MICROBIOLOGICAL RISK ASSESSMENT SERIES

5

Risk assessment of *Listeria monocytogenes* in ready-to-eat foods

TECHNICAL REPORT

For further information on the joint FAO/WHO activities on microbiological risk assessment, please contact:

Food Quality and Standards Service
Food and Nutrition Division
Food and Agriculture Organization of the United Nations
Viale delle Terme di Caracalla
I-00100 Rome, Italy

Fax: +39 06 57054593
E.mail: jemra@fao.org

Web site: http://www.fao.org/es/esn

or

Food Safety Department
World Health Organization
20, Avenue Appia
CH-1211 Geneva 27
Switzerland

Fax: +41 22 7914807
E.mail: foodsafety@who.int

Web site: http://www.who.int/foodsafety

Cover design : Food and Agriculture Organization of the United Nations
and the World Health Organization.

Cover picture:

The designations employed and the presentation of the material in this publication do not The designations employed and the presentation of the material in this publication do not imply the expression of any opinion whatsoever on the part of the World Health Organization nor of the Food and Agriculture Organization of the United Nations concerning the legal or development status of any country, territory, city or area or of its authorities, or concerning the delimitation of its frontiers or boundaries.

The views expressed herein are those of the authors and do not necessarily represent those of the World Health Organization nor of the Food and Agriculture Organization of the United Nations nor of their affiliated organization(s).

The World Health Organization and the Food and Agriculture Organization of the United Nations do not warrant that the information contained in this publication is complete and shall not be liable for any damage incurred as a result of its use.

WHO Library Cataloguing-in-Publication Data

Risk assessment of *Listeria monocytogenes* in ready-to-eat foods : technical report.

(Microbiological risk assessment series ; no. 5)

1. Listeria monocytogenes - pathogenicity 2.Listeria monocytogenes – growth and development 4.Food contamination 5.Food handling 6.Risk assessment - methods 7.Listeria infections I.World Health Organization II.Food and Agriculture Organization of the United Nations III.Series

ISBN 92 4 156262 5 (WHO) (LC/NLM classification: QW 142.5.A8)
ISBN 92 5 105 127 5 (FAO)
ISSN 1726-5274

CONTENTS

PART 3. EXPOSURE ASSESSMENT

PART 4. EXAMPLE RISK ASSESSMENTS

Acknowledgements

The Food and Agriculture Organization of the United Nations and the World Health Organization would like to express their appreciation to all those who contributed to the preparation of this report. Special appreciation is extended to the risk assessment drafting group for the time and effort that they freely dedicated to the elaboration of this risk assessment. Many people provided their time and expertise by reviewing the report and providing their comments, additional data and other relevant information. Both the risk assessment drafting group and the reviewers are listed in the following pages.

Appreciation is also extended to all those who responded to the calls for data that were issued by FAO and WHO and brought to our attention data which is not readily available in the mainstream literature and official documentation.

The risk assessment work was co-ordinated by the Joint FAO/WHO Secretariat on Risk Assessment of Microbiological Hazards in Foods. This included Sarah Cahill, Maria de Lourdes Costarrica and Jean Louis Jouve in FAO and Peter Karim Ben Embarek, Allan Hogue, Jocelyne Rocourt, Hajime Toyofuku and Jørgen Schlundt in WHO. During the development of the risk assessment additional support and feedback were provided by Lahsen Ababouch and Hector Lupin, Fishery Industries Division in FAO and Jeronimas Maskeliunas, Codex Secretariat. Publication of the report was coordinated by Sarah Cahill. Thorgeir Lawrence was responsible for editing the report for language and preparation for printing.

The preparatory work and the publication was supported and funded by the FAO Food Quality and Standards Service, the FAO Fishery Industries Division and the WHO Food Safety Department.

RISK ASSESSMENT DRAFTING GROUP

(in alphabetical order)

Robert Buchanan
Center for Food Safety and Nutrition, Food and Drug Administration,
United States of America

Roland Lindqvist
National Food Administration,
Sweden

Thomas Ross
University of Tasmania,
Australia

Mark Smith
Bureau of Biostatistics and Computer Applications, Health Products and Food Branch,
Health Canada, Canada

Ewen Todd
The National Food Safety & Toxicology Center, Michigan State University,
United States of America

Richard Whiting
Center for Food Safety and Nutrition, Food and Drug Administration,
United States of America

Assistance was provided to the drafting group by the following:

Elizabeth A. Junkins, **Michel Vigneault** and **William Ross**, Bureau of Biostatistics and Computer Applications, Health Products and Food Branch, Health Canada.

Jeff M. Farber, Bureau of Microbial Hazards, Health Products and Food Branch, Health Canada.

Sven Rasmussen, University of Tasmania.

Suzanne van Gerwen, Microbiology and Preservation Unit, Unilever Research, Vlaardingen, The Netherlands.

FDA/FSIS *Listeria Monocytogenes* Risk Assessment Team, Washington, DC, USA
(M. Bender, C. Carrington, S. Dennis, E. Ebel, A. Hitchens, N. Hynes, W. Long,
W. Schlosser, R. Raybourne, M. Ross, T. Rouse and R. Whiting)

Meat and Livestock Australia, for access to the report of Ross and Shadbolt (2001), and **Australian and New Zealand Food Authority,** for access to product composition data.

REVIEWERS

The risk assessment was reviewed on several occasions both during and after its elaboration, including through expert consultations and selected peer-reviews, and by members of the public in response to a call for public comments.

PARTICIPANTS IN EXPERT CONSULTATIONS

Amma Anandavally	Export Quality Control Laboratory, India.
Wayne Anderson	Food Safety Authority of Ireland, Ireland.
Olivier Cerf	Ecole Nationale Vétérinaire d'Alfort (ENVA), France.
Jean-Yves D'Aoust	Health Protection Branch, Health Canada, Canada.
Paw Dalgaard	Danish Institute for Fisheries Research, Ministry of Food Agriculture and Fisheries, Denmark.
Michael Doyle	Center for Food Safety, University of Georgia, United States of America.
Eric Ebel	United States Department of Agriculture, Food Safety and Inspection Service, United States of America.
Emilio Esteban	Centres for Disease Control and Prevention, United States of America.
Aamir Fazil	Population and Public Health Branch, Health Canada, Canada.
Lone Gram	Danish Institute of Fisheries Research, Technical University of Denmark, Denmark.
Steve Hathaway	MAF Regulatory Authority (Meat and Seafood), New Zealand
Matthias Hartung,	National Reference Laboratory on the Epidemiology of Zoonoses, Germany.
Inocencio Higuera Ciapara	Research Centre for Food and Development (CIAD), Mexico.
Andrew Hudson	The Institute of Environmental Science and Research Ltd, New Zealand.
David Jordan	New South Wales Agriculture, Wollongbar Agricultural Institute, Australia.
Fumiko Kasuga	National Institute of Infectious Diseases, Department of

	Biomedical Food Research, Japan.
Louise Kelly	Department of Risk Research, Veterinary Laboratories Agency, United Kingdom.
Julia A. Kiehlbauch	Microbiology Consultant, United States of America.
Günter Klein,	Division of Food Hygiene, Federal Institute for Health Protection of Consumers and Veterinary Medicine, Germany.
Susumu Kumagai	Graduate School of Agriculture and Life Sciences, The University of Tokyo, Japan.
Anna Lammerding	Population and Public Health Branch, Health Canada, Canada
Xiumei Liu	Institute of Nutrition and Food Hygiene, Ministry of Health, China.
Carol Maczka	United States Department of Agriculture, Food Safety and Inspection Service, United States of America.
Patience Mensah	Noguchi Memorial Institute for Medical Research, University of Ghana, Ghana.
George Nasinyama	Department of Epidemiology and Food Safety, Makerere University, Uganda.
Gregory Paoli	Decisionalysis Risk Consultants Inc. , Canada.
Irma N.G. Rivera	Departamento de Microbiologia, Universidade de São Paulo, Brazil.
Son Radu	Department of Biotechnology, University Putra Malaysia, Malaysia.
Dulce Maria Tocchetto Schuch	Laboratorio Regional de Apoio Animal – Lara / RS Agriculture Ministry, Brazil.
Eystein Skjerve	Department of Pharmacology, The Norwegian School of Veterinary Science, Norway.
Bruce Tompkin	ConAgra Inc., United States of America.
Suzanne Van Gerwen	Microbiology and Preservation Unit, Unilever Research, The Netherlands.
Michiel Van Schothorst	Wageningen University, The Netherlands.
Kaye Wachsmuth	United States Department of Agriculture, Food Safety and Inspection Service, United States of America.
Helene Wahlström	National Veterinary Institute,

	Sweden.
Charles Yoe	Department of Economics, College of Notre Dame of Maryland United States of America.

REVIEWERS IN RESPONSE TO CALL FOR PUBLIC COMMENT

K.E. Aidoo	School of Biomedical and Biological Sciences,Glasgow Caledonian University, Scotland.
Scott Cameron	Department of Public Health, University of Adelaide, Australia.
Andrew Hudson	The Institute of Environmental Science and Research Ltd, New Zealand.
Industry Council for Development	United Kingdom
O.O. Komolafe	Department of Microbiology, College of Medicine, Malawi.
Douglas Marshall	Department of Food Science & Technology, Mississippi State University, United States of America.
Bruce Tompkin	ConAgra Inc., United States of America.
United States Food and Drug Administration	United States of America.

PEER-REVIEWERS

Paolo Aureli	Food Department, Istituto Superiore della Sanità, Italy.
Sava Buncic	School of Veterinary Science, University of Bristol, United Kingdom.
Carmen Buchrieser	Laboratoire de Genetique des Microorganismes Pathogenes, Institut Pasteur, France.
Marie Teresa Destros	Depto Alimentos e Nutrição Experimental, FCF – USP, Brazil.
Veronique Goulet	Institut de Veille Sanitaire, Hopital de Saint-Maurice, France.
Lone Gram	Danish Institute of Fisheries Research, Technical University of Denmark, Denmark.

Arie Havelaar	Microbiological Laboratory for Health Protection, National Institute for Public Health and the Environment, The Netherlands.
Sophia Kathariou	Department of Food Science, North Carolina State University, United States of America.
Heejeong Latimar	United States Department of Agriculture, Food Safety and Inspection Service, United States of America.
Paul Martin	Laboratoire des Listeria, Institut Pasteur, France.
Robert L. McMasters	Department of Mechanical Engineering, Michigan State University, United States of America.
Thomas McMeekin	Centre for Food Safety and Quality, University of Tasmania, Australia.
Birgit Norrung	Danish Veterinary and Food Administration, Denmark.
Servè Notermans	TNO Nutrition and Food Research Institute, The Netherlands.
Jocelyne Rocourt	Food Safety Programme, World Health Organization, Switzerland.
Elliot T. Ryser	Department of Food Science and Human Nutrition, Michigan State University, United States of America.
Don Schaffner	Food Risk Analysis Initiative, Rutgers University, United States of America.
William Henry Sperber	Cargill, United States of America.
David Vose	David Vose Consulting, France.
Marion Wooldridge	Department of Risk Research, Veterinary Laboratory Agency (Weybridge), United Kingdom.

FOREWORD

The Members of the Food and Agriculture Organization of the United Nations (FAO) and of the World Health Organization (WHO) have expressed concern regarding the level of safety of food both at national and international levels. Increasing foodborne disease incidence over the last decades seems, in many countries, to be related to an increase in disease caused by microorganisms in food. This concern has been voiced in meetings of the Governing Bodies of both Organizations and in the Codex Alimentarius Commission. It is not easy to decide whether the suggested increase is real or an artefact of changes in other areas, such as improved disease surveillance or better detection methods for microorganisms in foods. However, the important issue is whether new tools or revised and improved actions can contribute to our ability to lower the disease burden and provide safer food. Fortunately new tools, which can facilitate actions, seem to be on their way.

Over the past decade, Risk Analysis – a process consisting of risk assessment, risk management and risk communication – has emerged as a structured model for improving our food control systems with the objectives of producing safer food, reducing the numbers of foodborne illnesses and facilitating domestic and international trade in food. Furthermore, we are moving towards a more holistic approach to food safety, where the entire food chain needs to be considered in efforts to produce safer food.

As with any model, tools are needed for the implementation of the risk analysis paradigm. Risk assessment is the science-based component of risk analysis. Science today provides us with in-depth information on life in the world we live in. It has allowed us to accumulate a wealth of knowledge on microscopic organisms, their growth, survival and death, even their genetic make-up. It has given us an understanding of food production, processing and preservation, and of the link between the microscopic and the macroscopic world and how we can benefit from as well as suffer from these microorganisms. Risk assessment provides us with a framework for organizing all this data and information and to better understand the interaction between microorganisms, foods and human illness. It provides us with the ability to estimate the risk to human health from specific microorganisms in foods and gives us a tool with which we can compare and evaluate different scenarios, as well as to identify the types of data is necessary for estimating and optimizing mitigating interventions.

Microbiological risk assessment can be considered as a tool that can be used in the management of the risks posed by foodborne pathogens and in the elaboration of standards for food in international trade. However, undertaking a microbiological risk assessment (MRA), particularly quantitative MRA, is recognized as a resource-intensive task requiring a multidisciplinary approach. Yet foodborne illness is among the most widespread public health problems, creating social and economic burdens as well as human suffering, making it a concern that all countries need to address. As risk assessment can also be used to justify the introduction of more stringent standards for imported foods, a knowledge of MRA is important for trade purposes, and there is a need to provide countries with the tools for understanding and, if possible, undertaking MRA. This need, combined with that of the Codex Alimentarius for risk-based scientific advice, led FAO and WHO to undertake a programme of activities on MRA at the international level.

The Food Quality and Standards Service, FAO, and the Food Safety Department, WHO, are the lead units responsible for this initiative. The two groups have worked together to develop the area of MRA at the international level for application at both the national and international levels. This work has been greatly facilitated by the contribution of people from around the world with expertise in microbiology, mathematical modelling, epidemiology and food technology to name but a few.

This Microbiological Risk Assessment series provides a range of data and information to those who need to understand or undertake MRA. It comprises risk assessments of particular pathogen-commodity combinations, interpretative summaries of the risk assessments, guidelines for undertaking and using risk assessment, and reports addressing other pertinent aspects of MRA.

We hope that this series will provide a greater insight into MRA, how it is undertaken and how it can be used. We strongly believe that this is an area that should be developed in the international sphere, and have already from the present work clear indications that an international approach and early agreement in this area will strengthen the future potential for use of this tool in all parts of the world, as well as in international standard setting. We would welcome comments and feedback on any of the documents within this series so that we can endeavour to provide Member countries, Codex Alimentarius and other users of this material with the information they need to use risk-based tools, with the ultimate objective of ensuring that safe food is available for all consumers.

Ezzeddine Boutrif
Food Quality and Standards Service
FAO

Jørgen Schlundt
Food Safety Department
WHO

ABBREVIATIONS USED IN THE TEXT

AIDS	Acquired Immunodeficiency Syndrome
a_w	Water activity
BHI	Brain-heart infusion
CCFH	Codex Committee on Food Hygiene
CDC	Centers for Disease Control and Prevention (USA)
CFU	Colony-forming units
CNS	Central nervous system
CSFII	Continuing Survey of Food Intakes by Individuals (USA)
EGR	Exponential growth rate
FAO	Food and Agriculture Organization of the United Nations
FDA	Food and Drug Administration (USA)
FSIS	Food Safety and Inspection Service [USDA]
ID_{50}	Dose of an infectious organism required to produce infection in 50 percent of the experimental subjects or exposed population.
HIV	Human Immunodeficiency Virus
IV	Intravenous
LD_{50}	The amount of an infectious organism or toxic agent that is sufficient to kill 50 percent of the exposed population within a certain time.
LLO	Listeriolysin O
LMRA	*Listeria monocytogenes* Risk Assessment [FDA/FSIS]
MPD	Maximum population density
MPN	Most probable number
MRA	Microbiological risk assessment
MSE	Mean square error
NaCl	Sodium chloride
NHANES	National Health and Nutrition Examination Survey (USA)
RLT	Relative lag time
RTE	Ready-to-eat
USDA	United States Department of Agriculture
WHO	World Health Organization
WPS	Water phase salt

Executive Summary

This risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods was undertaken to (i) respond to the request of the Codex Committee on Food Hygiene (CCFH) for sound scientific advice as a basis for the development of guidelines for the control of *L. monocytogenes* in foods; and (ii) address the needs expressed by Member countries for adaptable risk assessments that they can use to support risk management decisions and to conduct their own assessments.

The risk assessment was tailored to address three specific questions posed by the 33rd session of the CCFH (CAC, 2000) namely:

1. Estimate the risk of serious illness from *L. monocytogenes* in food when the number of organisms ranges from absence in 25 grams to 1000 colony forming units (CFU) per gram or millilitre, or does not exceed specified levels at the point of consumption.
2. Estimate the risk of serious illness for consumers in different susceptible population groups (elderly, infants, pregnant women and immunocompromised patients) relative to the general population.
3. Estimate the risk of serious illness from *L. monocytogenes* in foods that support its growth and foods that do not support its growth at specific storage and shelf-life conditions.

By answering these questions, this risk assessment aims to assist risk managers in conceptualizing how some of the factors governing foodborne listeriosis interact, thereby assisting the development of strategies to reduce the rates of illness.

The risk assessment comprises the four steps of hazard identification, hazard characterization, exposure assessment and risk characterization. A quantitative approach was taken and mathematical modelling employed to estimate the risks per serving and risk to a population in a year from the selected foods. The risk assessment focused on four RTE foods in order to provide examples of how microbiological risk assessment techniques can be used to answer food safety questions at an international level. The study was limited to foods at retail and their subsequent public health impact at the time of consumption. The impact of post-retail factors that could influence the risk to a consumer, such as temperature and duration of refrigerated storage, was also examined. This was considered sufficient to address the questions posed by the CCFH within the time frame and resources available to the risk assessors, and also reflects the situation that most of the currently available exposure data for *L. monocytogenes* relate to the frequency and extent of contamination at the retail level.

HAZARD IDENTIFICATION

Foodborne listeriosis is a relatively rare but serious disease with high fatality rates (20–30%) compared with other foodborne microbial pathogens, such as *Salmonella*. The disease largely affects specific segments of the population who have increased susceptibilities. Basically, *L. monocytogenes* is an opportunistic pathogen that most often affects those with a severe underlying disease or condition (e.g. immunosuppression, HIV/AIDS, chronic conditions such as cirrhosis that impair the immune system); pregnant women; unborn or newly delivered infants; and the elderly. *L. monocytogenes* is widely dispersed in the environment and foods. However, it was not until several large, common-source outbreaks of listeriosis occurred in North America and Europe during the 1980s that the significance of foods as the primary route of transmission for human exposure to *L. monocytogenes* was recognized (Broome, Gellin and Schwartz, 1990; Bille, 1990). An important factor in foodborne listeriosis is that the pathogen can grow to significant numbers at refrigeration temperatures when given sufficient time. Despite the fact that a wide variety of foods may be contaminated with *L. monocytogenes*, outbreaks and sporadic cases of listeriosis are predominately associated with RTE foods – a large, heterogeneous category of foodstuffs that can be subdivided in many different ways and vary from country to country according to local eating habits; availability and integrity of the chill chain; and regulations specifying, for example, the maximum temperature at retail level. Although listeriosis is a relatively rare disease, the severity of the disease and the very frequent involvement of industrially processed foods, especially during outbreaks, mean that the social and economic impact of listeriosis is among the highest of the foodborne diseases (Roberts, 1989; Roberts and Pinner, 1990). Listeriosis is mainly observed in industrialized countries and it is not known whether the differences in incidence rates between developed and developing countries reflect true geographical differences, differences in food habits and food storage, or differences in diagnosis and reporting practices.

HAZARD CHARACTERIZATION

The hazard characterization provides a description of the pathogen and host characteristics that contribute to an infection by *Listeria,* the public health outcomes of infection with this pathogen, the foods most commonly associated with listeriosis, and a description of the dose-response relationship. Various clinical manifestations are associated with listeriosis and these can be grouped in two categories: invasive listeriosis and non-invasive listeriosis. Invasive listeriosis are cases when initial infections of the intestinal tissue by *L. monocytogenes* leads to invasion of otherwise sterile body sites, such as the pregnant uterus, the central nervous system, or the blood, or combinations. Invasive listeriosis is characterized by a high case-fatality rate, ranging from 20 to 30% (Mead et al., 1999) and sequelae may follow listeriosis infections (McLauchlin, 1997), though their incidence is rarely estimated (Rocourt, 1996). Non-invasive listeriosis (referred to as febrile listerial gastroenteritis) has been observed during a number of outbreaks where the majority of cases developed symptoms of gastroenteritis, such as diarrhoea, fever, headache and myalgia, after a short period of incubation (Dalton et al*.,* 1997; Salamina et al*.,* 1996; Riedo et al*.,* 1994; Aureli et al., 2000). These outbreaks have generally involved the ingestion of high doses of *L. monocytogenes* by otherwise healthy individuals. The incidence rate and factors that govern the onset of this non-invasive form are not known. As a result, this risk assessment only considered invasive listeriosis as the outcome of exposure.

Dose-response data from human volunteer studies with *L. monocytogenes* or from volunteer studies with a surrogate pathogen do not exist. Therefore dose-response relations have been developed and evaluated based on expert elicitations, epidemiological or animal data, or combinations of these. These dose-response relations, which were reviewed and summarized in this work, cover the spectrum of biological end-points, i.e. infection, morbidity and mortality, and have, to varying degrees of sophistication, been evaluated using human epidemiological data. All models assume that each microbial cell acts independently, and that a single bacterial cell has the potential to cause disease. However, none of the available models were fully able to meet the needs of the current risk assessment in relation to the parameters examined and simplicity of calculation. For these reasons, alternative approaches were developed and evaluated for this risk assessment.

The approach used took advantage of the epidemiological data and detailed exposure assessment available in the *Listeria* risk assessment developed in the United States of America (FDA/FSIS, 2001). The model contains one parameter, r, which is the probability that a single cell will cause invasive listeriosis. This parameter was estimated from the pairing of population consumption patterns (exposure) with epidemiological data on the number of invasive listeriosis cases in the population. The estimated r-value, which will vary with the data sets used and the assumptions made, was then used in the exponential model to estimate specific risks given the number of *L. monocytogenes* consumed.

EXPOSURE ASSESSMENT

A full farm-to-fork risk assessment was not required to address the questions posed by the CCFH. Thus, the focus of the exposure assessment models was to account for changes in the frequency and extent of contamination in the food between retail marketing and the point of consumption. This simplified the modelling and reduced the model uncertainties, thereby decreasing the ranges around the final risk estimates. The models developed describe the growth or decline of *L. monocytogenes* between the time of purchase and consumption, using information and models for the growth rate and the lag time of *L. monocytogenes* as affected by storage temperature and food composition, the maximum growth of *L. monocytogenes* supported by the food, and the distribution of retail and home storage times and temperatures. Calculating the numbers of *L. monocytogenes* actually consumed also required consideration of how much of and how often the food is eaten (i.e. the size and the number of servings).

RTE foods are a broad and diverse food category, prepared and stored in different ways and under different conditions, some of which support growth of *L. monocytogenes* and others that do not support growth at specific storage and shelf-life conditions. As it was therefore not possible to consider all RTE foods, four foods – pasteurized milk, ice cream, fermented meat and cold smoked fish – were selected to illustrate how the different factors mentioned above interact to affect the risk of acquiring listeriosis. Pasteurized milk is a food that is widely consumed, has very low frequencies and levels of contamination with *L. monocytogenes* but allows growth of the organism during storage. Ice cream is similar to milk but does not permit growth of *L. monocytogenes* during storage. Fermented meat products are often contaminated with *Listeria* and are produced without any lethal processing step, but their final composition prevents growth of the microbe during storage. Cold-smoked fish is frequently contaminated with *L. monocytogenes*, has no lethal processing step and permits growth during an extended storage period.

Several "what-if" scenarios were also considered in the case of milk and smoked salmon. These hypothetical scenarios have specific changes made to one or more of the exposure factors to demonstrate how the factors interact to affect the risk. In conducting the exposure assessments for these four foods, different databases were available and the modellers used slightly different techniques. These techniques are explained in the main risk assessment document and illustrate that there are numerous approaches that may be taken depending on the available data and the judgment of the risk assessors.

The outputs from the exposure assessment included a distribution of *L. monocytogenes* in the food at the point of consumption (frequency of contamination) and also the amount consumed (number of servings per year and size of servings).

RISK CHARACTERIZATION

The outputs from the exposure assessment were fed into the dose-response model to develop the risk characterization portion of the risk assessment to calculate the probability of contracting listeriosis. The outputs are described in terms of estimates of risk per million servings for the healthy and susceptible populations. The risk per serving and number of servings were used to estimate the number of illnesses in a specified population per year.

The mean risk estimates of the number of illnesses per 10 million people per year and the risk per serving for pasteurized milk, ice cream, fermented meats and smoked fish are shown in Table 1. For milk, for example, the risk per serving was low (5.0×10^{-9} cases per serving), but the very high frequency of consumption resulted in milk making substantial contributions to the total number of predicted cases of illness. In contrast, for smoked fish the risk per serving was estimated to be high (2.1×10^{-8} cases per serving). However, consumption of this product is modest (1 to 18 servings per year), and consequently the total number of cases of listeriosis was moderate.

Table 1

The mean risk estimates of the number of illnesses per 10 million people per year and the risk per serving for four ready-to-eat foods.

Food	Cases of listeriosis per 10 million people per year	Cases of listeriosis per 1 million servings
Milk	9.1	0.005
Ice cream	0.012	0.000014
Smoked fish	0.46	0.021
Fermented meats	0.00066	0.0000025

RESPONSE TO QUESTIONS POSED BY THE CCFH

These risk assessments were used to address the specific questions posed by the 33rd session of the CCFH. The replies to these questions are summarized below.

Question 1: *Estimate the risk of serious illness from* L. monocytogenes *in food when the number of organisms range from absence in 25 g to 1000 colony forming units (CFU) per gram or millilitre, or does not exceed specified levels at the point of consumption.*

Two approaches were taken: (i) the predicted risk per serving and predicted number of cases of listeriosis annually were estimated for a "worst-case" scenario by assuming that all servings had the maximum level being considered (0.04, 0.1, 1, 10, 100 and 1000 CFU/g); (ii) a more realistic, but also more complex, approach was to use a distribution of the levels of *L. monocytogenes* in foods when consumed rather than an absolute value to estimate the risk per serving and the predicted number of cases of listeriosis annually.

Comparisons between these two approaches indicated that there were vast differences in the estimated number of cases when one considers the worst-case scenario as opposed to a scenario that attempts to also consider the frequency and extent of contamination actually encountered in RTE foods. These two scenarios demonstrated that as either the frequency of contamination or the level of contamination increases, the risk and the predicted number of cases also increase. These scenarios assume that ingestion of a single cell has the possibility to cause illness. Thus, if all RTE foods went from having 1 CFU/serving to 1000 CFU/serving, the risk of listeriosis would increase 1000-fold (assuming a fixed serving size). Conversely, the effect of introducing into the food supply 10 000 servings contaminated with *L. monocytogenes* at a level of 1000 CFU/g would, in theory, be compensated by removing from the food supply a single serving contaminated at a level of 10^7 CFU/g.

In interpreting these results and the actual effect of a change in the regulatory limits for *L. monocytogenes* in RTE foods, one also has to take into account the extent to which non-compliance with established limits occurs. Based on data available for the United States of America, where the current limit for *L. monocytogenes* in RTE foods is 0.04 CFU/g, the estimated number of cases for listeriosis for that population was 2130 (baseline level used in the United States *Listeria* risk assessment). If a level of 0.04 CFU/g was consistently achieved, one could expect less than 1 case of listeriosis per year. This, in combination with available exposure data, suggests that a portion of RTE food contains a substantially greater number of the pathogen than the current limit and that the public health impact of *L. monocytogenes* is almost exclusively a function of the foods that greatly exceed the current limit. Therefore it could be asked if a less stringent microbiological limit for RTE foods could be beneficial in terms of public health if it simultaneously fostered the adoption of control measures that resulted in a substantial decrease in the number of servings that greatly exceeded the established limit.

To examine this concept further, a simple "what-if" scenario was developed describing the impact on public health of the level of compliance to a microbiological limit. Two often discussed limits, 0.04 CFU/g and 100 CFU/g, were examined in conjunction with different "defect rates" (a defect rate is the percentage of servings that exceed the specified limit). To simplify the model, a single level of *L. monocytogenes* contamination, 10^6 CFU/g, was assumed for all "defective" servings. This assumption focuses the scenario on the group of defective servings that is responsible for the majority of listeriosis cases. Data demonstrate that at 100% compliance, the number of predicted cases is low for both limits, with an approximate 10-fold difference between them, that is 0.5 cases versus 5.7 cases. As expected the number of cases increases with an increasing frequency of defective servings. However, it is possible that public health could be improved if an increase in the regulatory limit in RTE foods resulted in a substantial decrease in the number of servings that greatly exceeded the established limit, i.e. if the rate of compliance increased.

To summarize, the risk assessment demonstrates that the vast majority of cases of listeriosis result from the consumption of high numbers of *Listeria*, and foods where the level of the pathogen does not meet the current criteria, whatever they may be (0.04 or 100 CFU/g). The model also predicts that the consumption of low numbers of *L. monocytogenes* has a low probability of causing illness. Eliminating higher levels of *L. monocytogenes* at the time of consumption has a large impact on the number of predicted cases of illness.

Question 2: Estimate the risk of serious illness for consumers in different susceptible population groups (elderly, infants, pregnant women and immunocompromised patients) relative to the general population.

These results showed that the probability of becoming ill from ingesting *L. monocytogenes* was higher for susceptible populations (immunocompromised; elderly; and perinatal) than the general population. The probability of becoming ill was also shown to vary between the sub-groups of the susceptible population. Based on susceptibility information available from the United States of America, it was determined that the elderly (60 years and older) were 2.6 times more susceptible relative to the general healthy population, while perinatals were 14 times more susceptible. Conditions that compromise the immune system also affect susceptibility to varying extents (Table 2). These results are consistent with the physiological observation that, as an individual's immune system is increasingly compromised, the risk of listeriosis at any given dose increases.

Table 2 Relative susceptibilities for different sub-populations based on French epidemiological data.

Condition	Relative susceptibility
Transplant	2584
Cancer-Blood	1364
AIDS	865
Dialysis	476
Cancer-Pulmonary	229
Cancer-Gastrointestinal and liver	211
Non-cancer liver disease	143
Cancer-Bladder and prostate	112
Cancer-Gynaecological	66
Diabetes, insulin dependent	30
Diabetes, non-insulin dependent	25
Alcoholism	18
Over 65 years old	7.5
Less than 65 years, no other condition	1

Question 3: Estimate the risk of serious illness from L. monocytogenes *in foods that support its growth and foods that do not support its growth at specific storage and shelf-life conditions.*

The risk assessment provides three approaches for answering the question: (i) the general consideration of the impact of the ingested dose on the risk of listeriosis; (ii) a comparison of four foods that were selected (according to diversity of prevalence and level of contamination, food composition and consumption patterns), in part, to evaluate the effect of *L. monocytogenes* growth or non-growth on risk; and (iii) the ability to conduct "what-if scenarios" for the evaluated foods that support growth of *L. monocytogenes*.

The results of the risk assessment show that the potential for growth of *L. monocytogenes* strongly influences risk, though the extent to which growth occurs is dependant on the characteristics of the food and the conditions and duration of refrigerated storage. Using the selected RTE foods, their ability to support the growth of *L. monocytogenes* appears to increase the risk of listeriosis 100- to 1000-fold on a per-serving basis. While it is not possible to present a single value for the increased risk for all RTE foods, because of the divergent properties of the foods, the ranges of values estimated in the risk assessment provide some insight into the magnitude of the increase in risk that may be associated with the ability of food to support the growth of *L. monocytogenes.* Control measures that focus on reduction of both frequency and levels of contamination have an impact on reducing rates of listeriosis. Controlling growth post-processing is one of these measures.

KEY FINDINGS

The most important key findings of the risk assessment as a whole are:

- The probability of illness from consuming a specified number of *L. monocytogenes* is appropriately conceptualized by the disease triangle, where the food matrix, virulence of the strain and susceptibility of the consumer are all important factors.
- The models developed predict that nearly all cases of listeriosis result from the consumption of high numbers of the pathogen.
- Based on the available data, there is no apparent evidence that the risk from consuming a specific number of *L. monocytogenes* varies for the equivalent population from one country to another. Differences in manufacturing and handling practices in various countries may affect the contamination pattern and therefore the risk per serving for a food. The public health impact of a food can be evaluated by both the risk per serving and the number of cases per population per year.
- Control measures that reduce the frequencies of contamination will have a proportional reduction in the rates of illness, provided the proportions of high contaminations are reduced similarly. Control measures that prevent the occurrences of high levels of contamination at consumption would be expected to have the greatest impact on reducing rates of listeriosis.
- Although high levels of contamination at retail are relatively rare, improved public health could be achieved by reducing these occurrences at manufacture and retail in foods that do not permit growth. In foods that permit growth, control measures such as better temperature control or limiting the length of storage periods will mitigate increased risk due to increases in *L. monocytogenes*.
- The vast majority of cases of listeriosis are associated with the consumption of foods that do not meet current standards for *L. monocytogenes* in foods, whether that standard is zero tolerance or 100 CFU/g.

LIMITATIONS AND CAVEATS

- The risk assessment focuses on four RTE foods and only examines them from retail to consumption.
- The risk characterization results are subject to uncertainty associated with a modelled representation of reality involving simplification of the relationships among prevalence, cell number, growth, consumption characteristics and the adverse response to consumption of some number of *L. monocytogenes* cells. However, the modelling is appropriate to quantitatively describe uncertainty and variability related to all kinds of factors and attempts to provide estimates of the uncertainty and variability associated with each of the predicted levels of risk.
- The amount of quantitative data available on *L. monocytogenes* contamination was limited and restricted primarily to European foods.
- Data on the prevalence and number of *L. monocytogenes* in foods came from many different sources, which adds to uncertainty and variability. Also, assumptions had to be made with regard to distribution of the pathogen in foods.
- The data used for prevalence and cell numbers may not reflect changes in certain commodities that have occurred in the food supply chain during the past ten years.
- The consumption characteristics used in the risk assessment were primarily those for Canada or the United States of America.
- The r-values and their distributions were developed using epidemiological data on the current frequency of *L. monocytogenes* strain diversity observed, with their associated virulence. If that distribution of virulence were to change (as reflected by new epidemiological data), the r-values would have to be re-calculated.
- There is uncertainty associated with the form of the dose-response function used, and with the parameterization. Also, the dose-response section of the hazard characterization is entirely a product of the shape of the distribution of predicted consumed doses in the exposure assessment component of the *Listeria* risk assessment undertaken in the United States of America (FDA/FSIS, 2001). Therefore its validity is dependant on the validity of the FDA/FSIS exposure assessment, and changes to that exposure assessment should lead directly to changes in the parameter, r.
- Predictive modelling was used to model the growth of *L. monocytogenes* in RTE foods, between the point of retail and the point of consumption, and the exposure assessment was based on information derived from those models. It is known that models may overestimate growth in food, and so reliance on such a model can result in an overestimation of the risk.

CONCLUSION

This risk assessment reflects the state of knowledge on listeriosis and on contamination of foods with *L. monocytogenes* when the work was undertaken, in 2002. New data is constantly becoming available, but in order to complete this work it was not possible to incorporate the very latest data in the risk assessment. A future iteration of the work would incorporate such new data.

The risk assessment provides an insight into some of the issues to be addressed in order to control the problems posed by *L. monocytogenes,* and approaches for modelling a system to evaluate potential risk management options. It addresses the specific questions posed by the CCFH and provides a valuable resource for risk managers in terms of the issues to be considered when managing the problems associated with *L. monocytogenes,* and alternative or additional factors or means to consider when addressing a problem. For example, if a limit is being established, then the technical feasibility of achievable levels of compliance must also be considered. While the available data were considered adequate for the current purposes, the risk assessment could be improved with additional data of better quality for every factor in the assessment. For example, quantification provides new perspectives on the risk posed by exposure to different doses of *L. monocytogenes*. The gaps in the database have been identified and could be used as a basis for establishing priorities for research programmes. The risk assessment improves our overall understanding of this issue and can therefore pave the way for risk management action to address this problem at the international level.

REFERENCES CITED IN THE EXECUTIVE SUMMARY

Aureli, P., Fiorucci, G.C., Caroli, D., Marchiaro, B., Novara, O., Leone, L. & Salmoso, S. 2000. An outbreak of febrile gastroenteritis associated with corn contaminated by *Listeria monocytogenes*. *New England Journal of Medicine*, **342**: 1236–1241.

Bille, J. 1990. Epidemiology of listeriosis in Europe, with special reference to the Swiss outbreak. pp. 25–29, *in:* A.J. Miller, J.L. Smith and G.A. Somkuti (eds). *Topics in Industrial Microbiology: Foodborne Listeriosis*. New York NY: Elsevier Science Pub.

Broome, C.V., Gellin, B. & Schwartz, B. 1990. Epidemiology of listeriosis in the United States. pp. 61–65, *in:* A.J. Miller, J.L. Smith and G.A. Somkuti (eds). *Topics in Industrial Microbiology: Foodborne Listeriosis*. New York NY: Elsevier Science Pub.

CAC [Codex Alimentarius Commission]. 2000. Report of the 33rd Session of the Codex Committee on Food Hygiene (CCFH), Washington, DC, 23–28 October 2000.

Dalton, C.B., Austin, C.C., Sobel, J., Hayes, P.S., Bibb, W.F., Graves, L.M. & Swaminathan, B. 1997. An outbreak of gastroenteritis and fever due to *Listeria monocytogenes* in milk. *New England Journal of Medicine,* **336**: 100–105.

FDA/FSIS [U.S. Food and Drug Administration/Food Safety and Inspection Agency (USDA)]. 2001. Draft Assessment of the relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods. Center for Food Safety and Applied Nutrition (FDA) and Food Safety Inspection Service (USDA) (Available at: www.foodsafety.gov/~dms/lmrisk.html). [Report published September 2003 as: Quantitative assessment of the relative risk to public health from foodborne *Listeria monocytogenes* among selected categories of ready-to-eat foods. Available at: www.foodsafety.gov/~dms/lmr2-toc.html].

McLauchlin, J. 1997. The pathogenicity of *Listeria monocytogenes*: A public health perspective. *Reviews in Medical Microbiology*. **8**: 1–14.

Mead, P.S., Slutsker, L., Dietz, V., McCraig, L.F., Bresee, J.S., Shapiro, C., Griffin, P.M. & Tauxe, R.V. 1999. Food-related illness and death in the United States. *Emerging Infectious Diseases*, **5**: 607–625.

Riedo, F.X., Pinner, R.W., De Lourdes Tosca, M., Cartter, M.L., Graves, L.M., Reeves, M.W., Weaver, R.E. et al**.** 1994. A point-source foodborne outbreak: Documented incubation period and possible mild illness. *Journal of Infectious Diseases,* 170: 693–696.

Roberts, D. 1989. *Listeria monocytogenes* in foods – results of two PHLS [Public Health Laboratory Service] surveys. (In Annual General Meeting and Summer Conference, [see FSTA (1990) 22 6A42].) *Journal of Applied Bacteriology*, **67**(6): xix.

Roberts, T. & Pinner, R. 1990. Economic impact of disease caused by *L. monocytogenes*. pp. 137–149, *in:* A.J. Miller, J.L. Smith and G.A. Somkuti (eds). *Topics in Industrial Microbiology: Foodborne Listeriosis*. New York NY: Elsevier Science Pub.

Rocourt, J. 1996. Risk factors for listeriosis. *Food Control*, **7**: 192–202.

Salamina, G., Dalle Donne, E., Niccolini, A., Poda, G., Cesaroni, D., Bucci, M., Fini, R., Maldin, M., Schuchat, A., Swaminathan, B., Bibb, W., Rocourt, J., Binkin, N. & Salmasol, S. 1996. A foodborne outbreak of gastroenteritis involving *Listeria monocytogenes*. *Epidemiology and Infection*, **117**: 429–436.