive samples corresponded to the presence/absence data. A normal curve for the log cfu/g was chosen because studies enumerating spoilage flora that are at sufficiently high levels to observe the curve showed that this distribution was appropriate (Kilsby and Pugh, 1981; Gill *et al*., 1996).

Step 1: Characterize the distribution of *Listeria monocytogenes* across food categories

Seventeen studies were selected for quantitative analysis (Table III-9). All of these studies had at least ten samples with enumerated values. The levels of *Listeria monocytogenes* in the

samples were transformed to log scale and the data for each study were fit using a normal distribution (Figure III-3). The mean level of *Listeria monocytogenes* (log cfu/g) and the standard deviation of the contamination data sets were calculated. This process was repeated for the 17 studies with adequate enumeration data.

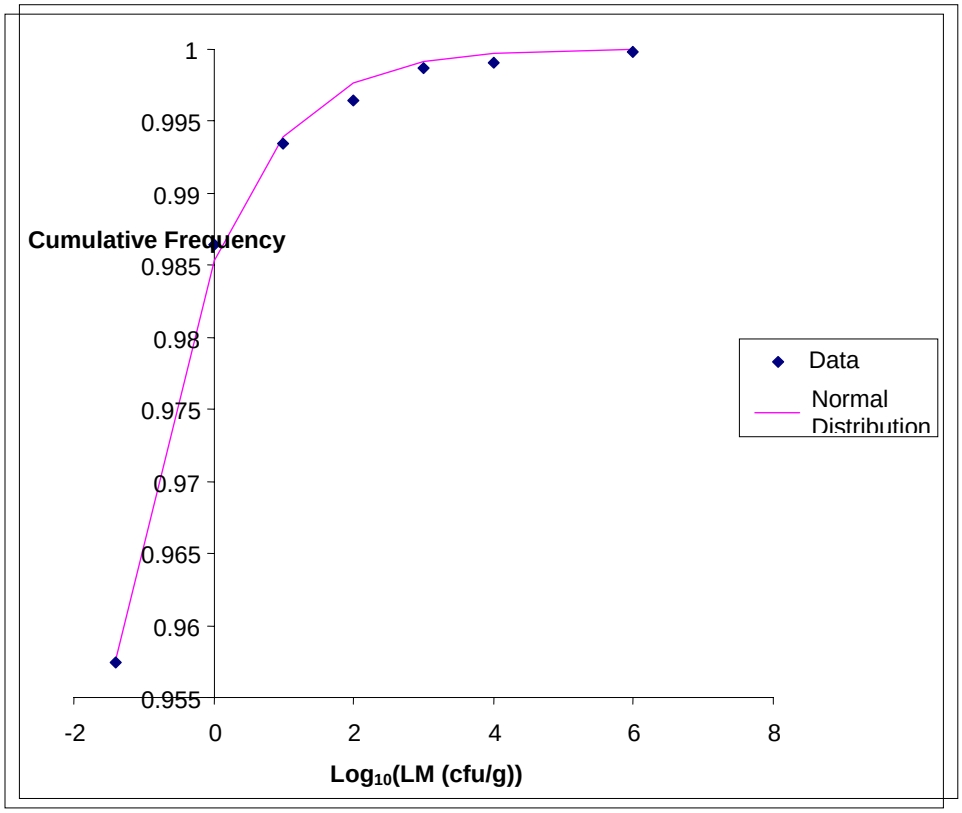


Figure III-3. A Lognormal Distribution for *Listeria monocytogenes* in Smoked Seafood

The standard deviations and mean levels of *Listeria monocytogenes* (log cfu/g) are summarized in Table III-9**.** The standard deviations of the distribution for each study ranged from 1.1 to 10.7 although most were less than 5.0.

Table III-9. Selected Studies Used to Characterize the Distribution of *Listeria monocytogenes* in Food at Retail

Study Reference[a]	Food Category	Number of Samples		Calculated Mean Level LM[a] (log cfu/g)	Estimated Standard Deviation[b]
		Total Tested	Total Positive		
Rawles, 1995	Cooked RTE Crustaceans	126	10	-9.9	6.4
NFPA, 2002	Deli Meat (CA)	4600	28	-12.2	4.3
NFPA, 2002	Deli Meat (MD)	4599	54	-7.7	2.8
WNYJWG, 1991	Deli-type Salad	149	21	-12.5	10.7
NFPA, 2002	Deli-type Salad (CA)	5504	126	-4.2	1.4
NFPA, 2002	Deli-type Salad (MD)	5606	191	-4.3	1.6
Hayes, *et al*., 1992	Frankfurter	40	12	-1.9	1.1
Morris and Ribeiro, 1991	Pâté	73	37	-1.2	4.0
Morris and Ribeiro, 1992	Pâté	216	75	-2.9	3.9
Jørgensen and Huss, 1998	Preserved Fish	91	23	-4.6	5.3
NFPA, 2002	Semi-soft Cheese	1623	23	-5.6	1.9
Cortesi, *et al*., 1997	Smoked Seafood	165	32	-4.4	3.5
Jørgensen and Huss, 1998	Smoked Seafood	420	163	-2.1	2.8
Dominguez *et al*., 2001	Smoked Seafood	170	38	-4.8	4.6
NFPA, 2002	Smoked Seafood	2687	114	-6.7	3.1
Loncarevic *et al*., 1995	Soft Ripened Cheese	31	13	-2.0	3.9
NFPA, 2002	Vegetables	2963	22	-8.9	3.1

[a]NFPA = National Food Processors Association; WNYJWG = West and North Yorkshire Joint Working Group; LM = *Listeria monocytogenes.*
[b]Standard Deviation of the log data.

These standard deviations were used to characterize the variation and uncertainty of the distribution of *Listeria monocytogenes* concentration in the food categories. The ranges of standard deviations used are given in Table III-10. A default range of 2 to 5 standard deviations was used for all food categories unless additional information was available to refine the uncertainty. Refined standard deviation ranges were used for four food categories (smoked seafood, pâté and meat spreads, deli meats, and deli-type salads) based on information as described in Table III-10. For example, the range of standard deviations assigned to Smoked

Seafood is narrower than the default range based on consideration of the standard deviations from four enumeration studies for this food category.

Table III-10. Standard Deviation Ranges for Each Food Category

Food Category	Standard Deviation Range	Comment
Default	2 to 5	This range was used as a default for all food categories (except Smoked Seafood, Pâté and Meat Spreads, Deli Meats, and Deli-type Salads) for which there was little or no empirical basis for estimating a distribution.
Smoked Seafood	2.8 to 4.6	This range encompasses the range for the four enumeration studies of smoked seafood samples.
Pâté and Meat Spreads	3.8 to 4.8	The standard deviation values for these products fit in a relatively narrow range and were generally higher than for other food categories.
Deli Meats	3.8 to 4.8	The standard deviation values for these meat products fit in a relatively narrow range and were generally higher than for other food categories
Deli-type Salads	1.5 to 2.5	The standard deviations for Deli Salads from the 2002 NFPA study were low (1.4; 1.6) in samples collected from both California and Maryland. A much higher value (10.7) was indicated by West Yorkshire study conducted 20 years ago in the U.K. Since the latter study is probably less representative of the current United States food supply, it was acknowledged by slightly raising the maximum range indicated by the NFPA study.

Step 2: Characterize the Uncertainty Distribution

The set of presence/absence studies for each food category was used to generate a discrete uncertainty distribution (a histogram) for the frequency of detectable contamination. First, the presence/absence data were used to generate a single estimate of the fraction of positive samples (i.e., a rate-concentration estimate) for each study. The concentration level was equal to the detection limit of the analysis (typically 0.04 cfu/g; based on 1 organism per 25 g sample). Next, the individual studies were adjusted (weighted) to account for sample size, geographic region of food origin, and date of collection. In addition, some data sets were obtained by sampling at the manufacturer instead of at retail. These data sets were adjusted to allow for growth between

manufacture and retail. With this adjustment the data collected at manufacture would then have the same percentage of positive samples but they were assigned higher cfu/g values.

Adjust for sample size, geographic location, and study date

The relevance of a particular contamination data set to represent current United States retail foods for the purposes of this risk assessment was a difficult judgment. If abundant, quantitative, recent and United States data were available, only this data would be used in the risk assessment. However, for most food categories these data were not available. Therefore, all data sources were used and weights were assigned to each data set so that the more relevant sets were given greater importance in this risk assessment. These weights were obtained from a panel comprised of government subject matter experts (Carrington and Dennis, 2001).

The individual studies were weighted by sample size, geographic region, and study date as follows in Equation 1.

$$\text{Study Weight} = n * gw * dw \qquad \text{Equation [1]}$$

Where:

n is the total number of samples in the study. A larger study would provide a better estimate of the percentage of positive samples than a small study.

gw is the geographic weight. A value of 1 was used unless the study was conducted in a region and food category for which there is little or no contribution (importation) to the United States food supply, in which case a value of 0.3 was used.

dw is the weight for the date of the study. Evidence exists that improved sanitation and HACCP programs have reduced the contamination of foods since the recognition of the public health problem from *Listeria monocytogenes* in the 1980's. A value of 1 was used for studies published within the past three years, a value of 0.7 was used for studies published between 1993 and 1999, while a value of 0.4 was used for studies published before 1993.

The width of the probability interval assigned to each study was proportional its relative weight as shown in Equation 2.

$$\text{Study Probability} = \text{Study Weight} / \text{Total Weight} \qquad \text{Equation [2]}$$

where Total Weight is the sum of all the Study Weights for the food category.

Adjustment of older data for food categories without large recent studies

About half of the food categories had large studies that were conducted within the past three years. As a result of the weighting scheme used to weight the studies, these recent studies usually received at least half the probability interval, dominating the analysis. Ten food categories had only older studies and those studies tended to have higher prevalence rates. The higher prevalence ranges may result from higher actual contamination levels or non-representative sampling. In either case, the data may tend to overestimate current *Listeria monocytogenes* concentrations, thereby biasing these categories compared to categories with recent data. To represent the uncertainty of this bias, the impact of large new studies on prevalence of *Listeria monocytogenes* was evaluated (Table III-11). Ratios were calculated by dividing the weighted pooled prevalence of 1999 and earlier data (percentage positive samples) by the weighted pooled prevalence of data for all years. A ratio less than 1 indicates that the prevalence of contaminated samples is currently higher than in the past. The reduction ratio values were used to adjust the food categories for which recent, large studies were not available. Specifically, the set of values in Table III-11 were used as an uncertainty distribution to reduce the number of positive values from older studies in categories without newer data. The food categories adjusted with the ratios to account for the lack of newer data include: Preserved Fish, Cooked RTE Crustaceans, Fruits, Hard Cheese, Processed Cheese, and Cultured Milk Products.

Table III-11. Prevalence Reduction Ratios for *Listeria monocytogenes* Using Study Age

Food Category	Prevalence Reduction Ratio[a]
High Fat and Other Dairy Products	0.9
Raw Seafood	1.0
Fluid Milk, Unpasteurized	1.0
Soft Ripened Cheese	1.8
Semi-soft Cheese	1.8
Vegetables	2.1
Deli-type Salads	2.3
Fluid Milk, pasteurized	2.6
Deli Meats	3.4
Fresh Soft Cheese	8.7
Frankfurters	9.7
Ice Cream and Frozen Dairy Products	31.3

[a]Prevalence reduction ratio = percentage of positive samples from data collected prior to 1999 divided by the total data set for each food category.

Adjustment for growth between production and retail for samples taken at manufacturing/ production

Some studies collected samples at manufacturing/ production prior to the point of retail (see Appendix 7). Since growth can be anticipated between production and purchase, the prevalence of positive samples for those data sets from sampling at manufacture were adjusted with estimates derived from the growth models (see section, "Modeling: Exponential Growth Rates").

The temperature ranges and storage times for the food categories are presented in Table III-12. These values were estimated as likely to be encountered between manufacture and retail. Because the distributions are narrow, rectangular distributions were used for storage time and for the temperature range. The median value from the growth models were used to adjust the contamination level but not the frequency of the presence/absence data. If, for example, the estimated growth was 0.5 logs prior to retail, a study with 5% positive at 0.04 cfu/g (-1.394 log) at manufacture would become 5% positive at 0.13 cfu/g (-0.884 log) at retail [0.5 log + -1.394 log = -0.894 log]. The contamination level was therefore increased from 0.04 cfu/g to 0.13 cfu/g to account for the possible growth of *Listeria monocytogenes* in food between production and retail.

Table III-12. Estimated Storage Temperature and Duration Between Manufacture and Retail and Predicted Median Growth

Food Category	Temperature Range[a] (°C)	Storage Time[a, b] (days)		Median Growth [c]
		Minimum	Maximum	(log cfu)
SEAFOOD				
Smoked Seafood	1 to 5	10	30	1.08
Raw Seafood	1 to 5	1	3	0.11
Preserved Fish		**Not applicable[b]**		
Cooked RTE Crustaceans	1 to 5	1	3	0.28
PRODUCE				
Vegetables	1 to 5	1	10	0.10
Fruits		**Not applicable[b]**		
DAIRY				
Fresh Soft Cheese		**Not applicable[b]**		
Soft Unripened Cheese		**Not applicable[b]**		
Soft Ripened Cheese	1 to 5	10	30	0.04
Semi-Soft Cheese		**Not applicable[b]**		
Hard Cheese	1 to 5	10	45	-0.94
Processed Cheese		**Not applicable[b]**		
Pasteurized Fluid Milk	1 to 5	1	3	0.20
Unpasteurized Fluid Milk		**Not applicable[b]**		
Ice Cream and Frozen Dairy Products		**Not applicable[b]**		
Cultured Milk Products		**Not applicable[b]**		
High Fat and Other Dairy Products	1 to 5	3	10	0.24
MEATS				
Frankfurters	1 to 5	10	30	1.03
Dry/ Semi-dry Fermented Sausage		**Not applicable[b]**		
Deli Meats	1 to 5	10	30	1.86
Pâté and Meat Spreads	1 to 5	1	7	0.34
COMBINATION FOODS				
Deli-type Salads		**Not applicable[b]**		

[a] Rectangular distributions were used for both the temperature range and storage times.
[b] Not applicable because none of the samples were collected at manufacture so growth between manufacture and retail was not calculated for these food categories.
[c]Median growth (log cfu) is calculated by multiplying the storage times and the exponential growth rates (see Section "Modeling: Growth Between Retail and Consumption").

Step 3: Integration of Prevalence Data and Quantitative Analysis

Frequency distributions for *Listeria monocytogenes* concentration for each food category were generated by integrating the standard deviation estimates with the rate estimates for detectable *Listeria monocytogenes*. This was accomplished with a 300 iteration simulation in which pairs of values were randomly selected from a uniform distribution of the standard deviations (Table III-10) and the weighted collection of the presence/absence data sets for each food category (including those at 0.04 cfu/g at retail and those adjusted for pre-retail growth). For each of the 300 pairs of values, a mean of the log cfu/g value was calculated (using the Excel Goal Seek procedure) to find the geometric mean that matches the cumulative frequency of positive samples at the detection limit of the assay (0.04 cfu/g or the adjusted value) with the selected standard deviation. Therefore, for each food category, 300 contamination curves were generated. The average frequency for each contamination level was determined to create the variability of contamination levels. The standard deviation of the frequencies for each contamination level became the uncertainty of the distribution for the contamination data.

Example of the Modeling for *Listeria monocytogenes* in Food at Retail Using Smoked Seafood

Step 1. Characterize the distribution of *Listeria monocytogenes* across food categories

Data from NFPA (2002) for Smoked Seafood is used to illustrate this step. As shown in Figure III-3, at the 0.04 cfu/g (-1.4 on log scale) contamination value, 0.958 (95.8%) of the samples (2573/2687) contain less than or equal to that contamination level. Sixty-seven more samples had levels < 0.1 cfu/g and eleven samples were contaminated at less than or equal to 1 cfu/g (0.0 on log scale). Therefore the fraction of negative samples is 0.986 [(2573 + 67 + 11)/2687]. This procedure is repeated for the samples that had higher levels of contamination. A normal curve was fitted to the data points by least-squares and the mean and standard deviation were estimated as –6.7 and 3.1, respectively. This process was repeated for the 17 selected enumeration studies and the resulting means and standard deviations are summarized in Table III-9.

Step 2. Characterize the uncertainty distribution for the frequency of detectable contamination

- Adjust for sample size, geographic location, and study date. The study weight and study probability are calculated as described by Equations 1 and 2 using the total number of samples in the study (n), the geographic weight (gw), and the weight for the date of the

study (dw). These values are shown for Smoked Seafood in Table III-13. For example for the Aguado *et al*., 2001 study, the study weight is 52 (52 x 1 x 1) and the study probability is 0.009 (52/6034.7).

- Adjustment of older data for food categories without large recent studies. This step is not applicable for smoked seafood as recent large studies were available. However an adjustment was made using the range of prevelance ratios given in Table III-11 for Preserved Fish, Cooked RTE Crustaceans, Fruits, Hard Cheese, Processed Cheese, and Cultured Milk Products.

Adjustment for growth between production and retail for samples taken at manufacturing. In Table III-13 the 'collection' column indicates which studies were collected at manufacturing/ product and at retail. For the studies collected prior to retail, the level of *Listeria monocytogenes* was increased to account for anticipated growth between manufacturing and retail. From Table III-12, the mean exponential growth for smoked seafood of 0.15 logs/day at 5°C was multiplied by a uniform distribution (minimum of 1 day, most frequent of 10 days, and maximum of 30 days of storage) and the median of this resulting distribution was 1.08 logs. The fraction of positive samples (0.04 cfu/g or -1.4 log cfu/g) at manufacture was increased to a fraction of positive samples with a value of 0.48 cfu/g (-0.32 log cfu/g) at retail (-1.4 log + 1.08 log = -0.32 log cfu/g). In Step 3 described below, the procedure for the fitting of the contamination distribution the fraction of positive samples remained the same but the contamination level was now represented by a value of –0.32 log cfu/g for these studies.

Table III-13. Prevalence Studies of *Listeria monocytogenes* in Smoked Seafood

Study Reference	n[a]	# neg[b]	gw[c]	dw[d]	Collection[e]	Study Weight[f]	Cumulative Probability[g]	LM% negative[h]
Aguado *et al*., 2001	52	36	1	1	R	52	0.009	0.69
Baek *et al*., 2000	68	65	1	1	R	68	0.020	0.96
Cortesi *et al*., 1997	165	133	1	0.7	R	115.5	0.039	0.81
Dauphin *et al*., 2001	36	20	1	1	R	36	0.045	0.56
Dillon *et al*., 1994	258	246	1	0.7	R	180.6	0.075	0.95
Dominguez *et al*., 2001	170	132	1	1	R	170	0.103	0.78
Eklund *et al*., 1995	61	13	1	0.7	P	42.7	0.110	0.21
Ericsson *et al*., 1997	9	6	1	0.7	R	6.3	0.111	0.67
Farber, 1991b	32	22	1	0.4	P	12.8	0.113	0.69
Garland, 1995	285	284	1	0.7	P	199.5	0.146	1.00
NFPA, 2002	2687	2573	1	1	R	2687	0.592	0.96
Guyer and Jemmi, 1990	64	60	1	0.4	P	25.6	0.596	0.94
Hartemink and Georgsson, 1991	31	30	1	0.4	R	12.4	0.598	0.97
Heinitz and Johnson, 1998	1080	929	1	0.7	P	432	0.669	0.86
Hudson *et al*., 1992	26	13	1	0.4	R	10.4	0.671	0.50
Inoue *et al*., 2000	92	87	1	1	R	92	0.686	0.95
Jemmi, 1990	820	732	1	0.4	R	328	0.741	0.89
Jørgensen and Huss, 1998	420	257	1	0.7	R	294	0.790	0.61
Maija *et al*., 2001	232	222	1	1	R	232	0.828	0.96
Miettinen, *et al*., 2001	25	22	1	1	R	25	0.832	0.88
Ng and Seah, 1995	2	1	1	0.7	R	1.4	0.832	0.50
Norton *et al*., 2000	38	32	1	1	P	38	0.839	0.84
Norton *et al*., 2001	96	85	1	1	P	96	0.855	0.89
Oregon State Dept of Agriculture, 2001	168	167	1	1	R	168	0.882	0.99
Scoglio *et al*., 2000	21	18	1	1	R	21	0.886	0.86
Teufel and Bendzulla, 1993	380	353	1	0.4	R	152	0.911	0.93
Vogel *et al*., 2001a	324	231	1	1	P	324	0.965	0.71
Vogel *et al*., 2001b	200	65	1	1	P	200	0.998	0.33
Yamazak *et al*., 2000	13	10	1	1	R	13	1.000	0.77
TOTAL						6034.7		

[a] n = total number of samples in the study

[b] # neg= total number of non-detectable samples in the study (i.e., <0.04 cfu/g)

[c] gw= geographic weight. A value of 1 was used unless the study was conducted in a region and food category for which there is little or no contribution (importation) to the United States food supply, in which case a value of 0.3 was used.

[d] dw= weight for the date of the study. A value of 1 was used for studies published within the past three years; a value of 0.7 was used for studies published between 1993 and 1999; and a value of 0.4 was used for studies published before 1993.

[e] Collecction. R= sample collected at retail; and P = sample collected at production/ manufacturing

[f] Study weight = n x gw x dw

[g] Cumulative probability.

[h] LM% negative = percentage of *Listeria monocytogenes* below the method of detection (i.e., <0.04 cfu/g)

Step 3: Integration of Prevalence Data and Quantitative Analysis

The *Listeria monocytogenes* concentration model for Smoked Seafood is presented in Figure III-4. The model estimates are compared to the prevalence studies and the enumeration data. The median (50th percentile), lower (5th percentile) and upper (95th percentiles) bounds reflect the *Listeria monocytogenes* concentration model (i.e., the set of Lognormal disitribution parameter values). Each data point in the "Prevalence Studies" data set represents an individual study (weighted for sample size and other study characteristics as described in Step 2). The data points in the "Enumeration Studies" data set are pooled from four different studies as noted in Table III-5. The prevelance studies at the –0.32 log cfu/g level represent the studies collected at manufacturing/ production and were adjusted for potential growth between production and retail.

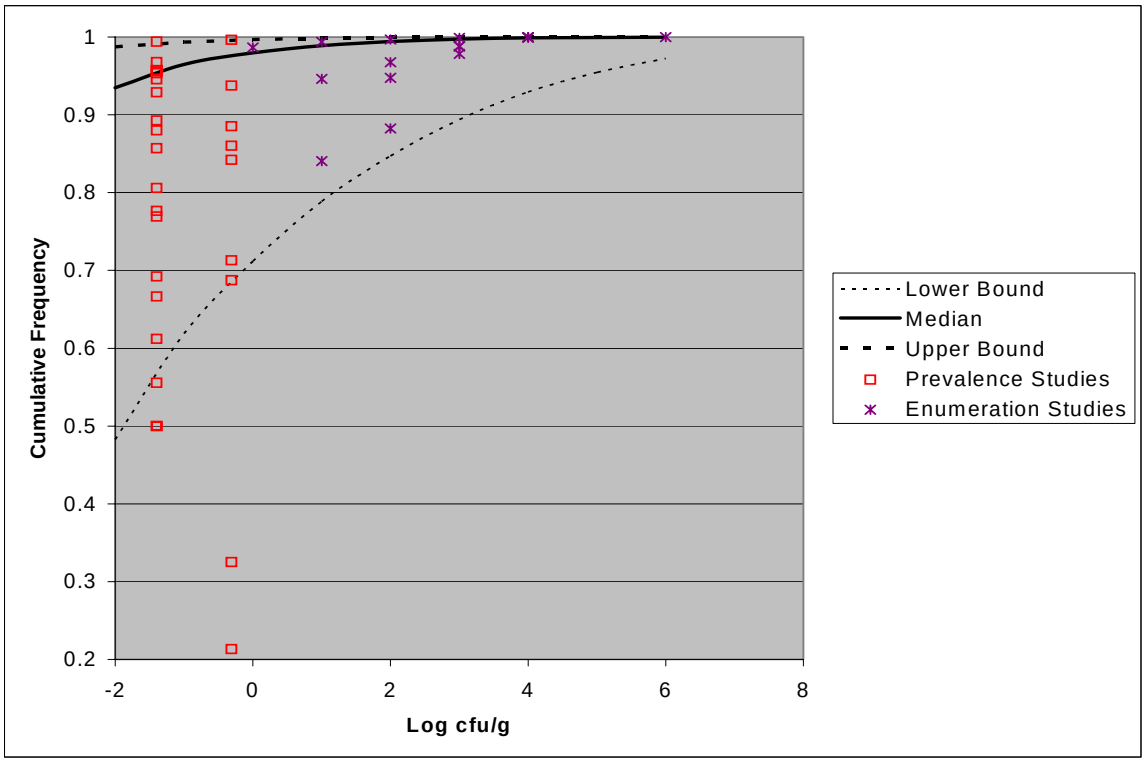


Figure III-4. Modeled Contamination Data for Smoked Seafood Food Category

Results: Modeled Contamination at Retail

Table III-14 shows the modeled distributions for *Listeria monocytogenes* contamination for the 23 food categories at retail. The first column of data in Table III-14 provides the median percentage of servings with less than one organism per serving, this estimate is not the same as undetectable values (<0.04 cfu/g) because different foods have different serving sizes. The predicted median of the servings having less than one organism of *Listeria monocytogenes* per serving ranged from 91.3 to 99.9% for the various food categories. In other words, less than 0.1 to 8.7% of the servings had one or more *Listeria monocytogenes* per serving, depending on the food category. The 5^{th} and 95^{th} percentiles provide information to estimate the uncertainty distributions for each of these median values. Although some servings of all food categories are likely to be contaminated at the retail level, servings of certain food categories (*e. g.,* Smoked Seafood, Raw Seafood, Deli Meats, Dry/Semi-Dry Fermented Sausages, and Deli Salads) were the most likely to be contaminated. Other columns in Table III-14 provide the percentage of servings with higher levels of contamination. Most frequently, the food categories are contaminated with 1 to 1000 cfu/serving. The calculations in the risk assessment model used 0.5 log intervals (referred to as bins) instead of the 3 log intervals shown in Table III-14.

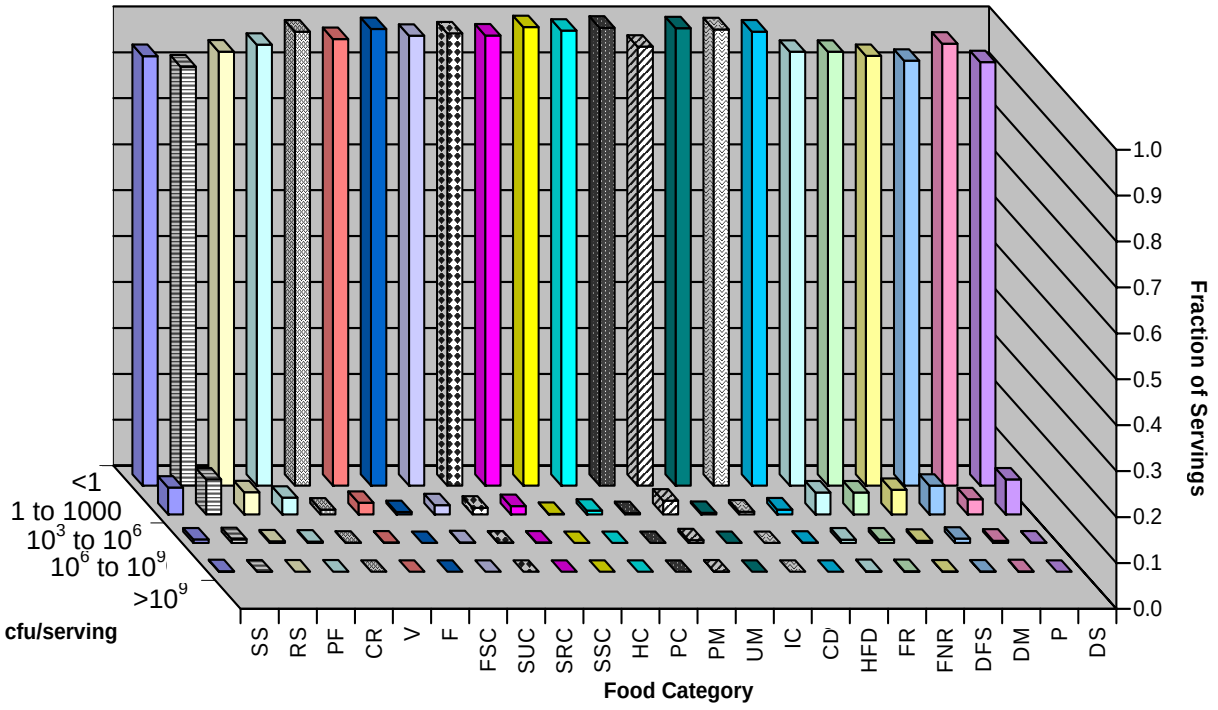
The bar chart in Figure III-5 provides a graphic depiction of the modeled distributions. Most of the servings for each food category are in the <1 cfu/serving level (back row of bars). As the level of contamination per serving rises (moving into the front rows of bars), the fraction of servings decreases markedly for most of the food categories.

Thus, for the Smoked Seafood category, the fraction of servings at <1, 1 to 10^3, 10^3 to 10^6, 10^6 to 10^9, and >10^9 cfu/serving are about 93.6, 5.8, 0.8, 0.1, and 0.0% of servings, respectively. The sum of the fractions of servings for a food category do not necessarily equal 100% because of rounding and because adding medians is not mathematically correct.

Table III-14. Modeled Percentage Distribution of Food Servings Contaminated with *Listeria monocytogenes* at Retail

Food Category	Median Percentage of Servings Contaminated at Different Levels									
	<1 cfu/serving		1 - 1000 cfu/serving		10^3 - 10^6 cfu/serving		10^6 - 10^9 cfu/serving		> 10^9 cfu/serving	
	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]
Seafood										
Smoked Seafood	93.6	(51.6, 98.7)	5.8	(0.9, 28.5)	0.8	(0.1, 12.8)	0.1	(<0.1, 5.9)	<0.1	(<0.1, 0.1)
Raw Seafood	91.3	(87.2, 98.6)	7.6	(1.3, 11.4)	0.8	(0.1, 1.7)	<0.1	(<0.1, 0.3)	<0.1	(<0.1, <0.1)
Preserved Fish	94.5	(70.8, 99.8)	4.8	(0.2, 20.4)	0.4	(<0.1, 4.1)	<0.1	(<0.1, 0.8)	<0.1	(<0.1, <0.1)
Cooked Ready-to-Eat Crustaceans	96.0	(93.9, 97.0)	3.6	(2.7, 6.0)	0.3	(0.1, 0.6)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
Produce										
Vegetables	98.9	(98.7, 99.0)	1.1	(0.9, 1.3)	0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Fruits	97.3	(70.4, 99.8)	2.5	(0.2, 22.0)	0.1	(<0.1, 6.8)	<0.1	(<0.1, 1.3)	<0.1	(<0.1, <0.1)
Dairy										
Fresh Soft Cheese	99.5	(95.1, 99.7)	0.5	(0.3, 4.8)	<0.1	(<0.1, 0.5)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
Soft Unripened Cheese,	98.0	(90.0, 99.9)	2.0	(0.1, 8.6)	0.2	(<0.1, 3.3)	<0.1	(<0.1, 0.7)	<0.1	(<0.1, <0.1)
Soft Ripened Cheese	98.5	(83.4, 99.9)	1.4	(0.1, 13.4)	0.1	(<0.1, 2.9)	<0.1	(<0.1, 0.4)	<0.1	(<0.1, <0.1)
Semi-soft Cheese	98.0	(90.8, 98.6)	1.8	(1.2, 7.2)	0.1	(<0.1, 1.5)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)
Hard Cheese	99.9	(97.8, 100.0)	0.1	(<0.1, 2.0)	<0.1	(<0.1, 0.2)	<0.1	(<0.1,< 0.1)	<0.1	(<0.1, <0.1)
Processed Cheese	99.1	(97.5, 99.9)	0.8	(0.1, 2.4)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Pasteurized Fluid Milk	99.7	(97.8, 99.9)	0.3	(0.1, 2.0)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Unpasteurized Fluid Milk	96.1	(90.0, 100.0)	3.3	(<0.1, 8.5)	0.3	(<0.1, 4.0)	<0.1	(<0.1, 0.9)	<0.1	(<0.1, 0.1)
Ice Cream and Frozen Dairy Products	99.6	(99.3, 99.8)	0.4	(0.2, 0.6)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Cultured Milk Products	99.4	(94.0, 99.9)	0.6	(0.1, 5.5)	<0.1	(<0.1, 0.5)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
High Fat and Other Dairy Products	98.9	(98.3, 99.1)	1.1	(0.7, 1.7)	0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Meats										
Frankfurters (reheated)	94.5	(88.5, 95.5)	4.8	(3.6, 9.4)	0.7	(0.7, 2.0)	0.1	(0.1, 0.5)	<0.1	(<0.1, <0.1)
Frankfurter (not reheated)	94.5	(88.5, 95.5)	4.8	(3.6, 9.4)	0.7	(0.7, 2.0)	0.1	(0.1, 0.5)	0.1	(<0.1, <0.1)
Dry/Semi-dry Fermented Sausages	93.6	(77.7, 97.6)	5.4	(2.1, 19.7)	0.5	(<0.1, 4.1)	<0.1	(<0.1, 1.1)	<0.1	(<0.1, <0.1)
Deli Meats	92.5	(87.8, 99.3)	6.3	(0.7,11.1)	1.0	(<0.1, 1.3)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)
Pâté and Meat Spreads	96.2	(79.7, 98.0)	3.3	(1.8, 14.9)	0.5	(0.2, 4.5)	0.1	(<0.1, 1.2)	<0.1	(<0.1, <0.1)
Combination Foods										
Deli-type Salads	92.2	(86.5, 97.7)	7.6	(2.3, 13.3)	0.1	(<0.1, 0.4)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)

[a] The 5th and 95th percentiles uncertainty levels, respectively.



LEGEND			
SS =	Smoked Seafood	PC =	Processed Cheese
RS =	Raw Seafood	PM =	Pasteurized Fluid Milk
PF =	Preserved Fish	UM =	Unpasteurized Fluid Milk
CR =	Cooked Ready-To-Eat Crustaceans	IC =	Ice Cream and Frozen Dairy Products
V =	Vegetables	CD=	Cultured Milk Products
F =	Fruits	HFD	High Fat and Other Dairy Products
FSC =	Fresh Soft Cheese	FR =	Frankfurters (reheated)
SUC =	Soft Unripened Cheese	FNR =	Frankfurters (not reheated)
SRC =	Soft Ripened Cheese	DFS =	Dry/Semi-Dry Fermented Sausages
SSC =	Semi-soft Cheese	DM =	Deli Meats
HC =	Hard Cheese	P =	Pâté and Meat Spreads
		DS =	Deli Salads

Figure III-5. Modeled Distribution of *Listeria monocytogenes* Contamination Levels in Food Servings at Time of Retail

Modeling: Growth Between Retail and Consumption

Most of the contamination data used in this risk assessment were from samples collected at retail. Because *Listeria monocytogenes* can grow slowly at refrigeration temperatures, a growth module was incorporated into the exposure assessment to account for the potential growth of the organism in the food during storage in the home, prior to consumption. The growth model provides an estimate of the numbers of *Listeria monocytogenes* in the food at the time of consumption.

The growth model included the initial level of *Listeria monocytogenes* in the foods at retail where the food is purchased, the storage temperature in the home refrigerator, the exponential growth rate of *Listeria monocytogenes* in a food stored at a specific temperature, the storage time in the home and the maximum growth (stationary phase). Inoculated food studies, where growth of *Listeria monocytogenes* inoculated into a food was measured, showed that maximum growth at low refrigeration temperatures (<5°C) was often less than growth in the same foods at higher temperatures. It was also concluded that refrigeration temperature and storage time are not independent factors. High storage temperatures and long storage times would not be likely to occur because this combination would lead to obvious spoilage and the food would not be consumed. The output from the growth model was a frequency distribution of the log cfu/g for each food category at the time of consumption.

Exponential Growth Rates

The square root model for exponential growth rate (EGR) was chosen because of its simplicity and general acceptance as indicated by the documented use in the microbiology literature (Ratkowsky *et al.*, 1982). A straight line results when the square root of the EGR is graphed for different growth temperatures. The equation for the model is:

$$\sqrt{EGR} = a(T - T_0) \qquad \text{Equation [3]}$$

where **EGR** is the exponential growth rate ($\log_{10}$ cfu/day), **T** is the growth temperature (°C), $\mathbf{T_0}$ is the extrapolated minimum notational growth temperature (°C), and **a** is the slope parameter for *Listeria monocytogenes* in the specific food. T_0 values were estimated from four sources (Alavi *et al*., 1999; Duh and Schaffner, 1993; USDA, 1997 Pathogen Modeling Program; Wijtzes *et al*., 1993) and an average of these values (-1.18°C) was used in the model.

Different storage temperatures were used in the studies from the published literature that reported growth of *Listeria monocytogenes* in various foods. Therefore, using the data from these studies, equivalent EGRs ($\log_{10}$ cfu/day) at 5°C were calculated. The equation, presented as Equation 4, is a ratio and rearrangement of Equation 3. The slope factor (a) is a constant and cancels out in the equation.

$$\frac{EGR_5}{EGR_{lit}} = \left\lceil \frac{a(T_5 + 1.18)}{a(T_{lit} + 1.18)} \right\rceil^2 = \left\lceil \frac{6.18}{(T_{lit} + 1.18} \right\rceil^2 \qquad \text{Equation [4]}$$

where:

$\mathbf{EGR_5}$ is the converted growth rate at 5°C,

$\mathbf{EGR_{lit}}$ is the growth rate from the inoculated pack study,

$\mathbf{T_5}$ is set to 5°C to standardize the EGRs, and

$\mathbf{T_{lit}}$ is the temperature used in the literature.

If a category had five or more data points, variation was modeled by fitting statistical distributions to the resulting values (using the software program ParamFit). Paramfit employs ten different distribution models: Beta, Cauchy, Exponential, Gamma, Logistic, Lognormal, Normal, Rectangular (Uniform), Triangular, and Weibull. There is no theoretical support for any one distribution to be more appropriate than any other distribution. Therefore, the range of values generated by each of the ten statistical distributions reflects the uncertainty.

The 10 distribution models are used to construct a probability tree for the predictive model. Within an iterative simulation, the frequency of use of each model is allocated according to its relative model weight which is calculated as follows:

$$\text{Model Weight} = (((1 + n / Pn)^{O}) \times ((1 - gof)^{H}) \qquad \text{Equation [5]}$$

where
n = number of observations
Pn = number of model parameters
gof = Goodness-of-Fit
O = an arbitrary constant to describe parameter penalty, a value of 19 was used
H = An arbitrary constant to modify and provide a better fit, a value of 141 was used

ParamFit uses least residual squares for the predicted percentiles as the optimization criteria. The ratio of the sum of residual squares to the sum of total squares for the predicted percentile is used as a goodness-of-fit statistic. This approach fits the middle of the distribution, so that outliers have less impact on the shape of the distribution.

In some food categories (such as Dry/Semi-dry fermented sausages and Deli-type Salads), the *Listeria monocytogenes* levels decline at a slow rate. The rate of decline was modeled with the same square root model (Equation 3) as for growth with a negative slope (**a**) and a negative EGR. Negative EGR values from the literature were combined with positive data to create one distribution, which was fitted to the growth models as explained earlier. The rate of decline was adjusted for temperature, after being converted to a positive value, by the same ratio method of Equation 4. Increasing the storage temperature above 5°C increases the rate of decline and conversely temperature decreases below 5°C decrease the rate of decline. This approach agrees with the USDA Pathogen Modeling Program (USDA, 1997), which predicts faster rates of decline at higher storage temperatures. This relatively simple approach to modeling growth versus decline (survival) sufficiently accounted for the relatively slow rates of declines encountered in this risk assessment.

If all of the growth values were positive, the data were fit with all ten distributions and the four with the highest weights were used in the probability tree. If some of the growth values were negative (reflecting a possible decline in *Listeria monocytogenes* numbers), then the data were

only fit with the Beta, Cauchy, Normal, Triangular, and Rectangular distributions as these are the only distributions of the ten that will accept negative values. Of these five distributions those with the three highest weights were used.

Several of the food categories had only two or three data points. Under this circumstance, probability trees were constructed with equiprobable rectangular or normal distribution. The maximum and minimum values were used as the parameters for the rectangular distribution. A standard algebraic formula was used to calculate the mean and standard deviation of the normal distribution.

Details on the variations and uncertainties used in the risk assessment for each food category are provided in Appendix 5. A value of zero for the EGR at all refrigeration temperatures is assigned to food categories that did not support growth (such as ice cream) and in which the pathogen levels remained stable over an extended period.

As an example, data from the Smoked Seafood food category (see Appendix 5) will be used to illustrate how the exponential growth rate of *Listeria monocytogenes* was calculated. Briefly, the data sets of EGR values at 5 °C are placed in order of ascending magnitude. Figure A5.1.2 (see Appendix 5) titled 'Cumulative Distribution for the Exponential Reference Growth Rate (EGR) at 5 °C,' is a cumulative frequency graph where the x-axis is the EGR in $\log_{10}$ cfu/day and the y-axis is the fraction of data points from the literature with that value or lower (values are from Appendix 8). Different statistical distributions are fitted to the cumulative frequency distribution with the residual sums of squares for each frequency distribution used to weight the distributions. The probability column from Table A5.1.6 (see Appendix 5) indicates the weights for the four best-fitting distributions. In this example, the Lognormal and Gamma distributions have 40 and 31% of the weight, respectively. The Beta and triangular distributions had poorer fits and carried relatively little weight (16 and 13%, respectively). The probability of each growth model dictates the frequency of selection of each distribution for use in each uncertainty iteration during a Monte Carlo simulation (Cassin, *et al*., 1998; Vose, 1998). The variation predominantly reflects the shape(s) of the most heavily weighted statistical distribution.

Post-Retail Storage Times

The distribution of storage times were multiplied by the EGR to provide an estimate of the amount of *Listeria monocytogenes* growth occurring between retail purchase of the food and its consumption. Some foods are consumed on the day of purchase whereas others remain in the home refrigerator for lengthy periods of time. This is a major source of variability in the estimate of growth and ultimately, in the numbers of *Listeria monocytogenes* consumed. The variation in storage time was described using a modified BetaPert distribution (Figure III-6). A BetaPert distribution is defined by minimum, most likely, and maximum values. The distribution was modified by increasing the weight for the central value from 4 to 7. This modification reduced the frequency of values in the extended tails. The storage times were not used in the modeling for foods where *Listeria monocytogenes* does not grow. The uncertainty was described using a ±20% uniform distribution for the most frequent value and a ±50% uniform distribution for the maximum value, with a 100% correlation between the two distributions.

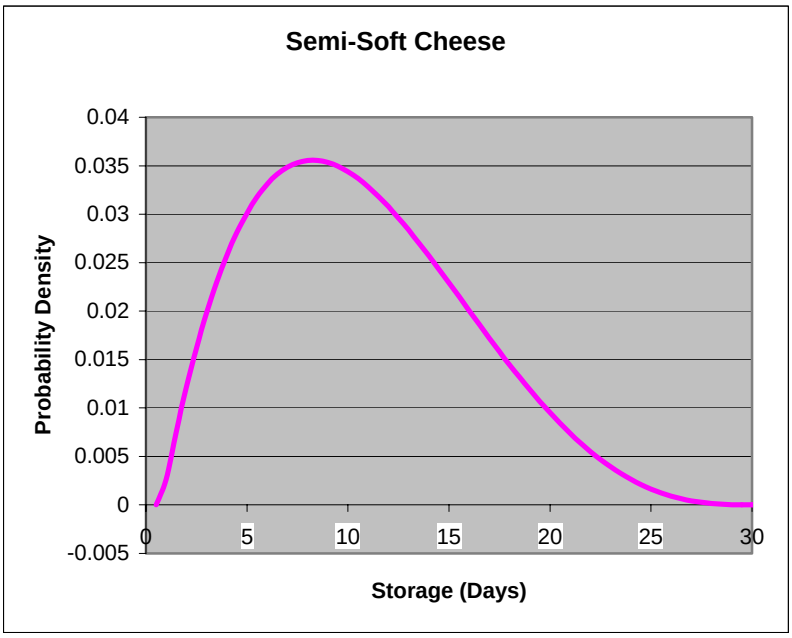


Figure III-6. Example of a Modified BetaPert Distribution

Frankfurters and Deli Meats

The survey sponsored by the American Meat Institute (AMI, 2001) asked for the average time consumers keep frankfurters and deli meats in the refrigerator. For example, 4% of the survey responders indicated that they stored frankfurters for an average of 11 to 14 days (Table III-6). This means that those responders consumed some individual servings of frankfurters after shorter storage times and others were kept longer than 14 days. While this is helpful information, what was needed for the model was the likely distribution of storage times for individual servings of frankfurters and deli meats. Thus, AMI data estimates inter-household variation. To get information on intra-household variation, consumers could be asked how long they stored the product the last time it was consumed. In order to introduce intra-household variation to the AMI data set, a log normal distribution was applied to the empirical AMI data points. The magnitude of the intra household variation, expressed as the Geometric Standard Deviation (GSD), ranged from 0.4 to 0.6 to be consistent with the data from the USDA/FSIS hotline study. The USDA hotline study asked for the 'last storage time' (Wachsmuth, 2000).

Figure III-7 shows a comparison of the USDA/FSIS hotline data (used in the draft risk assessment) and the AMI survey (indicated in the figure as 'individual average' data). The uncertainty bounds (GSD 0.2 to 0.6) are also shown in Figure III-7.

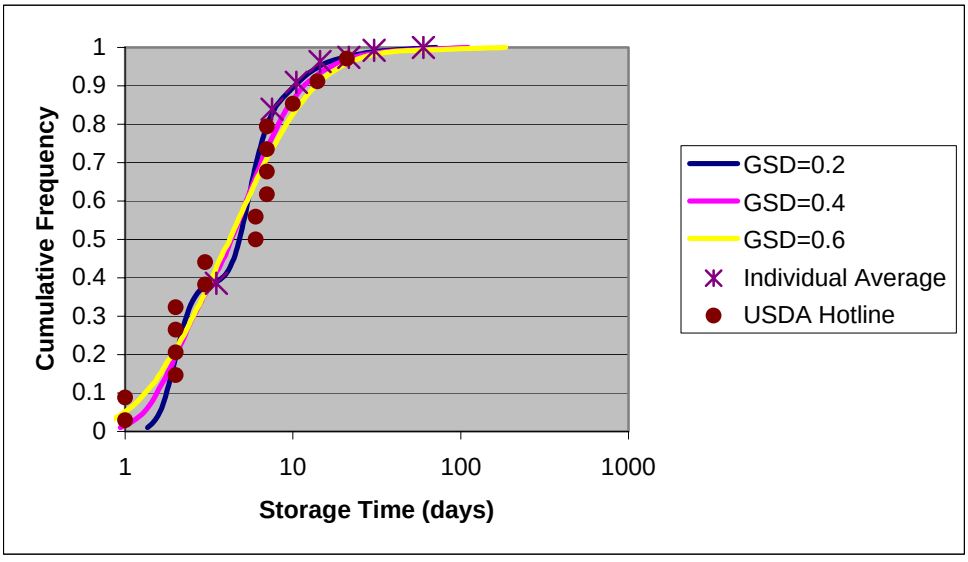


Figure III-7. Storage Time Distribution for Frankfurters and Deli Meats

Deli-type Salads

The data and assumptions behind growth estimates in deli salads were reexamined after the 2001 draft risk assessment. Data provided by Johnson (1993) and studies conducted in FDA's laboratories (Eblen, 2002a) showed that *Listeria monocytogenes* populations decline during the refrigerated storage of most deli foods. This is a consequence of processor-made salads having sufficient acidity and other preservatives to prevent growth. Locally- or store-made salads may not have these ingredients. The FDA studies indicated that growth only occurred in the shrimp and crab seafood salads. With the assistance of industry production data (Mitchell, 2001) the split between store-made and processor-made salads was estimated to be 15:85. It was also estimated that shrimp and crab salad are less than 10% of the total salad sales. Therefore, a triangular distribution of (1, 1.5, 3) was used to represent the fraction of deli salads that support growth and the uncertainty in that estimate. The growth rate at 5°C averaged 0.122 logs/day in the salads that supported growth and the declining rate averaged 0.143 logs/day in the majority of salads that did not support growth.

Modeling: Interaction of Storage Times and Temperatures

Increases in the levels of *Listeria monocytogenes* were calculated as the product of the EGR (which is dependent on the refrigeration temperature) and storage time. The Monte Carlo simulation program randomly selects different values from each calculated distribution. Both temperature and time distributions are concentrated toward the center of their ranges, 4°C and 8 days, respectively for Smoked Seafood. As a result, the most frequent estimates of growth would reflect these conditions. The simulation process would also select, at a lower frequency, the combination of low refrigeration temperatures and short storage times. Such combinations would result in relatively little growth. Similarly, the process could select high refrigeration temperatures and long storage times, 10°C and 45 days, which would result in extensive growth. However, this combination of temperatures and times would likely result in the food showing obvious spoilage and hence would not be consumed. Modeling the refrigeration temperature and storage time distributions as independent distributions was not believed to be appropriate. Therefore, the uncertainties in the mode and maximum storage times were negatively correlated

to the temperature. For example, for Smoked Seafood, this means the mode ranged from 6 to 10 days. When refrigeration temperature was 10°C, the time was 6 days and when the temperature was 0°C the time was 10 days. The maximum storage time similarly ranged from 15 to 45 days for 10°C and 0°C storage, respectively. This means that at higher temperatures the distribution for storage times is much compressed relative to the distribution at lower temperatures.

Maximum Growth Levels

Growth is the product of the EGR (at a specific temperature) and the storage time. For each iteration of the Monte Carlo simulation, the logarithm of growth estimated during storage was added to a contamination level at retail. No lag phase was calculated; it was assumed that the *Listeria monocytogenes* cells were already in the food and adjusted to the food's environment during the period before retail purchase. The only change made from retail to storage was to a new refrigerator temperature. This relatively small change would take several hours for a packaged food and the cells would effectively adjust as the temperature changes.

The populations for the stationary phases of *Listeria monocytogenes* in foods were obtained from the published literature and were used to establish limits for the maximum calculated growth levels for each food category (Appendix 8). If the calculated level for *Listeria monocytogenes* exceeded the maximum level, the maximum value was used. The literature also indicated that the maximum growth level is dependent upon the storage temperature. However, there were only a few studies in the literature that provided for the growth in a food to the stationary phase at more than one temperature to permit accurate estimation of this behavior.

Duffes *et al.* (1999) showed maximum levels (cfu/g) in smoked salmon to be less than 10^5 at 4°C and $10^{8.1}$ at 8°C. Pelroy *et al.* (1994a) found maximum levels in smoked salmon to be 10^5 and $10^{6.5}$ at 5 and 10°C, respectively. Maximum populations were reported in cream as 10^7 and $10^{7.5}$ at 4 and 8°C, respectively (Rosenow and Marth, 1987); in butter it was reported as $10^{5.5}$ and 10^6 at 4 to 6 and 13°C, respectively (Olsen *et al*, 1988); and in lettuce it was reported as 10^5 to $10^{5.5}$ and $10^{6.5}$ to $10^{7.5}$ at 5 and 10°C, respectively (Beuchat and Brackett, 1990b). In addition to direct

comparisons, a collection of individual growth studies also showed this tendency to grow to higher population levels at higher temperatures.

The maximum growth levels (cfu/g) used were applied across all food categories with 10^5, $10^{6.5}$ and 10^8 used as maximums for temperatures of <5, 5 to 7 and >7°C, respectively. For milk, sufficient data were found in the literature for growth levels of 10^7, $10^{7.5}$ and 10^8, to use as the maximums for the three temperatures, respectively. A uniform range of one logarithm was used to represent the uncertainty for each of the maximum growth levels. The calculated growth levels were added to the retail contamination levels during each iteration of the model, and these new levels of *Listeria monocytogenes* contamination in food were compared to the maximum growth level. If the calculated growth level exceeded the maximum growth level in any iteration, the amount of growth was reduced to the maximum growth level.

Modeling: Thermal Inactivation

Frankfurters have been implicated in outbreaks of listeriosis although they are generally reheated before serving. Because they are precooked during manufacturing, frankfurters are considered to be a RTE food. Reheating will kill *Listeria monocytogenes* in food. Frankfurters are usually, but not always, reheated before consumption. Therefore, a thermal inactivation step was included in the model for frankfurters. The frequency of insufficient reheating and the extent of inactivation of *Listeria monocytogenes* when not properly reheated were estimated in this step of the model.

No data describing the prevalence or extent of under-reheating of frankfurters has been published. However, the Georgetown survey (n=90) found approximately 1% of the respondents did not reheat their frankfurters (Wachsmuth, 2000). In an FSIS Hotline survey, 14% of the respondents indicated that at least one household member has eaten frankfurters directly from the package (Wachsmuth, 2000).

Some frankfurters are frozen by the consumer when they are brought home from the retail store. Information on the proportion of frozen frankfurters from the AMI survey and FDA Food Safety survey (Lando, 2003) led the risk assessment team to assign a uniform distribution from 3.0 to 8.7 % to represent this proportion and its uncertainty. To the frozen portion of frankfurters, the growth of *Listeria monocytogenes* would be set to zero, that is, the bacteria don't grow or die during the frozen storage. The time of storage would be irrelevant. It was assumed that all of the frozen frankfurters would be reheated before consumption. Therefore, the distribution of *Listeria monocytogenes* inactivation used for part of the non-frozen frankfurters was applied to all of the frozen frankfurters.

The final distribution of *Listeria monocytogenes* consumed per serving in reheated frankfurters is the summation of the respective proportions of the frankfurters stored frozen and reheated and the frankfurters stored refrigerated and reheated. The number of cases per annum was calculated from the total number of frankfurter servings and the proportion of the total in these two groups. The distribution of *Listeria monocytogenes* consumed per serving in non-reheated frankfurters represents the remaining proportion, represented by a triangular distribution of (4, 7, 10) percent of the non-frozen frankfurter servings (uncertainty distribution).

It was recognized that frankfurters are reheated in boiling water and microwave ovens more frequently than grilling, and that frankfurters are more likely to be contaminated on the surface than the interior. The Georgetown survey showed that 20% of the frankfurters were microwaved; the percentage of all responses for the FSIS Hotline was 19.4% with 4.7% microwaved less than 1 minute (Wachsmuth, 2000). In a preliminary experiment conducted by FDA/CFSAN, the heating of frankfurters by microwave ovens was measured with low (600 W) and high (1,100 W) powered microwave ovens (Buchanan, 2000). Four types were tested, including chicken frankfurters, low salt frankfurters, and two different size diameter frankfurters. Using various combinations of the two microwave power settings and four types of frankfurters, it was shown that the surface temperature increased faster than the center temperature. Heating for 25 seconds in the high power oven (1,100 W) and 40 seconds in the lower power oven (600 W) raised the surface temperature to at least 59 °C and, in some cases, raised the surface temperatures to over 70 °C. There is no information on what combinations of heating times and

temperatures are actually realized by consumers, but this preliminary experiment suggests that microwave heating is likely to be sufficient to cause substantial inactivation of any *Listeria monocytogenes* that might be present.

Inadequate data were found with which to directly model thermal inactivation in the frankfurters that receive some heating by microwaving, boiling, frying, grilling, broiling or other means. Therefore, data from inactivation of *E. coli* O157:H7 in hamburgers were adapted (Juneja *et al.,* 1997). These authors determined that survival of *E. coli* O157:H7 after cooking to maximum temperatures ranging from 54 to 77ºC (129 to 171 ºF) may be estimated by this equation:

$$\log_{10} \text{survivors} = 20.53 - 0.12\ T \qquad \text{Equation [6]}$$

The maximum cooking temperature to calculate the decrease (T) is in degrees Fahrenheit. Because the initial contamination was 6.6 logs, the equation can be rearranged to calculate the decrease in contamination and applied to any initial level of contamination. The temperature was also converted into degrees Celsius:

$$\log_{10} \text{reduction} = 0.216\ (T - 46.4) \qquad \text{Equation [7]}$$

A standard deviation of 0.5 logs was used to represent the uncertainty in the estimated reduction. This value reflects the sampling error from a similar experiment with *E. coli* O157:H7 (Jackson *et al*., 1996) where a 4.1 $\log_{10}$ reduction was observed after cooking to 68.3ºC.

Reductions in *Listeria monocytogenes* levels were calculated by estimating a distribution of cooking temperatures with a triangular distribution having a minimum of 54 ºC, most frequent temperature in the range of 69 to 73 ºC, and a maximum of 77 ºC. The four-degree range for the most frequent temperature represents uncertainty in the cooking temperature distribution. Table III-15 contains the resulting cumulative distribution in log reductions for the frankfurters that were given some reheating. The remainder had no reduction in *Listeria monocytogenes* after the growth modeling.

Table III-15. Cumulative Distribution of the Reduction (log_{10}) of *Listeria monocytogenes* in Reheated Frankfurters

Percentile	Median Reduction, log_{10} cfu/g [a]
1st	0.00 (0.00, 0.00)
5th	2.09 (1.90, 2.29)
10th	2.63 (2.52, 2.77)
25th	3.50 (3.38, 3.62)
50th	4.49 (4.32, 4.63)
75th	5.30 (5.13, 5.45)
90th	5.89 (5.78, 6.01)
95th	6.18 (6.05, 6.29)
99th	6.68 (6.57, 6.77)

[a] Values in parentheses are the 5th and 95th uncertainty levels.

Results: Modeled *Listeria monocytogenes* Levels in Food at Consumption

The estimated levels of *Listeria monocytogenes* at consumption are presented on Table III-16. This table has the same format as the table for *Listeria monocytogenes* contamination at retail (Table III-5), and may be directly compared to it to observe the shift in levels of *Listeria monocytogenes* after storage and/or heating. The median percentage of servings that fall within designated ranges of *Listeria monocytogenes* levels per serving are presented. The actual simulation modeling was at narrower levels (every logarithm and half-logarithm cfu/serving). The 5 and 95% values for the distributions for *Listeria monocytogenes* levels in each food are also given. These distributions indicate the uncertainty in the value for each median. The distribution observed with the values across a row gives the variation in *Listeria monocytogenes* levels expected for each food category. Because these medians are from skewed uncertainty distributions and because of rounding errors, a row may not sum to exactly 1.00.

As shown previously with the retail contamination estimates, every food category has some fraction of servings with at least 1 cfu/serving. The food categories range from 0.1% in hard cheeses to 8.7% in raw seafood. The column in Table III-16 showing 10^6 to 10^9 *Listeria monocytogenes* per serving is the level where the occurrence of listeriosis would be expected to be most likely. Smoked Seafood, Cooked RTE Crustaceans, Frankfurters not reheated, Deli Meats, and Pâté and Meat Spreads categories comprise a group of foods estimated to have the greatest likelihood of containing 10^6 to 10^9 *Listeria monocytogenes* per serving. These levels are

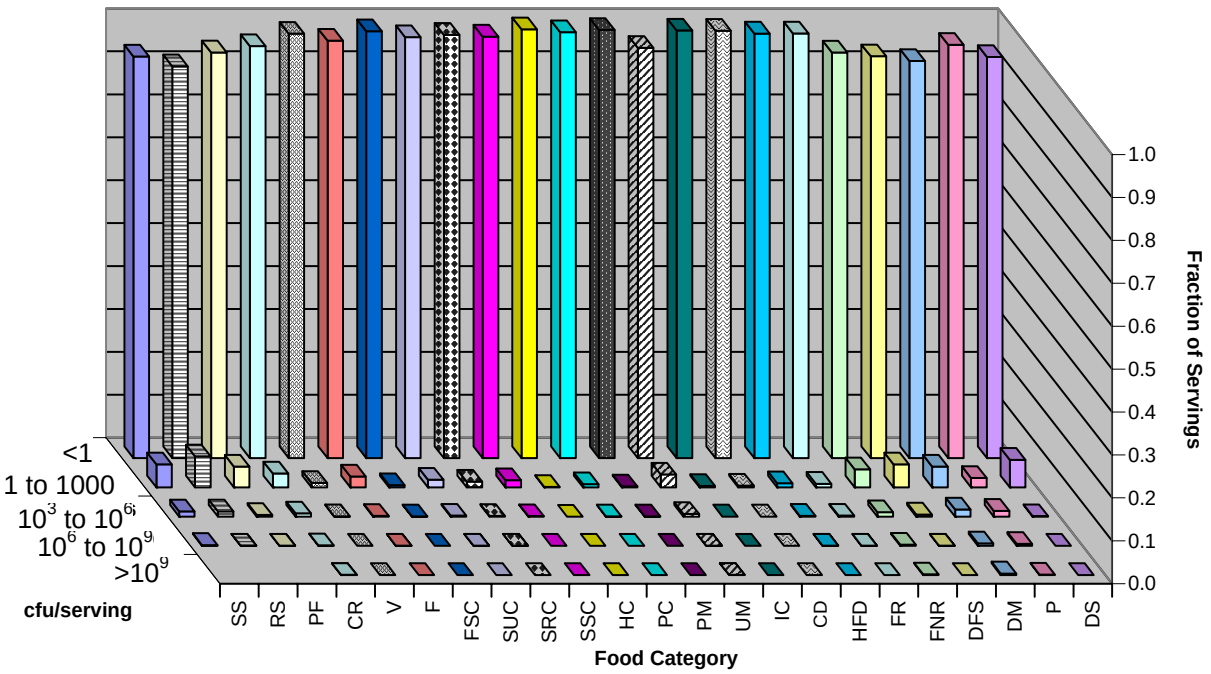
illustrated in Figure III-8. The row in the rear represents the fraction of servings with <1.0 cfu *Listeria monocytogenes*. All of the food categories have more than 90% of their servings in this contamination range. The rows have increasing levels of contamination toward the front of the figure.

Comparing corresponding values in Tables III-14 and III-16 shows the predicted effect of storage on the levels of *Listeria monocytogenes* at consumption. Cooked RTE Crustaceans, Frankfurters (not reheated), Deli Meats, and Pâté and Meat Spreads have some of the most dramatic changes. For example, at retail, 1.0% of Deli Meat servings would be in the 10^3 to 10^6 cfu/serving group. This increases to 1.6% at the time of consumption. In addition, the reduction in *Listeria monocytogenes* from reheating frankfurters is evident by comparing the <1, 1-1000 and 10^3 to 10^6 cfu/serving groups in Table III-16. The levels of *Listeria monocytogenes* in foods that do not permit growth, such as ice cream, do not show a change in comparing the values in Table III-14 (at retail levels) and Table III-16 (at consumption levels).

Table III-16. Modeled Percentage Distribution of Food Servings Contaminated with *Listeria monocytogenes* at Time of Consumption

Food Category	Median Percentage of Servings Contaminated at Different Levels[a]									
	<1 cfu/serving		1 - 1000 cfu/serving		10^3 - 10^6 cfu/serving		10^6 - 10^9 cfu/serving		> 10^9 cfu/serving	
	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]	Median	Percentiles[a]
Seafood										
Smoked Seafood	93.6	(51.6, 98.7)	5.3	(0.8, 24.6)	1.2	(0.2, 15.0)	0.2	(<0.1, 8.2)	<0.1	(<0.1, 0.5)
Raw Seafood	91.3	(87.3, 98.6)	7.2	(1.2, 10.8)	1.2	(0.1, 2.2)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)
Preserved Fish	94.5	(70.8, 99.8)	4.8	(0.2, 20.4)	0.4	(<0.1, 4.1)	<0.1	(<0.1, 0.8)	<0.1	(<0.1, <0.1)
Cooked Ready-to-Eat Crustaceans	96.0	(93.9, 97.0)	3.2	(2.5, 5.5)	0.7	(0.4, 1.0)	0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)
Produce										
Vegetables	98.9	(98.7, 99.0)	1.0	(0.9, 1.3)	0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Fruits	97.3	(70.4, 99.8)	2.5	(0.2, 21.4)	0.2	(<0.1, 7.5)	<0.1	(<0.1, 1.4)	<0.1	(<0.1, <0.1)
Dairy										
Fresh Soft Cheese	99.5	(95.2, 99.7)	0.5	(0.3, 4.5)	<0.1	(<0.1, 0.7)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
Soft Unripened Cheese	98.1	(90.1, 99.9)	1.8	(0.1, 7.5)	0.2	(<0.1, 3.7)	<0.1	(<0.1, 1.0)	<0.1	(<0.1, <0.1)
Soft Ripened Cheese	98.6	(84.0, 99.9)	1.3	(0.1, 12.8)	0.1	(<0.1, 3.0)	<0.1	(<0.1, 0.4)	<0.1	(<0.1, <0.1)
Semi-soft Cheese	98.2	(91.4, 98.8)	1.7	(1.1, 6.9)	0.1	(<0.1, 1.3)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)
Hard Cheese	99.9	(98.3, 100.0)	0.1	(<0.1, 1.6)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Processed Cheese	99.2	(97.8, 99.9)	0.7	(0.1, 2.1)	<0.1	(<0.1, 0.2)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Pasteurized Fluid Milk	99.7	(97.8, 99.9)	0.2	(0.1, 1.8)	<0.1	(<0.1, 0.4)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Unpasteurized Fluid Milk	95.6	(90.0, 99.6)	3.0	(0.4, 7.6)	0.6	(<0.1, 5.1)	<0.1	(<0.1, 1.3)	<0.1	(<0.1, 0.2)
Ice Cream/Frozen Dairy Products	99.6	(99.3, 99.8)	0.4	(0.2, 0.6)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
Cultured Milk Products	99.6	(95.8, 99.9)	0.4	(0.1, 3.8)	<0.1	(<0.1, 0.3)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)
High Fat and Other Dairy Products	98.9	(98.3, 99.2)	0.9	(0.6, 1.5)	0.2	(0.1, 0.4)	<0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
Meats										
Frankfurters (reheated)	98.9	(97.3, 99.1)	0.8	(0.7, 2.1)	0.2	(0.2, 0.5)	0.1	(<0.1, 0.1)	<0.1	(<0.1, <0.1)
Frankfurters (not reheated)	94.5	(88.5, 95.5)	4.2	(3.1, 8.1)	1.0	(1.0, 2.5)	0.3	(0.2, 0.8)	0.1	(0.1, 0.3)
Dry/Semi-dry Fermented Sausages	93.6	(77.7, 97.6)	5.4	(2.1, 19.7)	0.5	(<0.1, 4.1)	<0.1	(<0.1, 1.1)	<0.1	(<0.1, <0.1)
Deli Meats	92.5	(87.8, 99.3)	4.8	(0.5, 8.6)	1.6	(0.1, 2.4)	0.5	(<0.1, 0.7)	0.3	(<0.1, 0.6)
Pâté and Meat Spreads	96.3	(79.8, 98.0)	2.2	(1.2, 8.6)	1.3	(0.6, 7.8)	0.4	(0.2, 3.8)	0.1	(<0.1, 0.6)
Combination Foods										
Deli-type Salads	93.5	(88.7, 98.2)	6.4	(1.8, 11.1)	0.1	(<0.1, 0.3)	<0.1	(<0.1, <0.1)	<0.1	(<0.1, <0.1)

[a] The 5th and 95th percentiles uncertainty levels, respectively.



LEGEND			
SS =	Smoked Seafood	PM =	Pasteurized Fluid Milk
RS =	Raw Seafood	UM =	Unpasteurized Fluid Milk
PF =	Preserved Fish	IC =	Ice Cream and Frozen Dairy Products
CR =	Cooked Ready-To-Eat Crustaceans	CD=	Cultured Milk Products
V =	Vegetables	HFD	High Fat and Other Dairy Products
F =	Fruits	FR =	Frankfurters (reheated)
FSC =	Fresh Soft Cheese	FNR=	Frankfurters (not reheated)
SUC =	Soft Unripened Cheese	DFS =	Dry/Semi-Dry Fermented Sausages
SRC =	Soft Ripened Cheese	DM =	Deli Meats
SSC =	Semi-soft Cheese	P =	Pâté and Meat Spreads
HC =	Hard Cheese	DS =	Deli-type Salads
PC =	Processed Cheese		

Figure III-8. Three Dimensional Graph of the Modeled Distribution of *Listeria monocytogenes* Levels of Contamination at the Time of Consumption for the Food Categories

An approximation of the overall frequency of consumption of *Listeria monocytogenes* by the United States population can be made by multiplying the fraction of servings in each food category-dose bin (Table III-16) by the annual number of servings in each food category (Table III-2). The numbers of servings are then summed for each dose for all of the food categories. Table III-17 shows that most of the servings have less than one *Listeria monocytogenes* and the number of contaminated servings decreases with increasing levels of contamination. If the number of contaminated servings is divided by the United States population (2.6×10^8), an approximation of how frequently the "average person" would encounter these levels of *Listeria monocytogenes* each year can be calculated. This "average" person would consume a serving with 10^3 to 10^6 microorganisms 2.4 times per year, 10^6 to 10^9 microorganisms once every two years and more than 10^9 microorganisms once every three years.

Table III-17. Number of Servings of Food per Year Containing Various Levels of *Listeria monocytogenes*

***Listeria monocytogenes* Levels in Food (per serving)**	**Number of Servings (per year in the United States)**	**Number of Servings (per person per year)**
0	3.3×10^{11}	1300
1 to 1000	4.9×10^9	19
1×10^3 to 1×10^6	6.2×10^8	2.4
1×10^6 to 1×10^9	1.3×10^8	0.5
$> 1 \times 10^9$	7.3×10^7	0.3