Food and Agriculture Organization of the United Nations

World Health Organization

AD HOC EXPERT CONSULTATIONS ON RISK ASSESSMENT OF MICROBIOLOGICAL HAZARDS IN FOODS

Joint FAO/WHO Expert Consultation on Risk Assessment of Microbiological Hazards in Foods

Hazard identification, exposure assessment and hazard characterization of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood

WHO Headquarters, Geneva, Switzerland

23 - 27 July 2001

ACKNOWLEDGEMENTS

The Food and Agriculture Organization of the United Nations and the World Health Organization would like to express their appreciation to the expert drafting groups (see Annex 3) for the time and effort which they dedicated to the preparation of thorough and extensive technical documents on exposure assessment and hazard characterization. The deliberations of this expert consultation were based on these documents.

The documents on which this report was based will undergo further development, a public comment period and a scientific peer review. Therefore, the information made available through this report and other sources is subject to revision until the risk assessments have been finalized and published by FAO and WHO. FAO and WHO declines any responsibility for errors and omissions in the information and data provided.

Contents

1. INTRODUCTION

The Food and Agriculture Organization of the United Nations (FAO) and the World Health Organization (WHO) convened an Expert Consultation on Risk Assessment of Microbiological Hazards in Foods in WHO Headquarters, Geneva, Switzerland from 23 - 27 July 2001. The list of participants is presented in Annex 1.

Dr Jorgen Schlundt, Coordinator, Food Safety Programme, Department of Promotion of the Human Environment, Sustainable Development and Healthy Environments Cluster in WHO opened the consultation on behalf of the two sponsoring organizations. In welcoming the participants Dr Schlundt stated that FAO and WHO are in the forefront of the development of risk based approaches for the management of public health hazards in food. In doing so, they are extending the experience and expertise developed in risk assessment of chemical hazards to microbiological hazards.

There is heightened global awareness of microbiological food safety and the need to reduce significantly the occurrence of foodborne illnesses. Dr Schlundt noted that the highest governing body of WHO, the World Health Assembly, recognized this fact when it met in May 2000 and for the first time issued a resolution that focused on the microbiological problems relating to food safety. The resolution also highlighted the need for science based involvement of public health considerations within the future standardization work in this area. Dr Schlundt also highlighted the activities of the Codex Alimentarius Commission (CAC) in the area of microbiological risk assessment. In response to requests from the CAC as well as to the needs of their member countries, FAO and WHO had embarked on a programme of activities with the objective of conducting risk assessments on specific pathogen-commodity combinations.

The consultation elected Dr. Servé Notermans (the Netherlands) as Chairperson of the expert consultation. Dr Henrik Wegener (Denmark) was appointed as Rapporteur. The consultation also appointed a chairperson and rapporteur for each of the working groups. Professor Tom Humphrey (United Kingdom) and Dr. George Nasinyama (Uganda) were nominated as Chairperson and Rapporteur respectively for the working group on *Campylobacter* spp. in broiler chickens. Professor Tom McMeekin (Australia) and Dr Ron Lee (United Kingdom) were nominated as Chairperson and Rapporteur respectively for the working group on *Vibrio* spp. in seafood.

2. BACKGROUND

Risk assessment of microbiological hazards in foods has been identified as a priority area of work for the CAC. At its 32nd session the Codex Committee on Food Hygiene (CCFH) identified a list of pathogen-commodity combinations for which it required expert risk assessment advice. In response, FAO and WHO, jointly launched a programme of work with the objective of providing expert advice on risk assessment of microbiological hazards in foods to their member countries and to the CAC.

Dr. Hajime Toyofuku, WHO, and Dr. Sarah Cahill, FAO provided participants with history of the FAO, WHO and Codex activities on microbiological risk assessment including the background to the current work. In their presentation, they also highlighted the objectives and expected outcomes of the current meeting. Dr Jean-Louis Jouve, Chief, Food Quality and Standards Service, FAO provided the expert consultation with guidance on how to conduct their review and evaluation of the background documents.

The FAO/WHO programme of activities on microbiological risk assessment aims to serve two customers - the CAC and the FAO and WHO member countries. The CAC, and in particular the CCFH, has requested sound scientific advice as a basis for the development of guidelines and recommendations as well as the answers to specific risk management questions on certain pathogen-commodity combinations. Member countries on the other hand need specific risk assessment tools to use in conducting their own assessments and, if possible, some modules that can be directly applicable to a national risk assessment.

To implement this programme of work, FAO and WHO are convening a series of joint expert consultations. To date two expert consultations have been convened to address risk assessment of *Salmonella* spp. in broiler chickens and eggs and *Listeria monocytogenes* in ready-to-eat foods. In March 2001 FAO and WHO initiated risk assessment work on *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood, both of which were identified as priority issues by the CCFH (see Annex 2). Two *ad hoc* expert drafting groups were established to examine the available relevant information on the abovementioned pathogen-commodity combinations. These groups prepared background documentation on hazard identification, exposure assessment and hazard

characterization of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood. These documents were reviewed and evaluated by the joint expert consultation.

The purpose of this report is to present the summary of the draft documents on hazard identification, hazard characterization and exposure assessment on *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood as well as the discussions and recommendations of the expert consultation.

3. OBJECTIVES OF THE CONSULTATION

The consultation examined the information provided by the expert drafting groups on hazard characterization and exposure assessment of *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood with the following objectives;

1. To critically review the documents prepared by the *ad hoc* expert drafting groups giving particular attention to:

- the scope of the work and the approach taken or the proposed approaches to undertake the risk assessments of these pathogen-commodity combinations;
- the assumptions on which the exposure assessments and hazard characterizations are or will be based;
- the associated uncertainty and variability;
- the data needed to improve and complete the work.

2. To provide scientific advice to FAO and WHO member countries on the risk assessment of *Campylobacter spp.* in broiler chickens and *Vibrio* spp. in seafood based on the available documentation and the discussions during the expert consultation.

3. To identify the areas in which risk management guidance is needed from the CCFH to further define the future direction of the work.

4. SUMMARY OF THE GENERAL DISCUSSIONS

The drafting groups presented overviews of the hazard identification, exposure assessment and hazard characterization components of the risk assessments on *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood to the expert consultation. A summary of this and the discussions of the expert consultation are given in sections five and six of this report.

The expert consultation acknowledged and expressed its appreciation for the body of work that had been carried out by the drafting groups and the quality of the material presented.

The consultation formed two working groups addressing *Campylobacter* and *Vibrio* respectively. The composition of the two working groups is outlined in the tables below.

<u>*Campylobacter* spp. in broiler chickens</u>

Independent experts	**Expert members of the drafting groups**
Louis Anthony Cox, United States	Bjarke Bak Christensen, Denmark
Marja-Liisa Hänninen, Finland	Aamir Fazil, Canada
Tom Humphrey, United Kingdom	Emma Hartnett, United Kingdom
Servé Notermans, The Netherlands	Anna Lammerding, Canada
Susana María de los Milagros Jiménez, Argentina	Greg Paoli, Canada
Paul Mead, United States	Hanne Rosenquist, Denmark
George Nasinyama, Uganda	
Henrik Wegener, Denmark	

Vibrio spp. in seafood

Independent experts	Expert members of the drafting groups
Awa Kane Aïdara, Senegal	Angelo DePaola, United States
Dorothy-Jean McCoubrey, New Zealand	I. Karunasagar, India
Ron Lee, United Kingdom	Ken Osaka, Japan
Tom McMeekin, Australia	John Sumner, Australia
Noel Murray, New Zealand	Mark Walderhaug, United States
Mitsuaki Nishibuchi, Japan	
Mark Tamplin, United States	
Paul Brett Vanderlinde, Australia	
Shigeki Yamamoto, Japan	

The expert consultation discussed the approaches taken by the two expert drafting groups to respond to the risk management questions posed by the 33rd session of the CCFH and found that the approaches in general were sound. It was recognized that there are inherent challenges and problems relating to developing "globally applicable risk assessments" based on national risk assessments, or, in the absence of national risk assessments, from relevant data available in different countries. Also, the expert consultation realized that the current FAO/WHO microbiological risk assessment work could not reach the level of detail achievable in national microbiological risk assessment work. This was due to the need for it to be widely applicable but also due to the limited resources available to the sponsoring organizations.

The expert consultation recognized that the availability of suitable data for microbiological risk assessment was a critical issue. For example, it identified data in relation to food consumption patterns and food handling practices in different countries as a very important issue for the development of internationally applicable risk assessment tools. In relation to data availability the expert consultation noted that the FAO/WHO "Calls for data" were attempting to address this. However, it felt that the current process had limitations and was unlikely to reach the attention of all relevant data contributors due to language barriers and the fact that their distribution was almost exclusively by electronic means. It was considered that this process could be improved by addressing the language and distribution issues and also by providing potential contributors with detailed guidelines for data collection and a template for data submission.

The expert consultation recommended that dialogue between risk assessors and risk managers should be enhanced to provide timely feedback on model creation and documentation and to better serve the needs of the risk managers. The consultation suggested that presentations by a representative member of the expert drafting groups at the CCFH would be a productive means of increasing understanding of the potential uses and limitations of microbiological risk assessment among risk managers and enabling the CCFH to better identify their risk assessment needs.

The consultation noted that currently the risk assessment work that had been carried out on both *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood was weighted towards the situation in developed countries, primarily due to differences in data availability in developing and developed countries. However, due to the international nature of this work the expert consultation recommended that both expert drafting groups attempt to further include the situation in developing countries in the draft risk assessments.

4.1 *CAMPYLOBACTER* SPP. IN BROILER CHICKEN

The expert consultation found that the approach taken and the assumptions made in the draft *Campylobacter* risk assessment were acceptable. However, it was noted that there were a large number of uncertainties relating to important areas in the farm-to fork model, primarily due to lack of data to develop and validate the models.

The expert consultation acknowledged that in the current draft of the *Campylobacter* farm-to-table model the various components were not yet fully integrated, and needed further development before estimates of the risk to public health and the efficacy of interventions to reduce *Campylobacter* could be generated. Although, this was felt to be a limitation in reviewing the model, the expert consultation recognized the capacity of the farm-to-table model to identify gaps in data and felt that it could be used to stimulate relevant research on *Campylobacter*.

Furthermore, when finalized and further validated the expert consultation was of the opinion that it would constitute an important contribution to the management of risks to public health posed by *Campylobacter* spp. in broiler chickens.

The expert consultation identified several areas in which it recommended the expert drafting group should focus particular attention in completing the risk assessment. These included the introduction of *Campylobacter* in poultry flocks, the application of specific interventions in poultry processing plants and the preparation of meals outside the home. Furthermore, it recommended that FAO and WHO identify means of validating the model when completed.

The expert consultation noted that risk management questions posed by the CCFH in relation to *Campylobacter* spp. in broiler chickens were identical to those formulated for *Salmonella* spp. in broiler chickens. However, due to the significant differences between these two pathogens the consultation felt that the preparation of a risk profile prior to the formulation of the risk management questions could have focused these questions to better address the particularities of *Campylobacter*.

4.2 *VIBRIO* SPP. IN SEAFOOD

The expert consultation accepted the logic of using an available model (that of *Vibrio parahaemolyticus* in United States oysters) as the basis for the development of globally applicable models for the same organism in oysters and other seafood products, and for its extension to other *Vibrio* spp.. It was stressed that appropriate data from a number of countries needed to be included in order to achieve this.

The expert consultation noted that appropriate data were needed in order to include differences in seafood consumption and preparation patterns, aquaculture and harvesting practices as well as biological effects introduced by different species of shellfish, crustacea and fish in the model. These were in addition to the more readily identifiable variables of water and air temperature, water salinity, prevalence and number of pathogenic vibrios in the environment and the proportion of strains presumed to be pathogenic.

It was noted that only example mitigations had been included in the model for *V. parahaemolyticus* in oysters in order to demonstrate the way in which these could be incorporated. There was a need for the risk managers to identify the various mitigations that should be included. It might not be necessary to have comprehensive models in order to identify the relative effects of different intervention strategies.

The expert consultation recognized the considerable resources needed for completion of the four separate *Vibrio* risk assessments identified to date and the difficulty of completing all of these to a satisfactory standard in the identified timeframe. There was a need to review the work involved and resources available and for the CCFH to identify which of the assessments were of greatest importance.

5. HAZARD IDENTIFICATION, HAZARD CHARACTERIZATION AND EXPOSURE ASSESSMENT OF *CAMPYLOBACTER* SPP. IN BROILER CHICKENS

5.1. EXECUTIVE SUMMARY

An understanding of *Campylobacter* spp., and specifically *Campylobacter jejuni* in broiler chickens is important from both public health and international trade perspectives. As a result, there is an urgent need to evaluate this pathogen-commodity combination by quantitative risk assessment methodology.

The objective of this work was to undertake the first steps of a risk assessment that will ultimately provide estimates for 1) the risk of illness from pathogenic *Campylobacter* spp. in broiler chickens consequential to a range of levels in raw poultry for the general population and various susceptible population groups and 2) the change in exposure and illness likely to occur for different interventions in primary production, processing, handling and preparation of poultry. This report describes the first three steps of the risk assessment. It will be concluded at a future date with the completion of the risk characterization. The final report will also include discussion and evaluation of potential intervention strategies and uncertainty analysis. Methods of validating such a model will be explored.

5.1.1 Hazard identification of *Campylobacter* spp. in broiler chickens

Campylobacter is the leading cause of zoonotic enteric infections in most developed countries. Human cases are usually caused by *C. jejuni* or to a lesser extent by *Campylobacter coli.* Most human *Campylobacter* infections are classified as sporadic cases or as part of small family related outbreaks. Identified outbreaks are not common. Information on the burden of human *Campylobacter* infections for the developing countries is very limited. However, it is likely that the rate of campylobacteriosis is especially high among children below 2 years of age causing substantial morbidity and eventually mortality.

The principal reservoir of pathogenic *Campylobacter* spp. is the alimentary tract of wild and domesticated mammals and birds. *Campylobacter* is commonly found in poultry, cattle, pigs, sheep, wild animals and birds, and in dogs and kittens. *C. jejuni* is predominantly associated with poultry and *C. coli* is predominantly found in pigs.

It is well recognized that poultry products can be contaminated with *Campylobacter*. However, *Campylobacter* is also found in beef, pork, other meat products, raw milk and milk products, fish and fish products, fresh vegetables and modified atmosphere packaged foods such as unsmoked bacon and salad vegetables.

Campylobacter may be transmitted from the reservoirs to humans by direct contact with contaminated animals or animal carcasses or indirectly through the ingestion of contaminated food or water. Case-control studies conducted worldwide have repeatedly identified handling raw poultry and eating poultry products as important risk factors for sporadic campylobacteriosis. Other food related risk factors that have repeatedly been identified include consumption of other meat types, undercooked or barbecued meat, raw seafood, unpasteurized milk or dairy products and drinking untreated surface water. Eating meat cooked outside the home (at restaurants) and not washing the kitchen cutting board with soap (indicating cross-contamination) have also been identified as risk factors. Other risk factors include exposures when travelling abroad, contacts with pets and farm animals, and recreational activities in nature. Person-to-person transmission is apparently infrequent, because infected humans constitute a minor reservoir for *C. jejuni*, and asymptomatic excretion of *Campylobacter* is uncommon.

5.1.2 Hazard characterization of *Campylobacter* spp. in broiler chickens

Introduction

This section focuses on evaluating the nature of adverse health effects associated with foodborne *Campylobacter* spp. and how to quantitatively assess the relationship between the magnitude of the foodborne exposure and the likelihood of adverse health effects occurring. In this document, human infection refers to the status of pathogen persistence and multiplication within the gastrointestinal tract with or without symptoms. Illness refers specifically to the state where overt symptoms occur as a result of the infection.

Objective

The objective and scope of the *Campylobacter* hazard characterization is to provide:

- a review of the characteristics of the host, organism and food matrix effects;
- a summary and review of available data and information on adverse health effects;
- a dose-response model based on human feeding study data.

Approach

Information was compiled from published literature and from unpublished data submitted to FAO and WHO by public health agencies and other interested parties. The first section of the document provides a description of the public health outcomes following infection and including sequelae, pathogen characteristics influencing its ability to elicit infection and illness, host characteristics that influence the acquisition of infection, and food-related factors that may affect the survival of *C. jejuni* in the human gastrointestinal tract.

The second section of the hazard characterization document presents the data and methods available to derive a dose-response relationship for *C. jejuni*. The ultimate goal was to derive a dose-response model that mathematically describes the relationship between the numbers of organisms that might be present in a food and consumed (the dose), and the human health outcome (the response). In order to achieve this, human feeding trial data for two strains of *C. jejuni* were used. The data were used to derive estimates for the probability of infection as well as the probability of illness.

Key findings

The current document has attempted to synthesize and summarize the information that was available either in the literature, or through the call for data to describe the factors that influence the likelihood of an individual becoming infected, ill, and developing sequelae. The quantification of the importance of most of these factors requires a substantial amount of additional research. Nevertheless, the information and analysis conducted does allow some advances to be made in estimating the risk from *Campylobacter* and putting into context the apparent importance of other contributory factors.

The probability of any pathogen initiating an infection is influenced to various degrees by three factors. These include the pathogen and host characteristics and the matrix or conditions of ingestion. The influence of specific components within these three factors was qualitatively described based on current thinking. Unfortunately, there is currently insufficient information on which to base a detailed analysis, that would allow fine distinctions to be drawn between, for example, the probability of illness upon ingestion of any one strain versus any other, or ingestion in milk versus water, or for an individual who is taking medication versus a very young child.

The adverse effects that can occur following infection with *C. jejuni* were also summarized. They included acute gastroenteritis and non-gastrointestinal sequelae such as reactive arthritis, Guillain-Barré syndrome (GBS), and Miller-Fisher syndrome. Reactive arthritis has been estimated to occur in approximately 1% of patients with campylobacteriosis. Guillain-Barré syndrome is a serious paralytic condition, which has been estimated to occur once in every 1000 cases. Finally, Miller-Fisher syndrome, which is considered to be a variant of Guillain-Barré syndrome is also reported to occur, however, there are no estimates on the frequency of the occurrence of this condition following campylobacteriosis.

The hazard characterization also describes dose-response models that can be used to mathematically describe and estimate the probability of infection following the ingestion of a dose of *C. jejuni*. The dose-response equations used were based upon the single-hit hypothesis, fit to human feeding trial data conducted using healthy volunteers and two strains of *C. jejuni* (Figure 5.1). It was proposed that pooling the infectivity data for the two strains from the feeding trial study may be appropriate, and this offers a new interpretation of the available information.

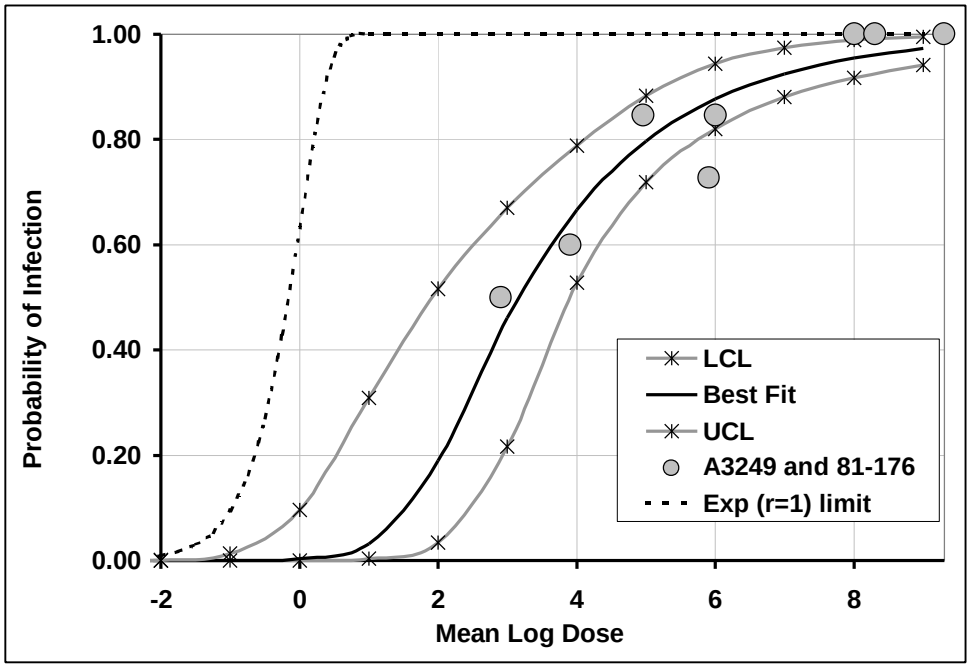

FIGURE 5.1 Beta-Poisson dose-response relationship for the probability of infection for *C. jejuni* based on human feeding trial data and two strains (A3249 and 81-176) and (model parameters, $\alpha = 0.21$, ß = 59.95)
LCL - Lower confidence limit UCL - Upper confidence limit

The probability of illness is conditional upon the probability of infection. Using the data from the human feeding trial study, there does not appear to be a clear trend for the behaviour of this conditional probability. When the data for both strains are pooled, the conditional probability tends to exhibit a dose-independent relationship (Figure 5.2). It is important to recognize that although the conditional probability is dose-independent, the ultimate probability of illness increases with ingested dose.

In conclusion, the probability of infection upon ingestion of a dose of *C. jejuni* can be estimated with the caveat that the data are from a feeding study involving healthy volunteers, and using a milk matrix and a limited number of *Campylobacter* strains. The impact of population immunity, sub-population susceptibilities or other factors cannot be quantified from the data. The probability of illness following infection can also be estimated using a dose-independent probability. Some researchers have proposed a decreasing conditional probability based on consideration of only one of the two *Campylobacter* strains. Again, the impact of other factors, such as susceptibility, on the probability of illness cannot be quantified due to a lack of adequate epidemiological data and resolution to this level. Finally, the progression of the illness to more serious outcomes and the development of some sequelae can be crudely estimated from the approximate proportions reported in the literature.

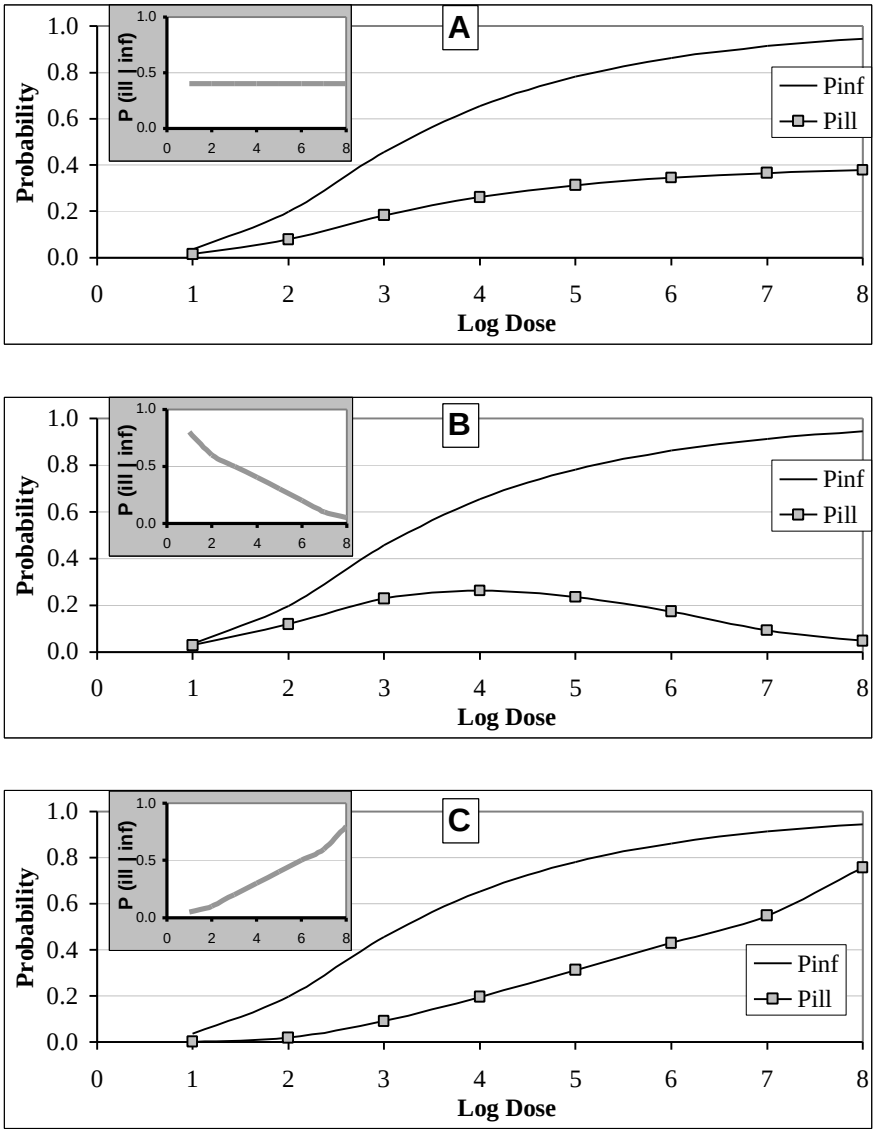

FIGURE 5.2 Hypothetical probability of illness curves, influenced by three alternative conditional probabilities. The conditional probability assumption is shown in the inset curves.

(A)Conditional probability independent of dose;

(B) Conditional probability decreasing with dose;

(C) Conditional probability increasing with dose.

Gaps in the data

- Data on strain variability regarding virulence/pathogenicity.
- Studies on the mechanisms of infectivity, virulence/pathogenicity in the human host.
- Quantitative information about infection and illness rates at low doses and with other strains of C. jejuni ranging from 10^0 to 10^9 organisms.
- Complete epidemiological data from outbreak studies including enumeration of *Campylobacter* in suspected food items or in drinking water, numbers of people exposed, attack rates, and demographics of those exposed, particularly immunocompromised population groups and, children under the age of five.
- Data describing the impact of acquired immunity resulting from recent exposure to *Campylobacter*.
- Studies to elucidate the true number of human infections caused by *Campylobacter*, including GBS etc., and to determine the burden of disease attributable to different sources of *Campylobacter*.
- To utilize an alternative approach to dose-response for management purposes in a country that lacks enumeration data in their system, several pieces of country specific information are required. Data are needed on the number of *Campylobacter* illnesses associated with chicken in that country in a time interval (e.g. per year). Coupled with this, the prevalence of contaminated chickens at a point in the farm-to-table chain is also required. The further down-stream the prevalence estimation (i.e. closer to the consumer) the more utility in the approach for risk mitigation strategies.

5.1.3 Exposure assessment of *Campylobacter* spp. in broiler chickens

Introduction

To evaluate the risk posed to the human population by the presence of *Campylobacter* spp. in broiler chickens, an exposure assessment model was developed. The aim of this assessment was to estimate the likelihood and magnitude of exposures to *Campylobacter* as a result of consumption of a chicken meal. Exposure assessments that consider *Campylobacter* spp. in broiler chickens have been developed independently by Canada, Denmark and the United Kingdom. However, each of these models specifically focused upon the localized situation.

Objectives

The objective of the exposure assessment was to develop a model that details the prevalence and numbers of *Campylobacter* throughout the production chain from farm-to-table. However, the model presented to the expert consultation only considered whole fresh broiler chickens, prepared for consumption by oven roasting in the home.

Approach

The approach taken was described in detail in the background document on *Campylobacter* spp. in broiler chickens that was prepared for and presented to the expert consultation. In its preparation, it was felt that the most efficient means of facilitating discussion at the expert consultation would be to limit the detailed descriptions within the exposure assessment section to five key stages; rearing, transport, processing, cross-contamination and cooking. These stages were chosen according to their level of development and their predicted importance in contributing to the overall confidence in the final risk assessment results. Other stages were described only superficially and will be included for discussion at future expert consultations.

Farm-processing

The exposure assessment was approached in a farm-to-table manner (Figure 5.3). The model framework is modular in nature and each stage of the supply chain is described by a distinct mathematical model. This provides a flexible tool for risk managers, which may be used to estimate the risk to public health and investigate the impacts of potential interventions. Only meals prepared in the home and cooked using oven-roasting are currently considered but this could be expanded in the future.

The exposure assessment begins by considering the *Rearing and Transport* module of the supply chain. The aim of this module is to estimate the probability that a broiler chicken will be colonized and the probability a bird will be contaminated on the exterior at the point of slaughter. The levels of colonization and external contamination associated with any given bird are also considered. From this point, the slaughter of the birds and

subsequent stages of processing are investigated, corresponding to the second module of the overall framework, *Slaughter and Processing*. The output from this module is the probability that a chicken carcass is contaminated with *Campylobacter* at the end of processing, and the associated level of contamination on such a product. The exposure assessment concludes with the final module, *Preparation and Consumption*. This module addresses the preparation of a product in the home environment and subsequent cooking. The result is an estimate of the probability an individual is exposed to at least one *Campylobacter*, along with a measure of the numbers of *Campylobacter* cells ingested. Each of these models is stochastic and incorporates the inherent uncertainty and variability associated with the model through the use of Monte-Carlo simulation. The integration of the modules outlined above then feeds into hazard characterization. Each of these modules is described in detail below.

FIGURE 5.3 Model framework for the risk assessment of *Campylobacter* spp. in broiler chickens.

Rearing and transport

To estimate the colonization and contamination status of a broiler chicken at the point of slaughter two parameters are used. These are a measure of the national prevalence of flocks that contain at least one colonized bird and the within-flock prevalence of such a flock. The probability that a bird is colonized at slaughter is then the product of these two factors. Here, a colonized bird is defined as a positive bird, and a flock that contains at least one positive bird is defined as a positive flock.

Data are often available to estimate the flock prevalence; however, data may not be available to estimate the within-flock prevalence of positive flocks. Therefore, a dynamic model describing the colonization process of a flock following exposure has been developed. In brief, the model assumes that the transmission of *Campylobacter* within a flock is initiated by an exposure event, which results in the colonization of a single bird. Once this first bird is successfully colonized, transmission ensues amongst the birds with which the first colonized bird makes contact on a daily basis, that is, the birds' social cluster. This continues until a threshold is reached where the level of contamination in the feed, and water supply is sufficient to result in the colonization of an exposed bird. From here onwards colonized birds appear randomly throughout the entire flock. This process continues until either all the birds become colonized or depopulation occurs and the birds are removed for slaughter.

The within-flock transmission dynamics may depend upon the source of *Campylobacter*. The above description applies to the colonization of the first bird as a result of some point source. However, if one considers exposure as a result of contaminated feed or water, under such circumstances a large proportion of the flock will be exposed and colonization of birds is likely to occur randomly throughout the flock. Further, if vertical transmission occurs it may be that initially there are several birds that are colonized and hence initiate the process of flock colonization. Therefore, the model is developed such that the dynamics of within-flock transmission are dependent upon the source of the organism. Colonization levels are estimated by use of experimental data.

A consequence of the colonization of a flock is the external contamination of the birds in that flock. This occurs either by self-contamination for a bird which is colonized and hence likely to become contaminated as a result of the excretion of *Campylobacter* in the bird's faeces or, for a bird not necessarily colonized, by contact with faeces containing *Campylobacter*. This contamination is then magnified during the transportation of the flock to the slaughter facility. This is as a result of the dispersal of contaminated faeces throughout the vehicle.

To predict the extent to which birds become contaminated on their exteriors, it is assumed that the colonization process of a flock can be described spatially on a lattice structure. Each bird has an associated location on the lattice. The colonization process is modelled as described above, such that at depopulation the location of each colonized bird is known on the lattice. In this way, the location of the colonized birds in the transport vehicles is also known and hence the consequence of the shedding of *Campylobacter* in the faeces and the impact this has upon the contamination of the exterior of a given bird can be predicted. Estimation of the levels of contamination are based upon experimental data, however, only one data set is currently available and hence does not allow for variability in transport times that may occur. As such, the model does not currently incorporate the length of time of

transport, although this may be an important factor in predicting the impact of transport on contamination levels. If such information should become available, it can be incorporated in future model development.

The contamination of the exterior of birds is not unique to positive flocks. Experimental studies suggest that birds from negative flocks can become contaminated on their exteriors at some point prior to slaughter. However, the frequency and extent of such contamination is currently unknown due to lack of data. An assumption is made that a bird from a negative flock can become contaminated with 1% of the contamination on a random bird from a positive flock. However, in the absence of data the impact this has upon the probability and levels of contamination on the exterior of a random bird at slaughter, as predicted by the model, and the validity of these predictions is unknown.

Slaughter and processing

The stages considered by the model in this module are stun and kill, scald, de-feathering, evisceration and wash and chill. The focus of the processing of broiler chickens is the level of external contamination on the bird/carcass and the manner in which this changes through processing. These changes occur due to reduction as a direct result of the processing stage itself, cross-contamination from other birds and self-contamination from the caecal contents of the bird.

To estimate the impact of the processing stages upon the level of contamination on a bird/carcass and the prevalence of contaminated carcasses a simulation model has been developed. This model utilizes the outputs from the rearing and transport module and generates a profile for a random bird. More specifically, a bird is assigned a colonization status, a contamination status and associated levels of colonization and contamination. The stochastic effects of each of the processing stages upon the level of contamination of a bird and hence the prevalence of contaminated birds is then predicted.

To model the stochastic effect of each stage quantitative measurements are required for the number of *Campylobacter* on a carcass before and after each of the processing stages considered. These data represent the variability between birds that occurs at each stage. Available data are sparse and only small data sets are currently available. The true extent of the inter-bird variability is uncertain. Further, published data sets often report only mean values of a number of samples. Numerous combinations of effects could have occurred to produce these mean values so further uncertainty is present. To quantify the level of uncertainty, second-order modelling is adopted through non-parametric methods that make no assumptions regarding the true form of the variability. The level of uncertainty in any model results as a consequence of the absence of data can be visualized. A further complication is the data reported do not sample the same bird before and after each stage, but random birds from the flock. Small sample sizes were used. For example, in one study only four birds were sampled. The extent of the effect a stage has on an individual bird level is therefore difficult to assess. The model development process has highlighted the importance of the availability of appropriate data, reported in a manner appropriate for use in quantitative risk assessment modelling.

The data incorporated into the model is taken from a variety of sources and hence involves a number of processing plants and production methods. In this way variability that exists between processing plants is incorporated in the model framework.

As a direct result of the nature of the data available, the model currently does not explicitly consider the evisceration process. However, a conceptual model has been developed for this stage of processing but was not presented to the expert consultation. Should more data become available this can easily be incorporated in to the current framework. The model considers different methods of chilling, these are air chilling, and water chilling with and without the addition of chlorine. In this way the model is adaptable to different processing systems.

Post-processing transportation and storage

These steps have been considered in the model development, but were not included in the document presented to the expert consultation. Future exposure assessment descriptions will include this module.

Preparation and consumption

Cross-contamination in the home

Two models for cross-contamination were developed. One describes exposure via fluid "drip" exuded from the uncooked broiler carcass and ingested via some pathway, for example on fingers, or via contact with other foods. This model is a mechanistic approach related to the water that a chicken gains through processing and is subsequently released. Loosely attached cells will enter into this fluid and may be inadvertently ingested.

The second is a "contact transfer" model that quantifies the number of *Campylobacter* cells transferred from the raw chicken to preparation surfaces (cutting board, utensils, etc.) or hands, and subsequently from the preparation surface to a prepared meal. The organisms may also be ingested directly by for example licking on fingers.

Cooking

Three modelling approaches were examined to describe the fate of *Campylobacter* during thermal heating by oven-roasting of whole carcasses. Once an appropriate modelling approach has been determined, the model can be extended to other cooking styles.

The first approach, the "internal temperature approach", was based on the calculation of thermal death through a sequence of time-steps during roasting of chicken. The temperature, which determines the thermal death in each step through standard D-value calculations, was based on observations of the time-temperature profile in the centre of the drumstick portion of a roasted chicken. The final temperature achieved during the cooking process was based on observed internal temperatures achieved in domestic kitchens.

The second approach, the "protected areas approach", was based on the designation of areas in the carcass where the least heat treatment is achieved, presumably due to increased thermal insulation from the heat source. In this approach, it was assumed that any *Campylobacter* outside of these designated areas are killed. The approach then required assumptions regarding the proportion of pathogens that are found in these areas, and the maximum temperature achieved in this area. The thermal inactivation was estimated by D-value calculation for the final temperature in this area and the assumed time spent at this temperature.

The third approach, the "heat transfer approach", was designed to predict the time-temperature profile at various depths below the surface of chicken based on a simplified thermodynamic model of heat transfer through chicken meat. This allows for prediction of thermal inactivation as a function of depth and time. The final consumer exposure was highly dependent upon the final temperature achieved at each depth and the assumed proportion of cells that were located at each depth in the carcass.

At the current stage of development, the models are being evaluated with respect to the validity of required assumptions, the degree of conservatism which is implicit in the approach and the relative value of complex versus simple models given the amount of uncertainty in the location of cells in or on carcasses with respect to thermal insulation.

Key Findings

On farm

The transportation of flocks to slaughter may be a crucial stage in predicting microbial levels at slaughter. However, this is an area often not considered by researchers and more data are needed.

Processing

It was recognized that much of the data that is reported in the literature is difficult to use in risk assessment due to the methods employed and the style of the reporting. As an example, data on changes in pathogen load on carcasses is frequently reported as the mean of log concentrations for only a few carcasses. The research community should consider the statistical power of such studies and be more critical of the ability of such study designs to give definitive results for the purpose of assessing risk and intervention efficacy.

Cross-contamination

At present two models have been developed, both of which consider the overall probability of exposure to *Campylobacter* during a food preparation event. Comparison of the models shows that, despite the different approaches taken, they seem to be mathematically equivalent with an appropriate choice of assumptions. One of the two models may be preferable at a later date depending on the data that will become available in future.

It is difficult to model cross-contamination based on the information currently available. Further improvement of the model and model validation may be extremely difficult given the complexity of cross-contamination, the many possible pathways by which it can occur, and the variability in the behaviour of individuals in the kitchen.

Cooking

Based on thermal inactivation calculations, it is difficult to reconcile the assumed importance of undercooking as a cause of human exposure with the assumption that contamination of broiler carcasses with *Campylobacter* is on the external or internal surface of the carcass (or very close to the surface). Resolution of this inconsistency requires the allocation of some amount of contamination to various places within the carcass where *Campylobacter* are significantly insulated from heat. While it is possible to demonstrate and to hypothesize that *Campylobacter* will, on occasion, be found in such a place, it is very difficult to quantify the frequency and extent of this particular mode of contamination. It may also call into question the importance of surface contamination with respect to consumer exposure due to undercooking. Clearly surface contamination will remain a key component for exposure via cross-contamination.

Gaps in the data

On-Farm

- Data on the routes of *Campylobacter* infection of broiler chicken flocks.
- Survey data on the prevalence of *Campylobacter* in slaughtered flocks and within flocks.
- Studies on the dynamics of within flock transmission.
- Data on the probability of contamination of a bird during transport.
- Data on production systems in different countries and regions.

Processing

- Prevalence and enumeration data for poultry before and after various processing steps such as scalding, defeathering, evisceration, washing and chilling.
- External contamination levels of broilers at slaughter from both positive and negative flocks and the relationship between this and time of transport.
- Prevalence and enumeration data comparing various methods of chilling - air chilling, water chilling, water chilling with chlorine, etc..
- Data describing the actual cross-contamination between positive and negative flocks and within positive flocks during the different slaughter processes.
- Prevalence and enumeration data comparing different scalding temperatures and different packaging methods.
- Data on the relationship between the concentration on neck skin samples and the concentration on the whole chicken in order to calculate a conversion factor.
- Data on the microbial implications of carcass deboning.

Cross-contamination

- Survey data and direct observational data on consumer practices in preparation and handling of chicken that especially detail to which extend different contamination pathways may contribute to exposure.
- Research data detailing quantities of *Campylobacter* that are transferred to and from surfaces and hands and from preparation surfaces to the final meal during preparation of chicken. Data on the concentration of *Campylobacter* in the fluid attached to the chicken.
- Survey data and direct observational data on preparation and handling practices of chicken in restaurants and other retail establishments
- .Survey data on consumption patterns.
- Data on the number of *Campylobacter* cells in the fluid attached to the chicken.

Cooking

- Distribution of pathogens in places other than the surface of chicken carcasses before and after cooking.
- Final temperatures achieved in oven roasting in different areas of the carcass.

- Importance of the cooling phase after removal from the oven.
- Relationship between observational definitions of undercooking (e.g. self-assessed, or "pink chicken") and the actual heat treatment experienced.

5.2. SUMMARY OF THE DISCUSSIONS

5.2.1 Hazard identification of *Campylobacter* spp. in broiler chickens

The quality of data on the human incidence of *Campylobacter* infection varies across countries, reflecting differences in surveillance systems and microbiological techniques used. Most of the available information was summarized in the document presented to the expert consultation but it was recommended that newly available sentinel surveillance data on *Campylobacter* infections in the Netherlands and the United Kingdom also be included. The expert consultation noted that there appears to be differences in the pathogenicity of *Campylobacter* species and that for example the apparent low pathogenicity of *C.. lari* should be mentioned. The expert consultation agreed that *Campylobacter* is an important source of human foodborne illness.

The expert consultation noted the importance of distinguishing between the reduction in prevalence and reduction in levels of contamination of *Campylobacter* on retail products. In discussing the infectivity of so called "viable-but-non-culturable" *Campylobacter* the expert consultation noted that there are conflicting data in the literature concerning this issue and felt that in some published work, the techniques used to assess "culturability" were not sufficiently sensitive.

Risk Factors

There are several sources of infection with *Campylobacter* spp. but the main one is believed to be poultry. However, this may differ from country to country or region to region. Therefore, it was suggested that additional data is obtained if possible or otherwise it should be generated. The relative importance of risk factors is not fully addressed and the expert consultation recommended the inclusion of epidemiological data in this section. For example, the results of recent and ongoing intervention studies such as those in Belgium and Iceland should be included as they become available. Risk factors for campylobacteriosis in developing countries may be different from those in developed countries and this should also be considered. Furthermore, the epidemiology of campylobacteriosis in developing countries and the role of acquired immunity needs to be addressed. The effect of seasonality on the prevalence of *Campylobacter* was noted and should be considered when discussing reservoirs of this pathogen.

There is a need for consistency in the use of terminology (e.g. birds are "colonized" but humans are "infected" with *Campylobacter).* In addition, it should be stated that infection precedes clinical symptoms and does not necessarily result in disease.

5.2.2 Hazard characterization of *Campylobacter* spp. in broiler chickens

Although acquired immunity is likely to play a role in the risk of human infection, the consultation agreed that 15-25 year olds may be more susceptible and or more frequently exposed. Age definitions need to be made clearer for the purposes of identifying population groups of increased susceptibility or exposure.

The consultation recognized that antimicrobial resistance might compromise treatment in patients with diarrhoea and bacteremia. It recommended that existing risk assessments on antimicrobial resistant *Campylobacter* be considered in the future development of this work.

Dose-response analysis

Dose-response data were only available from one feeding study on young, apparently healthy males, using only two strains of *C. jejuni*, both of which were clinical isolates. The limitation of developing a dose-response curve from such limited data was recognized. Although limited, the data showed a positive correlation between the exposure dose and the probability of infection. This correlation was not evident for disease, possibly due to the small sample size. Given the current data limitations, the expert consultation concurred with the decision to pool data from the two strains. However, more data were needed to establish a sound dose-response correlation in relation to illness. Some of those identified by the expert consultation included:

- data on dose-response for other sections of the population including the more susceptible human population groups;

- data for other strains of *Campylobacter* and the differences between strains in relation to dose-response; and,
- data on the effect of food matrices on dose-response.

The fact that milk was used as the food carrier for *Campylobacter* in the abovementioned feeding trial raised concern about the protective role played by the food matrix. However, in the absence of other data, it was assumed that the protection afforded by milk was at least as good as that of chicken.

The consultation agreed that the dose-response model developed was a reasonable one but should be applied with caution. It may overestimate the frequency of illness in developing countries due to acquired immunity, and may likewise underestimate the frequency of illness due to differences in susceptibility of a population group. If other strains of *Campylobacter* act with more or less efficacy than those used in the feeding trial the dose-response model may need to be modified accordingly.

5.2.3 Exposure assessment of *Campylobacter* spp. in broiler chickens

The strong need to develop a model for assessing exposure to *Campylobacter* was recognized. It was the opinion of the expert consultation that a model such as the one currently being developed may help to inform risk management decisions and to assess the risk to human health.

Colonization on the farm

An overview of data made available from case-control studies on the factors involved in the introduction of *Campylobacter* to poultry flocks on the farm should be presented. In addition, data are required on the major cause(s) of seasonality of *Campylobacter* colonization in broilers. This is currently unknown although some of the assumed risk factors are reduced in winter (e.g. numbers of wild birds are reduced, airflow is out of the houses, temperature is reduced, snow on earth etc.). There is a need to include information from some countries that are succeeding in reducing the prevalence of positive flocks (e.g., the United Kingdom, Sweden and Denmark). Farmers are becoming more successful in either excluding *Campylobacter* from the flock or delaying colonization. This means not all birds will be *Campylobacter* positive pre-slaughter. Information should be provided on the possibilities to limit the introduction of *Campylobacter* into a flock (biosecurity) and to mitigate its spread if introduced (immunization, feed additives, etc.).

Transportation

Transportation of broiler chickens may further spread contamination within the flock due to spreading of faecal material over the birds. Where the distance between the farm and the processing plant is short, contamination is largely restricted to the outside of the birds. It was the opinion of the expert consultation that this contamination is easily reduced during scalding and further processing. As such, it may be neglected as a source of *Campylobacter* on the final carcass. However, longer periods of transportation could influence cross-contamination, gut colonization and excretion levels. The model on transport needs to take this into consideration. In addition, the model should consider the potential effects of feed withdrawal on the amount of faeces produced and the levels of *Campylobacter* in faeces.

Other preharvest intervention strategies

The consultation recognized that the model should include intervention strategies. These may include:

- improved biosecurity;
- organic acids in feed;
- possible competitive exclusion or vaccination.

Slaughter and processing

During processing of chickens, the bacterial flora present on the outer surface of the broiler chickens will fluctuate. During scalding a proportion of the *Campylobacter* present will be washed off and a proportion killed by heat. In the succeeding processes (defeathering and evisceration), further contamination may occur. The predominant source of contamination is faecal spread during these processes. Once carcasses are contaminated, reduction of bacterial flora is limited. Therefore, in most Hazard Analysis Critical Control Point (HACCP) plans, faecal contamination during evisceration is a critical control point. Although the slaughtering techniques and equipment are steadily improving, faecal spread cannot be completely avoided.

It was recommended that the statement in the background document beginning 'Welfare of the live birds and carcass quality are top priorities...' be replaced to reflect the opinion of the expert consultation that economics are the main controlling factor in poultry processing. The model requires data on changes on *Campylobacter* numbers on poultry carcasses at critical points during processing. The consultation recommended that the section on poultry processing should include discussion on the survival of *Campylobacter* and consider that the strains may differ in their survival abilities.

The current model assumes that the effects of stunning and killing are negligible. This statement may need to be qualified in the light of information from the United States that indicates that water in the electric stunning bath can be *Campylobacter* positive. Birds may inhale this water and this may lead to systemic contamination.

Available data indicate that the processes that have been used during poultry slaughter have not significantly reduced *Campylobacter* contamination rates on chickens. It is also observed that when a *Campylobacter* positive flock is processed, contamination levels vary between 10^2 to 10^4 per gram of skin. As a consequence of this observation and the absence of processes that significantly reduce contamination, it was advised to consider simplifying the models.

Changes due to defeathering

Water usage and pressure force applied by the plucking fingers are considered important parameters affecting recontamination and cross-contamination during defeathering. For example, the present model does not take account of faecal material that may be introduced onto bird surfaces by the expulsion, during defeathering, of faeces from the gut as a result of physical pressure. However, such data may not be currently available.

Changes due to evisceration

The current model does not include the effects of evisceration. Although it was acknowledged this might have an impact if the viscera rupture, data on the evisceration step are currently only available for birds where the viscera did not rupture.

Effects of washing and other treatments

The model should allow for additional interventions geared at reducing the bacterial load after processing of carcasses including treatment with organic acids, irradiation and hot water baths.

Effects of chilling and freezing

Broiler chickens comprise whole birds or portions that can be either fresh or frozen. There are several methods for cooling carcasses. These include immersion cooling in a spin chiller and spray or air chilling. These processes influence both the water content of the meat and the bacterial load present. Data showed that the effect of air chilling on *Campylobacter* present was not significant. However, new developments in air chilled technologies demonstrate that a reduction in *Campylobacter* organisms is achievable. The relative proportions of contaminated chickens cooled under air compared to water chilled systems is country specific and can be handled in the model. The model can also represent effects of intervention measures such as use of chlorinated, ozonized or electrolyzed water. The expert drafting group informed the consultation that a model was also available on the effects of freezing but that this model was not included in the background document .

Post-processing changes

There is a need for the model to account for the period between the processing plant and the home. The model as presented to the expert consultation did not include a retail component and it was recommended that this be included in the further development of the model.

Home Preparation

As there are many differences in the preparation of broiler chicken products, data on this area would be relevant for assessing the final risk to the consumer and in identifying risk factors during preparation.

Differences in preparation between countries comprise, among others, the proportion of chicken prepared at private homes and restaurants and the method of preparation (conventional oven, fan oven, microwave, boiling, frying, barbecuing). The model should be adapted so that it is able to examine risk factors associated with preparation methods and commercial preparation of chicken for consumption in restaurants, hotels and institutions.

Model for cross-contamination

The consultation recommended that the term "drip" fluid should be explicitly defined in the context of the cross-contamination model developed. It was agreed that the concepts used in deriving both the "drip" and "contact transfer" models were plausible and, therefore, should be retained for further elaboration. However, validation of the models would be difficult due to lack of data. It was noted that the "contact transfer" model provides the possibility to model interventions.

The current "drip" model uses volumes of fluid between 0.5 and 1.5ml as the potential volume of liquid released from the chicken. Given that many air-chilled chickens are comparatively dry it was suggested that the lower limit should be reduced to 0.1ml for these types of chickens.

The consultation also proposed changing the name of the "drip" fluid model due to concerns that in countries where air chilling is used, and the drip from chicken is very low, this may lead to misinterpretation of the model.

There was also a need to acknowledge that some chickens are sold as portions and not whole carcasses. The model does not currently address this and this is a limitation. A further limitation is that the model deals with only one pathway, that of a whole chicken coming into the home and being roasted.

Exposure via cooked chicken

In light of epidemiological studies implicating "undercooked" chicken as a risk factor for human campylobacteriosis, it is reasonable to include a component for human exposure via this vehicle in the model. Of the three approaches presented, "internal temperature", "protected areas" and "heat transfer", it was felt that conceptually the latter was the most reasonable. However, because of lack of data for both the "heat transfer" and "protected areas" approaches, the consultation felt that it was important that all the approaches be retained in the model. This may also help to account for the many different ways in which chicken may be cooked. The consultation recommended that the collection of data was needed to further develop and validate the heat transfer model. A microbiological survey to determine the frequency of contamination of cooked chicken was discussed, however, the consultation felt that such a study would be impractical.

Consumption data

Due to differences in consumption patterns between countries and regions, specific surveys are needed in this area.

5.2.4 Conclusions and recommendations

General

The expert consultation commended the expert drafting group for the enormous amount of work done both before and during the expert consultation. In continuing their work, the consultation acknowledged and welcomed the drafting groups' plans to carry out uncertainty analysis of the final model and explore ways to validate such a model.

Hazard characterization and dose-response

Conclusions

The experts concluded that the present dose-response model is the best that can be produced with the existing limitations in data and should be put forward for public debate and for future validation. Validation of the model may come about through analytical epidemiological investigations and descriptive epidemiological studies.

Recommendations

Epidemiological studies that can serve to validate the dose-response model should be carried out and the data made available to the drafting group. Such studies may include carefully conducted outbreak investigations, intervention studies and other epidemiological approaches. To be of value such studies should collect information on attack rates among exposed persons, the amount of food ingested, the level of contamination within that food, and sampling strategies.

Exposure assessment

The strong need to further develop the model for assessing exposure to *Campylobacter* was recognized and the expert consultation was of the opinion that such a model will help to inform risk management decisions and to assess the risk to human health.

Conclusions

The expert consultation felt the early farm-to-table model was valuable in identifying data gaps and sampling strategies that can stimulate relevant research on *Campylobacter* in the different stages of the farm-to-table continuum. It is likely that components of the model will be capable of producing useful information within the next year. However, given the extensive nature of some of the data gaps, it is also likely that the development and validation of the full model will require longer than one year.

Recommendations

The expert consultation identified the following areas in the risk assessment as needing particular attention in the next year's work:

- The pre-harvest module
  - Introduction of *Campylobacter* into poultry flocks
  - Reducing levels of *Campylobacter* in the gut of poultry
- Processing
  - Effects of cooling and freezing, chlorination, lactic acid treatments and irradiation on levels of *Campylobacter*
- Distribution and processing from slaughterhouse to home (wholesale, retail)
  - Effects of storage conditions on colony forming unit (CFU) counts, cross-contamination, etc.
- Preparation of meals
  - Meals prepared outside the home should be considered in the model
  - Data to validate the cross-contamination and cooking elements in the model are needed.
- The situation in developing countries
  - The risk assessment does not to a very large extent take the situation in developing countries into account, primarily due to data gaps. These data gaps urgently need to be filled.

5.3 ISSUES TO BE BROUGHT TO THE ATTENTION OF FAO AND WHO

5.3.1 Risk management questions

In identifying *Campylobacter* in chicken as a priority area in which it required expert risk assessment advice the CCFH[1] outlined two risk management questions as follows:

1) Estimate the risk from pathogenic thermophilic *Campylobacter* in chicken (broilers) consequential to a range of levels in raw poultry for the general population and for various susceptible population groups (elderly, children, and immuno-compromised patients).

2) Estimate the change in risk likely to occur for each of the interventions under consideration including their efficacy.

[1] ALINORM 01/13A Report of the thirty third session of theCodex Committee on Food Hygiene *Washington DC, 23 -28 October 2000*

- Reduce the prevalence of positive flocks
  - Destruction of positive breeder and chicken flocks (broiler) flocks
  - Vaccination of breeding flocks
  - Competitive Exclusion
- Reduce the prevalence of positive birds at the end of slaughter
  - Use chlorine in water chilling of chicken (broilers)
  - Water chilling vs. air chilling for chicken (broilers)
- Evaluate the importance of various routes for introduction of pathogenic *Campylobacter* into flocks including feed, replacement birds, vectors and hygiene

The CCFH furthermore mentioned that a risk profile could be carried out to focus the work before embarking on a risk assessment.

The expert consultation noted that the risk management questions for the risk assessors were not very well tailored to the particular problem. A risk profile could have helped in identifying relevant risk management questions in particular in relation to interventions. Because of the lack of a risk profile, specific interventions were not identified at the outset. Nevertheless the drafting group has taken into consideration a range of different relevant interventions during their model development.

The expert consultation felt that the approach taken by the risk assessors to answer the risk managers' questions, the development of an integrated farm-to-table mathematical model, was a useful one, and potentially the best approach. The consultation furthermore acknowledged that major research gaps need filling to complete and validate the model. These data gaps are not likely to be filled in the short term.

5.3.2 Data

The expert consultation recommended that FAO and WHO promote the harmonization of methods used in both surveillance of human illness and food monitoring.

Due to the very limited data sets available for modelling dose-response and the difficulties of conducting further human feeding trials for *Campylobacter,* it was recommended that FAO and WHO promote the collection of quantitative data from outbreak investigations in member countries.

In relation to exposure assessment, FAO and WHO should promote the generation and collection of quantitative data throughout the food chain.

6. HAZARD IDENTIFICATION, HAZARD CHARACTERIZATION AND EXPOSURE ASSESSMENT OF *VIBRIO* SPP. IN SEAFOOD

6.1. EXECUTIVE SUMMARY

The objective of the work was to undertake the first steps of a risk assessment of *Vibrio* spp. in seafood products that would have the most impact on public health and/or international trade. These first steps involve taking a risk assessment developed by a member country, generalizing it, and testing its ability to provide predictions that are useful for other member countries. Furthermore, it is desirable to explore the capability of a risk assessment model to be adapted to different commodities and/or related organisms of international and national interest. The approach used by the drafting group was to quantify those illnesses caused by *Vibrio* spp. in different countries following the consumption of a range of seafoods. Three species, *Vibrio parahaemolyticus, Vibrio vulnificus* and *Vibrio cholerae* were considered as the species responsible for most illnesses caused by *Vibrio* spp. This report describes the approach suggested to undertake a risk assessment of these three species in specific seafood products.

With regard to pathogen-food commodity combinations, the expert drafting group proposed to undertake a detailed risk assessment of *V. parahaemolyticus* in oysters as a model was already available. The proposed work on *V. vulnificus* would demonstrate the application of the previous model to a different organism with appropriate modifications. Non-oyster associated *V. parahaemolyticus* is important in Japan and some other countries, and consideration of the organism with respect to finfish would give a different viewpoint of the same organism used in the first model, but with cross-contamination as an important factor. *V. cholerae* is an important pathogen in developing countries and the development of a model for the organism in shrimp would provide a tool to investigate a number of other scenarios. It would also enable an investigation of the risk associated with international trade of this product and of the problems caused with respect to the export market for potentially contaminated shrimp. Since shrimp are usually eaten cooked it would provide an additional example of the use of a cross-contamination model for another *Vibrio* spp..

6.1.1 Hazard identification of *Vibrio* spp. in seafood

Vibrio spp. are Gram-negative, facultatively anaerobic rod-shaped bacteria. The genus contains twelve species that can cause food-borne illness (Table 1), most of which is caused by *V. cholerae*, *V. parahaemolyticus* or *V. vulnificus* (Oliver and Kaper, 1997, Dalsgaard, 1998). Some species are primarily associated with gastrointestinal illness (*V. cholerae* and *V. parahaemolyticus*) while others can cause non-intestinal illness, such as septicaemia (*V. vulnificus*).

In tropical and temperate regions, disease-causing species of *Vibrio* occur naturally in marine, coastal and estuarine (brackish) environments and are most abundant in estuaries. Pathogenic vibrios can also be recovered from freshwater reaches of estuaries (Desmarchelier, 1997). The occurrence of these bacteria does not correlate with numbers of faecal coliforms and depuration of shellfish may not reduce their numbers. Based on data from the United States, there is a positive correlation between water temperature and both the number of human pathogenic vibrios isolated and the number of reported infections, a correlation particularly marked for *V. parahaemolyticus* and *V. vulnificus.*

TABLE 1: *Vibrio* spp. which cause, or are associated with, human infections (after Dalsgaard, 1998)

	Occurrence in human clinical specimens*	
	Intestinal	Non-intestinal
V. cholerae O1	++++	+
V. cholerae non-O1	++	++
V. parahaemolyticus	++++	+
V. fluvialis	++	-
V. furnissii	++	-
V. hollisae	++	-
V. mimicus	++	+
V. metschnikovii	+	+
V. vulnificus	+	+++
V. alginolyticus	-	++
V. carchariae	-	+
V. cincinnatiensis	-	+
V. damsela	-	+

* The symbol (+) refers to the relative frequency of each organism in clinical specimens and (-) indicated that the organism was not found

In Japan (Twedt, 1989; Japanese Ministry of Health, 2000) and eastern Asian countries *V. parahaemolyticus* has been recognised as a major cause of foodborne gastroenteritis. By contrast, in most countries outside of Asia, the reported incidence appears to be low, perhaps reflecting a different mode of seafood consumption. Gastroenteritis caused by this organism is almost exclusively associated with seafood consumed raw or inadequately cooked, or contaminated after cooking. In the United States prior to 1997 illness was most commonly associated with crabs, oysters, shrimp and lobster (Twedt, 1989; Oliver and Kaper, 1997). Four *V. parahaemolyticus* outbreaks associated with the consumption of raw oysters were reported in the United States in 1997 and 1998 (DePaola *et al.*, 2000). A new *V. parahaemolyticus* clone of O3:K6 serotype emerged in Calcutta in 1996. It has spread throughout Asia and to the United States elevating the status of *V. parahaemolyticus* to pandemic (Matsumoto *et al.*, 2000). In Australia, in 1990 and 1992, there were two outbreaks of gastroenteritis caused by *V. parahaemolyticus* in chilled, cooked shrimps imported from Indonesia (Kraa, 1995) and there was also a death in 1992 associated with the consumption of oysters.

V. vulnificus has been associated with primary septicaemia in individuals with chronic pre-existing conditions, following consumption of raw bivalves. This is a serious, often fatal, disease. To date, *V. vulnificus* disease has almost exclusively been associated with oysters (Oliver, 1989; Oliver and Kaper, 1997). Recently, *V. vulnificus* infections have been associated with a variety of raw seafood products in Korea and Japan (Personal Communication, Dr. Yamamoto, Japan).

Toxigenic *V. cholerae* O1 and O139 are the causative agents of cholera, a water- and food-borne disease with epidemic and pandemic potential. Non-O1/non-O139 strains may also be pathogenic but are not associated with epidemic disease. Non-O1 strains are generally nontoxigenic, usually cause a milder form of gastroenteritis than O1 and O139 strains,-and are usually associated with sporadic cases and small outbreaks rather than epidemics (Desmarchelier, 1997).

Outbreaks of cholera have been associated with consumption of seafood including oysters, crabs and shrimp (Oliver and Kaper, 1997). The largest outbreak was a pandemic in South America in the early 1990s when *V. cholerae* O1 caused more than 400,000 cases and 4,000 deaths, in Peru (Wolfe, 1992). Contaminated water used to prepare food, including the popular, lightly marinated fish *ceviche*, was the cause of the outbreak.

Given the foregoing, the drafting group concluded that four pathogen-product risk assessments should be progressed:

- *Vibrio parahaemolyticus* in raw oysters
- *Vibrio vulnificus* in raw oysters
- *Vibrio parahaemolyticus* in finfish consumed raw
- *Vibrio cholerae* in shrimp from developing countries for domestic and export consumption.

Accordingly, the drafting group prepared an exposure assessment and hazard characterization for each these pathogen-commodity combinations. The justification for each pathogen-commodity combination is contained in a "Statement of Purpose" included at the head of each assessment.

References[2]

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6.1.2 Hazard characterization of *Vibrio* spp. in seafood

Introduction

This section focuses on evaluating the nature of adverse health effects associated with *Vibrio* spp. in seafood and how to quantitatively assess the relationship between the magnitude of the foodborne exposure and the likelihood of adverse effects occurring. The hazard characterization presents dose-response curves for three important species of *Vibrio*: *V. parahaemolyticus, V. vulnificus* and *V. cholerae.* Infection by *V. parahaemolyticus,* and *V. cholerae* is characterized by an acute gastroenteritis. Therefore, the end-point of the dose-response curve is defined as gastroenteritis. *V. vulnificus* can occasionally cause mild gastroenteritis in healthy individuals, but for specific subpopulations *V. vulnificus* can cause a serious septicaemia that frequently leads to death in susceptible people. Therefore, the endpoint for the dose-response curve is defined as septicaemia.

Objective

The objective of the hazard characterization is to provide sufficient information to allow for a quantitative measurement of the public health risk from *Vibrio* spp. associated with the consumption of seafood and foods potentially contaminated by seafood. The quantitative measurement of public health risk is accomplished by the determination of dose-response relationships for each *Vibrio* spp. based upon the best available data. These data are often sparse and the resulting dose-response relationship is uncertain. This uncertainty of the dose-response curve is accounted for by representing the dose-response relationship in the form of a family of plausible data-derived dose-response curves.

Approach

Human volunteer studies are available for the construction of dose-response curves for *V. parahaemolyticus* and *V. cholerae*. These data were analysed using curve-fitting routines to find a best fit for the Beta-Poisson dose-response curve. Because of the sparse data for the human volunteer studies, multiple curve-fits are determined using resampling techniques. The resulting multiple Beta-Poisson dose-response curves may be used in the risk characterization. These multiple dose-response curves represent a key uncertainty in the risk assessment. For *V. vulnificus,* no human volunteer data were available and an alternate approach was attempted. The dose-response relationship was estimated by fitting a Beta-Poisson model using monthly *V. vulnificus* levels in the United States Gulf of Mexico oysters and estimated consumption of raw oysters with monthly reported cases of *V. vulnificus*-associated septicaemia in the United States. With further research, this risk relationship may be applied in a *V. vulnificus* risk assessment and validated with data on the distribution of *V. vulnificus* in raw oysters at the point of retail.

Key findings

A review of the literature was undertaken to identify and characterize the infectivity and genetic factors of *Vibrio* spp. to be modelled. *V. parahaemolyticus* and *V. cholerae* have both pathogenic and non-pathogenic forms based on the presence of specific virulence factors: *tdh* (thermostable direct hemolysin) and *trh* (*tdh*-related hemolysin) for *V. parahaemolyticus* and cholera toxin for *V. cholerae*. There is not adequate information to

differentiate between virulent and avirulent strains of *V. vulnificus*. Therefore, all *V. vulnificus* strains were considered to be equally pathogenic. Relevant factors with respect to the host and food matrix have been identified and where data are available may be incorporated into the model.

Reasonable Beta-Poisson dose-response parameters were obtained from data sets for all three organisms examined; however, the human volunteer studies characterize dose-response relationships for pathogens administered with a pH-neutralizing buffer rather than for pathogens administered with a food matrix.

Figure 6.1 shows the most probable dose-response curve for *V. parahaemolyticus*; however, the family of curves representing uncertainty that surrounds the curve is not shown. These data are from healthy human volunteer studies where gastrointestinal illness was used as the endpoint response.

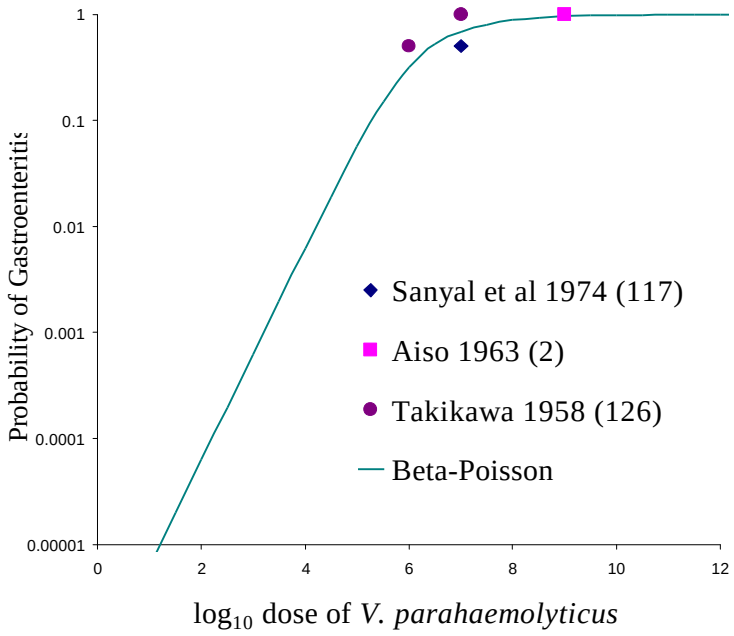

FIGURE 6.1 Beta-Poisson dose-response curve for *V. parahaemolyticus (*endpoint modelled is gastrointestinal illness*).*

Figure 6.2 shows the most probable fits of human volunteer data for several biotypes and serotypes of *V. cholerae*. Once again, uncertainty is represented by a family of curves surrounding each of the most probable curves, but this is not shown. The endpoint modelled is gastrointestinal illness. The data indicate that the consumption of *V. cholerae* with foods may significantly shift the dose-response curve to the right, indicating that a higher dose of *V. cholerae* is needed to cause illness in a comparable number of volunteers when the *V. cholerae* are consumed with food. It is unknown if specific food matrices have greater or lesser effects on the shift of the dose-response relation.

Figure 6.3 shows the dose-response relationship for *V. vulnificus* as estimated based upon exposure of the United States susceptible oyster-consuming population and the United States *V. vulnificus* epidemiology reports. As with the other *Vibrio* spp., a family of dose-response curves was generated using resampling techniques (bootstrap) but this was not shown. The derived curve is different compared to the other *Vibrio* spp. because a septicaemia endpoint is modelled instead of a gastrointestinal illness endpoint.

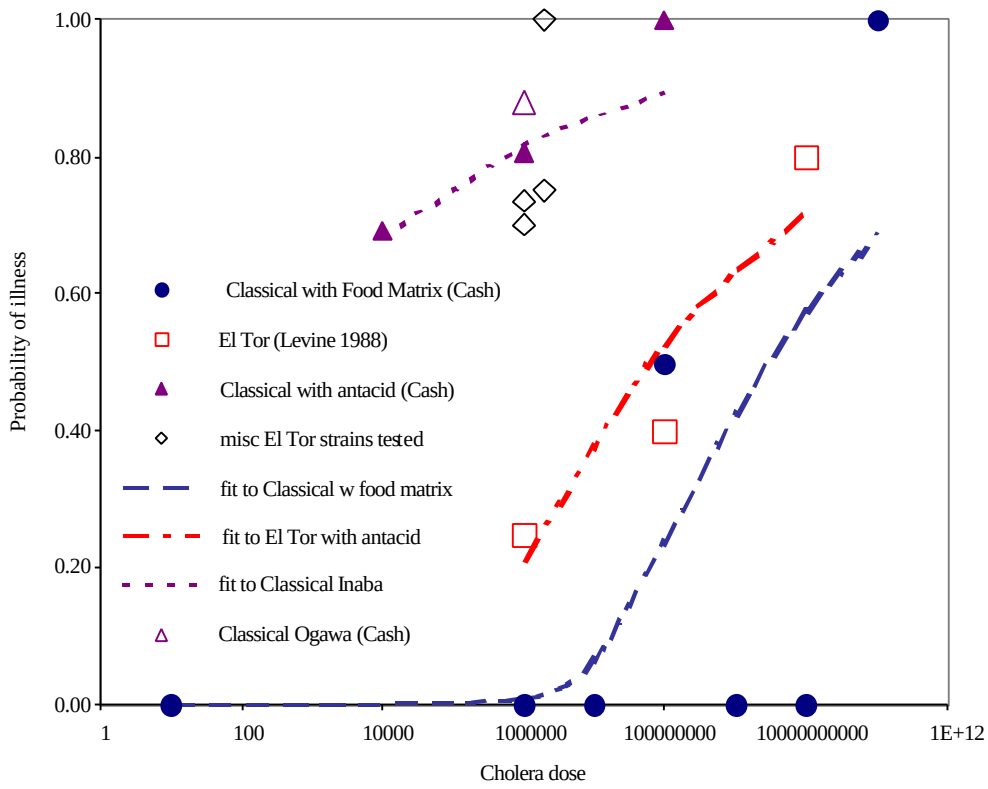

FIGURE 6.2. Beta-Poisson dose-response curves for *V. cholerae* (endpoint modelled is gastrointestinal illness)

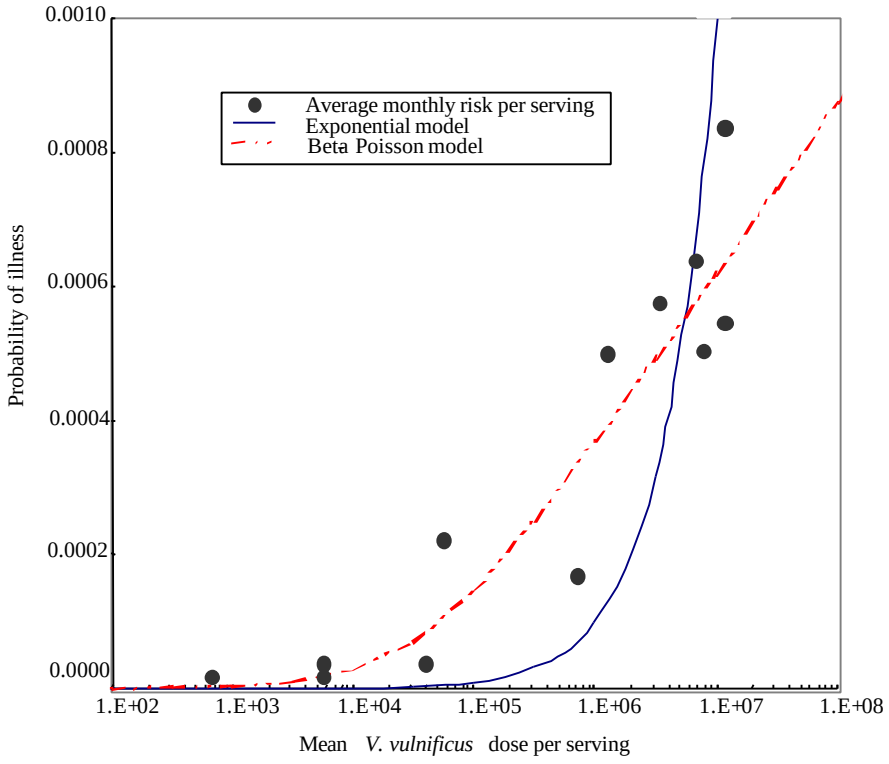

FIGURE 6.3. Beta-Poisson dose-response curve for *V. vulnificus* (endpoint is septicaemia)

Gaps in the data

- Few human volunteer studies have been carried out - more data are needed and these could be obtained through the collection of quantitative data from outbreak investigations.
- As there is only a small amount of available data on the effect of the food matrix on dose-response, additional data is required in this area.
- There is a need for ongoing *V. vulnificus* environmental and epidemiological surveillance data to test and to refine the *V. vulnificus* dose-response relationship.
- There is a lack of information for the characterization of human susceptibility and pathogen virulence variability.

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6.1.3. Exposure assessment of *Vibrio* spp. in seafood

6.1.3.1 Exposure assessment of *Vibrio parahaemolyticus* in raw oyster

Introduction

In the United States during 1997 and 1998 there were more than 700 cases of illness due to *V. parahaemolyticus*, the majority of which were associated with the consumption of raw oysters. In two of the 1998 outbreaks a serotype of *V. parahaemolyticus* previously reported only in Asia, O3:K6, emerged as a principal cause of illness for the first time. It was suggested that warmer than usual water temperatures were responsible for the outbreaks.

In 1999, the United States Food and Drug Administration (FDA) initiated a risk assessment to characterize the public health impact of consuming raw oysters contaminated with *V. parahaemolyticus*. The FDA Draft Risk Assessment on the Public Health Impacts of *V. parahaemolyticus* in Raw Molluscan Shellfish (FDA-VPRA) was released for public comment in 2001. The FDA-VPRA contains several key linkages between prevalence of *V. parahaemolyticus* in oysters and temperature, most notably temperature of harvest waters and of oysters throughout the post-harvest-retail-consumption continuum.

Temperature profiles in the oyster industry of other countries e.g. New Zealand, Australia and Japan indicated the opportunity for growth of pathogenic *V. parahaemolyticus* to potentially dangerous levels. However, the public health statistics of these countries do not reflect any impact due to this organism in oysters.

Accordingly, an exposure assessment will be undertaken on *V. parahaemolyticus* in oysters using data from Australia, Canada, Japan, New Zealand and the United States.

Objectives

The objectives are to:

- Quantify the exposure of consumers to pathogenic *V. parahaemolyticus* from consumption of raw oysters.

- Extend this exposure assessment to consumers in other countries that have oyster industries.

Approach

The approach being taken is to use the FDA-VPRA model as the base and further develop it to accommodate data inputs from other countries. This model incorporates all phases in the harvest - post-harvest - consumption continuum in three modules (Figs 6.4-6.6).

Data for the exposure assessment were obtained via a call for data issues by to FAO and WHO. The data were then analysed for incorporation into the risk assessment model.

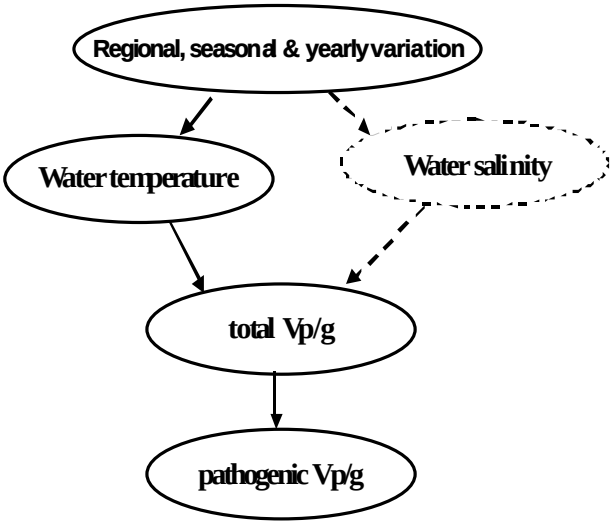

FIGURE 6.4: Harvest module for exposure assessment of *V. parahaemolyticus* in oysters

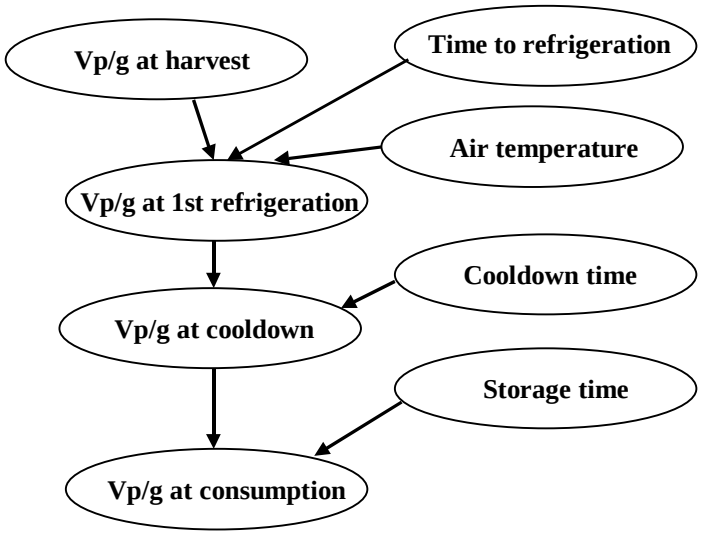

FIGURE 6.5: Post-harvest module for exposure assessment of *V. parahaemolyticus* in oysters

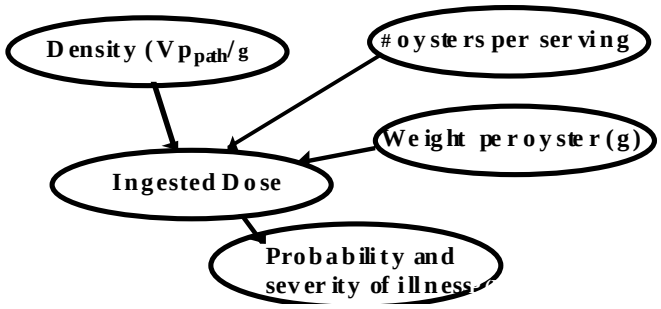

FIGURE 6.6: Consumption module for exposure assessment of *V. parahaemolyticus* in oysters

Key findings

Analysis of the data has not been completed but the key findings at the present stage are as follows:

- In Japan, while *V. parahaemolyticus* is the cause of significant seafood-based illness, oysters appear not to be an important vehicle. It may be that regulatory practices of controlling the total coliform number in harvesting seawater result in the curtailment of harvesting in the summer months.
- In Australia and New Zealand, some oyster-producing areas have seawater temperature and salinity combinations, together with post-harvest temperatures that are potentially favourable for proliferation of *V. parahaemolyticus*. This situation should also be considered in the model.
- It may be that not all oyster species are similarly vulnerable to proliferation of pathogenic *V. parahaemolyticus* in the post-harvest chain. For example, the Sydney Rock Oyster (*Saccostrea commercialis*) is particularly resistant to stress during post-harvest handling.

Gaps in the data

For each country the following gaps exist:

- Incidence/frequency of pathogenic *V. parahaemolyticus* in water and shellfish.
  - Factors that affect incidence of pathogenic *V. parahaemolyticus* in the environment.
  - Growth rate of *V. parahaemolyticus* within oysters at temperatures other than 26°C, including data on the potential differences in the growth rate of pathogenic strains versus total *V. parahaemolyticus* populations.
- Potential virulence factors of pathogenic strains other than *tdh* and *trh*, e.g. urease, enterotoxins.
- Consumer and industry handling practices for oysters.
- Consumption patterns of oysters in each country.

6.1.3.2 Exposure assessment of *Vibrio vulnificus* in raw oyster

Introduction

This document outlines the objectives and approach for modelling the risk of *V. vulnificus* from consumption of raw oysters. This pathogen-commodity pair was proposed by the European Community in the 33rd session of the CCFH.

The general approach and many of the parameters may be adopted from the FDA-VPRA, which is the only available risk assessment for a *Vibrio* spp. in raw oysters. Thus the approach taken by the drafting group was to elaborate on the FDA-VPRA. Due to the lack of appropriate data outside of the United States for many of the model inputs this assessment relies almost totally on data from this country. The approach for determining dose-

response uses exposure and illness frequency. Because of this approach there are some elements of hazard characterization included in the exposure assessment.

The choice of the United States data is intended only to provide an example on how to apply the exposure model to a different national situation. This model will be further tested when appropriate data from other countries or situations become available.

Objectives

- To adapt the FDA-VPRA model to assess the risk from *V. vulnificus* associated with the consumption of raw oysters.
- To identify the most appropriate data as well as the data gaps and limitations for modelling *V. vulnificus* in oysters.

Approach

- Examine *V. vulnificus* ecology and the epidemiology of *V. vulnificus* illness.
- Test the appropriateness of transferring inputs for harvest, postharvest and public health modules of FDA-VPRA to the risk assessment of *V. vulnificus* (Figure 6.7).
- Select data inputs and develop alternate approaches from FDA-VPRA to fit *V. vulnificus*.
- Validate the predicted exposure with a survey of *V. vulnificus* levels at retail.
- Determine the dose-response by establishing a mathematical relationship between exposure and illness for each month of the year within a defined geographical region.
- Develop a conceptual model (schematic diagram) of the *V. vulnificus* risk assessment model showing integration of all the modules.

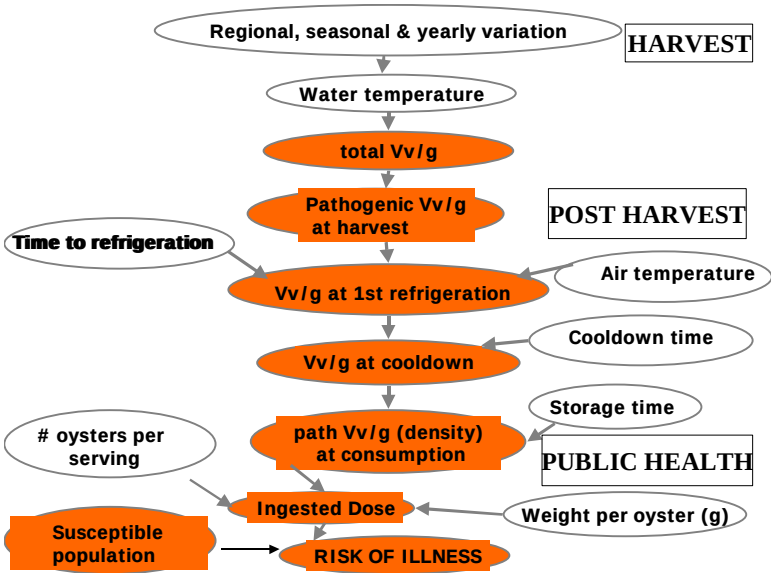

FIGURE 6.7 Schematic diagram of the *V. vulnificus* conceptual risk assessment model showing integration of all the modules. Inputs that are not shaded can be transferred directly from the FDA-VPRA and inputs that are shaded require additional data

Key findings

- The FDA-VPRA provides many of the needed inputs and is a useful framework to model the risk of *V. vulnificus* septicaemia from consumption of raw oysters.

- Reliable data are available for determining *V. vulnificus* exposure from consumption of raw United States Gulf Coast oysters during each month of the year (levels at harvest and consumption, growth and survival in raw oysters).
- Reliable monthly illness rates are available due to intensive epidemiological surveillance, the severity of primary septicaemia caused by this organism and the nearly exclusive association of illness in the United States to consumption raw Gulf Coast oysters.
- Aggregate population dose-response can be approximated from available data on differences in exposure from consumption of the United States Gulf Coast oysters and reported illness frequency during warm and cold months.
- The approach for determining dose-response circumvents the lack of data on frequency of virulent strains in raw oysters and uncertainty concerning the susceptible population by assuming that these do not vary from month to month.
- This modelling approach is appropriate for determining the potential effectiveness of specific mitigations.
- The dose-response relationship developed using data for raw oyster consumption in the United States may be used for:
    - Other foods for which the distribution of *V. vulnificus* at the point of consumption is known assuming there is no matrix effect
    - Other countries, but adjustments would need to be made for differences in the susceptible population compared to the United States.

Gaps in the data

- There is insufficient exposure data available for modelling risk of *V. vulnificus* illness from consumption of raw Gulf Coast oysters in the United States using the proposed approach.
- There is probably a wide variation in the susceptibilities within and among the various risk groups and this is not well understood. The major obstacle for expanding risk assessments to other foods is the lack of data on the distribution of *V. vulnificus* levels in these foods at the time of consumption and potential matrix effects.
- The incidence of specific risk factors in the population consuming a seafood of interest and exposure associated with this seafood are the primary data needed for applying this model to other countries.
- Validation of the model in a given region or country would require epidemiological data on monthly *V. vulnificus* primary septicaemia rates.

6.1.3.2 Exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw

Introduction

V. parahaemolyticus is a leading cause of seafood-based illness in Japan and other Asian countries. With the globalization of Japanese cuisine and the increased practice of eating raw fish and shellfish, there is increased possibility of *V. parahaemolyticus* infection.

Outbreaks due to *V. parahaemolyticus* associated with fish and shellfish other than oysters have been reported in some countries including the United States, Thailand, China (Taiwan) and Spain. Several reports exist on the high prevalence of the organism in a variety of seafoods, especially in finfish, lobster and shrimp. Therefore, eating raw fish and shellfish has potential risks for *V. parahaemolyticus* infection and it is important to assess the exposure of consumers to *V. parahaemolyticus* in finfish.

Objectives

The objective of this exposure assessment is to model and quantify the exposure of consumers to *V. parahaemolyticus* from consumption of raw finfish.

Approach

The approach being taken is to modify the FDA-VPRA model for *V. parahaemolyticus* in oysters to accommodate data inputs on other seafood from Japan and other countries.

The model has four modules (Figs. 6.8 - 6.11) accommodating the entire continuum from harvest and post-harvest and ending with consumption in the home or in the food service sector.

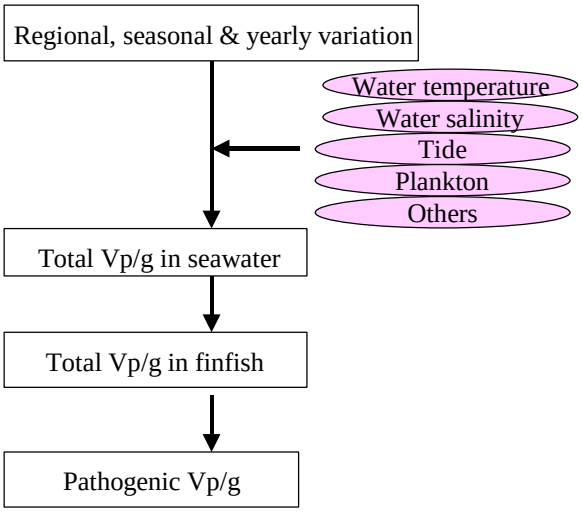

FIGURE 6.8: Schematic representation of the pre-harvest module for the exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw.

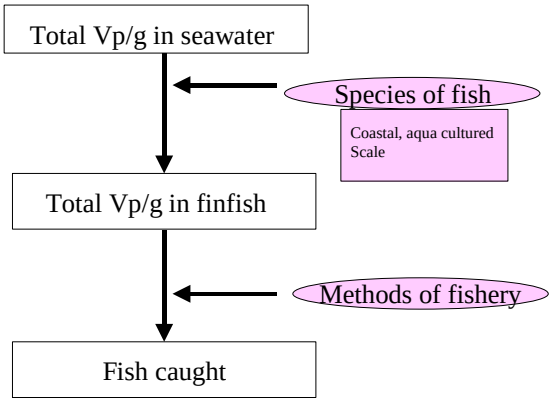

FIGURE 6.9: Schematic representation of the harvest module for the exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw.

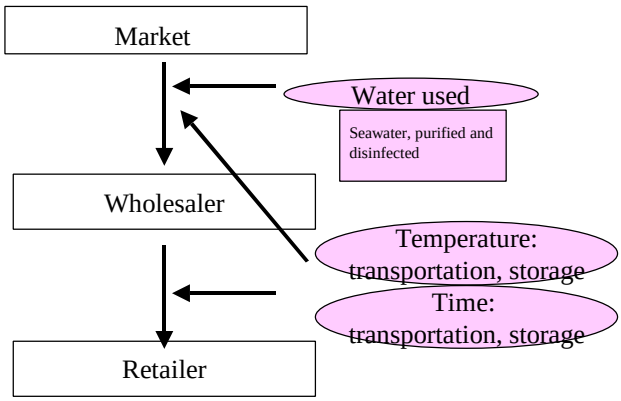

FIGURE 6.10: Schematic representation of the post-harvest module for the exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw.

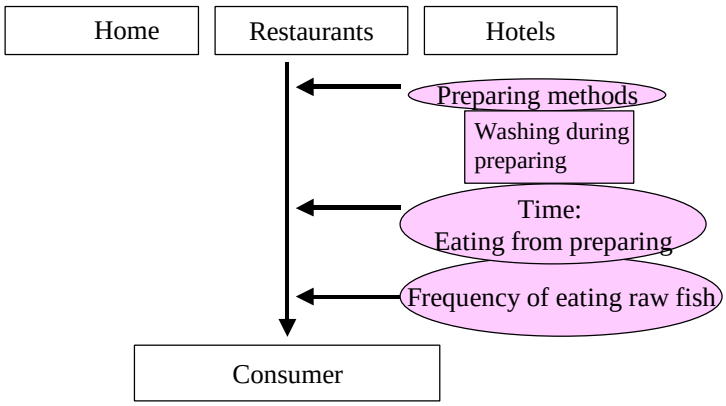

FIGURE 6.11: Schematic representation of the preparation and consumption module for the exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw.

Key findings

- *V. parahaemolyticus* density and prevalence in the seawater are influenced by seawater temperature, salinity of water, existence of plankton, tide and others.
- Many species of finfish could be contaminated with *V. parahaemolyticus* though prevalence and number of *V. parahaemolyticus* vary with species. Differences in prevalence and density seemed to be associated with the species and habitat (e.g. coastal or deep-sea).
- Coastal seawater used at landing and at market was shown to be highly contaminated with *V. parahaemolyticus* and this phase can be an important risk factor for contamination.
- This conceptual modelling approach would be appropriate for determining the potential effectiveness of mitigation strategies such as chlorinated water and thermal processing.
- The effect of time and temperature during transportation and storage may be less important than with raw oysters as *V. parahaemolyticus* was shown not to proliferate significantly until four hours at 25^{o}C on finfish samples.
- Washing the external surface of the fish and the visceral cavity with potable water reduced *V. parahaemolyticus* levels.

Gaps in the data

- Prevalence, number and proportion of pathogenic *V. parahaemolyticus* cells in various species of finfish
- Frequency of consumption of raw fish
- Transportation practices (time and temperature)

6.1.3.4 Exposure assessment of *Vibrio cholerae* in shrimps from developing countries for domestic and export consumption

Introduction

Seafood has been incriminated in cholera outbreaks. Shrimp is one of the most important seafood commodities in international trade and most shrimp come from developing countries. While high value shrimp is mostly exported by the developing countries to earn valuable foreign exchange, low value shrimp are consumed domestically. There have been outbreaks of cholera in many shrimp producing countries and such episodes have often adversely affected the international shrimp markets. In this context, it was felt that performing an exposure assessment for *V. cholerae* in shrimp intended for international trade as well as domestic markets was desirable since there are differences in the way shrimp are handled for these two markets.

Objectives

To perform an exposure assessment of *V. cholerae* in shrimp for domestic and export markets.

Approach

Shrimp may be contaminated with toxigenic *V. cholerae* during handling due to poor personal hygiene and washing with contaminated water. Occasionally *V. cholerae* O1 may be detected in brackish water aquaculture ponds where shrimp are grown. In the case of shrimp intended for export markets, generally specific hygienic practices are implemented to prevent contamination. Toxigenic *V. cholerae* is rarely isolated from shrimp imported from developing countries and there have only been one or two reported cases associated with shrimp products in developed countries (Infectious Agents Surveillance Report, National Institute of Infectious Disease, Japan, 1998) even though the total world shrimp production is about four million tons, of which 1,3 million are traded internationally with three quarters of this originating in developing countries (FAO. 1999). But in the domestic markets, contamination of seafood with toxigenic *V. cholerae* has been reported in a number of developing countries. Thus, the consultation suggested that the exposure assessments for shrimp intended for these two types of markets be performed separately as outlined below (Figures 6.12 and 6.13)

Shrimp intended for domestic markets in developing countries are often poorly iced, washed in landing centres where potable water is generally not available, sorted by hand and transported to local markets. Also, it may be prudent to consider the microbiological quality of the water from which shrimp are harvested. Contamination with toxigenic *V. cholerae* could occur through water used in processing establishments or through handling by asymptomatic carriers (Figure 6.12). In many developing countries, the domestic water supply is often not of potable quality and contamination with toxigenic *V. cholerae* could occur in kitchens of households and street vendors and also through raw-cook transfer. If such cooked contaminated shrimp are stored at ambient temperature, *V. cholerae* could multiply to infective doses.

Shrimp intended for export are generally iced immediately after harvest and transported in ice to well equipped processing establishments with hygienic controls, potable washing water, processors with clean gloves, clean handling tables, and HACCP plans etc.. In this setting, chances of contamination with toxigenic *V. cholerae* are very low. Shrimp frozen in such establishments are exported and thawed and cooked in the receiving country. Since *V. cholerae* are killed by cooking, the exposure in the importing country is likely to be negligible (Figure 6.13).

Key findings

Among *V. cholerae*, only serotype O1 and O139 are known to cause epidemic cholera. The major virulence factor of this organism is cholera toxin encoded by the *ctx* gene. Many environmental strains may be negative for this gene. *V. cholerae* is not known to colonise shrimp in its natural habitat. The primary source of *V. cholerae* is the faeces of persons infected with the organism. Asymptomatic carriers are also known to excrete the organism. *V. cholerae* reaches water through sewage and survives for long periods of time. However, the levels found in shrimp harvesting waters are generally low.

Contamination of shrimp with toxigenic *V. cholerae* could occur in the environment, through water used during processing or handling by asymptomatic carriers. *V. cholerae* is highly sensitive to gastric acid and therefore neutralization of gastric acid is found necessary to cause illness in human volunteers. *V. cholerae* is not known to multiply in raw shrimp, and it is sensitive to heat and eliminated during normal cooking of shrimp. If cooked shrimp is contaminated, the organism can multiply and reach infective doses if not adequately refrigerated.

Gaps in the data

- Data on the levels of toxigenic *V. cholerae* in natural waters and aquaculture environments.
- Levels of this organism or chlorine levels in the water supply in rural fish markets and in that used to process shrimp in developing countries.
- Data on the extent of cross-contamination and multiplication in cooked shrimp.
- Frequency and amount of shrimp consumption in both developing and developed countries.

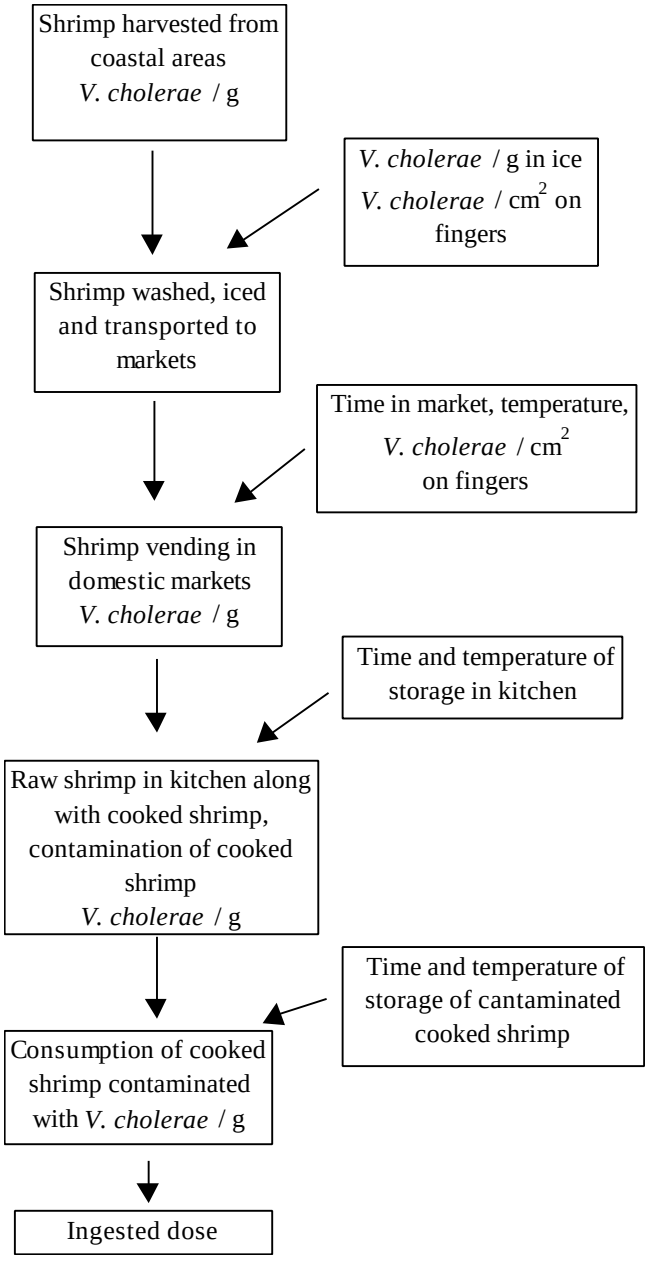

FIGURE 6.12: Model for exposure assessment of *V. cholerae* in shrimp for domestic consumption in developing countries.

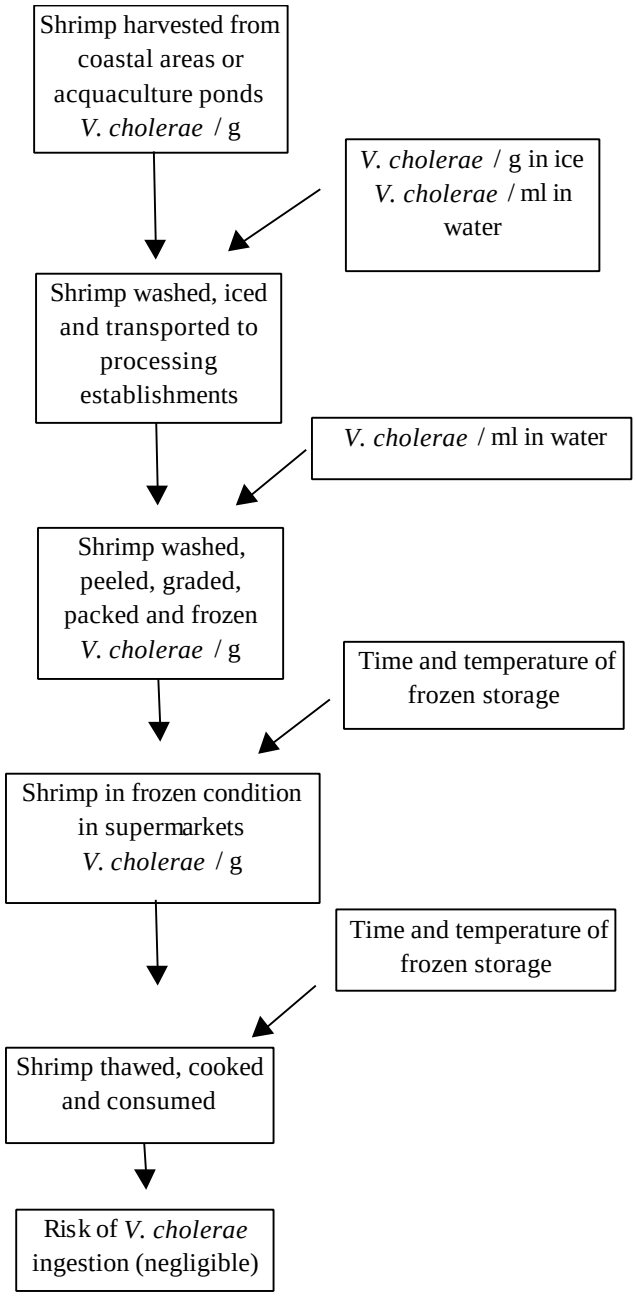

FIGURE 6.13. Model for exposure assessment for *V. cholerae* in shrimp for international markets.

6.2. SUMMARY OF THE DISCUSSIONS

6.2.1.Hazard identification of *Vibrio* spp. in seafood

The drafting group had not prepared a hazard identification document prior to the expert consultation. During the consultation the need to undertake this was identified and a document was subsequently prepared.

6.2.2 Hazard characterization of *Vibrio* spp. in seafood

The expert consultation noted that further information might become available which would modify the dose-response parameters for *V. parahaemolyticus*. This will be addressed when the data is forthcoming. With regard to *V. vulnificus* and *V. cholerae*, there was a need to identify the uncertainty in the dose-response relationships.

The consultation recommended that the information on the effect of age, sex and ethnic group, where available, should be presented in a consistent manner for the three *Vibrio* species. More recent consumption pattern data for these groups in the United States than that presented should be available and could therefore be included in the model.

Further information on dose-response relationships with respect to ingested food could be obtained by appropriate investigation of outbreaks. This has rarely been undertaken in a manner that contributed useful data. Ways should be considered to ensure that more useful information can be extracted from such events.

Consideration of the virulence factors of *V. parahaemolyticus* needs to include *tdh*-related haemolysin (*trh*). Cholera toxin needs to be clearly identified as the pathogenic entity for *V. cholerae* O1 and O139. A distinction between the biologically active proteins and the associated genetic factors needs to be made for each of these organisms.

6.2.3 Exposure assessment of *Vibrio* spp. in seafood

The consultation endorsed the approach used in selecting the pathogen-commodity combinations for detailed study but emphasized that the documents should clearly state the purpose for their selection.

6.2.3.1 Exposure assessment of *Vibrio parahaemolyticus* in raw oysters

A schematic representation of the process pathway used for the model should be included at the beginning of the risk assessment document before discussion of the model.

The expert consultation welcomed the approach to modelling presented in the exposure assessment document. It noted, however, that specific components of the model were developed based on data from the United States and that they may not therefore be suitable for application in different geographical areas. To facilitate a wider application of the model there should be instructions on the type and structure of data that would be needed in order to apply it in other geographical areas. The United States data would then be presented as an example data set and a basis for validation of the model under the United States conditions. Guidance could be given on the critical points in the model and the use of the United States data suggested where appropriate local data were not available, with appropriate riders on the constraints arising from this. It was noted that the United States data sets used in the exposure assessment were available in electronic format. As further data sets become available they should also be made available in this way.

The status of the availability of other data identified in the draft document, or sent in response to the FAO/WHO call for data was questioned. The identified data from Australia and Canada were available but data are still required from New Zealand on industry practices (e.g. time and temperature on boats). It was suggested that all the data received be placed in a table and identified as to whether it was inappropriate, of limited usefulness or good for inclusion in the risk assessment model. The expert drafting group agreed to undertake this for each pathogen-commodity combination individually.

The model used to predict the density of *V. parahaemolyticus* at harvest is an empirical model derived from data collected in the United States. There were a number of potential differences identified between the situation in the United States and other countries - cultivation and harvesting techniques, temperature and salinity of harvesting areas, consumption practices and frequency. Differences between water temperatures at surface and

depth could occur in some situations. Exposure to direct solar heating at low tide could alter the predicted effect of water temperature for intertidal fisheries. The proportion of pathogenic *V. parahaemolyticus* strains could conceivably vary in different geographical areas (it was noted that pathogenic strains may have been introduced into a United Kingdom estuary by discharge of waste from a plant processing imported seafood). The consultation recommended that ideally all of these factors be considered in the model.

At this stage the FDA-VPRA model incorporates only one potential explanatory variable, temperature. Salinity does not appear to be an influential variable in the United States so was not included in the final model. However, this may not be the case elsewhere and so this element may need to be retained. Other data elements should be investigated and incorporated into the model if they are found to be relevant. Local data should be used where possible to develop an empirical model to predict the density of *V. parahaemolyticus* at time of harvest. This will reduce the uncertainty that may arise from applying the FDA-VPRA model to environments outside that country.

The introductory section should be expanded to include greater consideration of the pathogenic strains. The consultation proposed the inclusion of two further paragraphs, one on the toxins and associated genes linked to pathogenicity and the other on the pandemic spread of a single clone and these have been provided by Prof. Nishibuchi from Japan. Pathogenic strains should be defined as those carrying the *tdh* and/or *trh* gene. The *tdh* and *trh* genes encode thermostable direct haemolysin (*tdh*) and *tdh*-related haemolysin (*trh*) respectively. Molecular epidemiologic studies have demonstrated that clinical strains usually carry the *tdh* gene, the *trh* gene, or both, whereas distribution of these genes in environmental strains is rare. The incidence (expressed in % of the total *V. parahaemolyticus* population) can be used in the proposed risk assessment model. The *trh*-positive strains are more frequently distributed in seafood than are *tdh*-positive strains (Personal Communication, Dr. Nishibuchi, Japan). Studies on the O3:K6 strains revealed that pandemic spread of infection by a new clone of *V. parahaemolyticus* is currently spreading around the world. The strains belonging to this clone have been isolated from clinical specimens in Asian countries and the United States and emergence of serovariants of the clone has been reported. Additional research and surveillance programmes are needed to identify and trace the spread of new epidemic strains as they emerge.

With regard to the proportion of pathogenic strains, it was emphasized that *tdh*-/*trh*+ isolates occur and it was noted that these strains occur with a greater frequency in Asia than in the United States. There were differences between studies in performance of the Kanagawa test - some gave good correspondence with molecular methods whereas others showed that a high proportion of false positives were obtained with the conventional test. It was thought that the molecular tests were preferable but there was some concern about recommending these for use in developing countries where laboratory facilities might be limited.

In order to improve the degree of confidence in the regressions of *V. parahaemolyticus* concentration on temperature and salinity, it will be necessary to include the appropriate model statistics. The drafting group indicated that the model's predictions were in good agreement with the measured concentrations from a retail survey. While it is possible that a number of compensating errors may have produced these results it provides good evidence that the model is valid. The model predictions, survey results and associated statistics should be included in the exposure assessment.

The exposure assessment for *V. parahaemolyticus* in raw finfish quoted models for the effect of temperature and salinity on the concentration of *V. parahaemolyticus* in raw oysters. The output from the present model should be compared to that of the Japanese models quoted in the finfish section. It should be determined whether the Japanese models have been subjected to validation. The latter models included a concentration factor relating the concentration of *V. parahaemolyticus* in seawater and oysters - caution was urged regarding this as such concentration factors were known to vary greatly for other bacteria with regard to both shellfish species and temperature.

Further explanation should be sought regarding the model assumptions relating to the growth of *V. parahaemolyticus* in oysters at various temperatures. It was confirmed that actual growth in oysters had only been experimentally observed at 26°C. These data were compared to a published study that had looked at the growth in broth at a number of temperatures. These temperatures included 26°C, at which temperature the growth rate in oysters was a quarter of that in broth. A triangular distribution of a factor between 3 and 5 had therefore been used for the relationship at other temperatures. Some concern was expressed that the growth rate in oysters could be even lower at lower temperatures because of greater competitive pressures - this would have the effect of making the present model conservative. It was also questioned as to whether the distribution of ambient air temperature during the day had been determined. In the current model, the temperature at noon had been used

rather than a distribution, which would be more complicated. The need for growth studies in oysters at other temperatures has been emphasized.

A difference in *V. parahaemolyticus* growth rate may exist between whole and shucked oysters. Growth may be slower in shucked oysters, partly because the pH drops after shucking, because they are usually kept on ice and because of the release of degradative enzymes. Shucked oysters had not been included in the FDA study because they were normally cooked in the United States - it was noted that this was not necessarily the case in other countries.

Uncertainty had been allowed for in the proportion of *V. parahaemolyticus* pathogenic strains included in the model. At present, an arbitrary symmetrical triangular distribution around the observed values had been incorporated to make some allowance for the uncertainty. In this regard, a question has been raised as to whether the proportion of pathogenic strains detected in retail samples from an area would be the same as that obtained from harvesting area samples. This might be due to the pathogenic strains in the former samples having grown above a detectable threshold. No conclusions were reached on this matter. It was queried as to whether an uncertainty had been incorporated into the model for *V. parahaemolyticus* die-off. This was not the case and a point estimate had been used.

There was discussion as to the role of antacids in *V. parahaemolyticus* infection. It was not known whether data on the use of antacids was available in many countries (in Australia, approximately 10% of the population are thought to be on prescribed acid lowering treatments). This may need to be taken into account, however, the exposure assessment currently considers the effect of susceptible populations with regard to serious sequelae after infection has taken place but not with respect to initiation of infection.

At this stage of the risk assessment, mitigation strategies were used in the model as examples of interventions and this should be clearly stated. Defining other mitigation strategies should be left to risk managers. A more comprehensive discussion on mitigation strategies, together with an appraisal of their effectiveness, may be undertaken at a later stage.

The water activity component in the equation in Section 4.1 of the background document could be replaced by a constant. It should be noted that the T_{min} and T_{max} values in the equation were theoretical and not actual, although they would be close to these.

6.2.3.2 Exposure assessment of *Vibrio vulnificus* in raw oyster

The consultation welcomed the proposal to extend the *V. parahaemolyticus* model to this organism and noted the clear identification of the work and data requirements. The present draft needs to be put into the format of the assessments on the other pathogen-commodity combinations. It was noted that *V. vulnificus* infections and deaths had been associated with other seafoods and reference to this needed to be included in the hazard identification (the information was provided to the expert drafting group by Dr. Yamamoto). Information from the draft European Commission report on *Vibrios* in seafood could also be incorporated when the report is publicly available. The progress of this assessment will depend on priorities identified by CCFH and the subsequent identification of appropriate resources. If a numerical model is produced (based initially on United States FDA data) then the United States Environmental Protection Agency (EPA) data would be provided by Dr Tamplin and this could be used to test the performance of the model using data collected from different sources.

6.2.3.3 Exposure assessment of *Vibrio parahaemolyticus* in finfish consumed raw

The consultation identified the need for the inclusion of further data on the prevalence of *V. parahaemolyticus* in seafood. Data from a study of seafood imported into Japan would be made available to the drafting group by Dr. Nishibuchi. These data can complement the *V. parahaemolyticus* data of the Japanese fish market. The imported seafood study included data on the proportion of *tdh*- and/or *trh*-positive strains. The data showed that for fresh imports, *V. parahaemolyticus* was most commonly found in tuna. The rate for tuna from that study and the data presented in the current draft document has implications for the progress of the model as there was currently an assumption that contamination rates would be highest for seafood from coastal and estuarine locations rather than deep sea.

In order to progress development of a model, it was proposed that a single appropriate finfish species, eaten raw, be identified. Expansion of such a model to include other species and the additional complications of undercooked seafood could then be undertaken later if the need and appropriate resources were identified.

In order to improve the degree of confidence in the regressions of *V. parahaemolyticus* concentration on temperature and salinity, it will be necessary to include the appropriate model statistics. Presentation of the data used in the development of the model would be useful. Data could be presented graphically as was done in the exposure assessment for *V. parahaemolyticus* in oysters. For the further development of models in this area it was necessary to compare the data already presented on contamination of fish surface and intestines in order to better identify the relative contribution of these sources to the contamination of the final product. Information was presented in the current draft on the effect of disinfecting process water: the data to be presented to the drafting group on the Japanese imported seafood included information on the chlorination of seafood in the producing countries. It should therefore be possible to include these as example mitigation steps as the model is developed further.

The variation of performance of *V. parahaemolyticus* isolation and enumeration methods in publications referenced in the assessment should be explicitly acknowledged even if the variation cannot be explicitly allowed for. This had been avoided in the FDA-VPRA as the study group had gathered all of its own data specifically to overcome this problem. The consultation agreed that standard methods should be adopted to facilitate the inclusion of data into risk assessment models.

Data on the frequency of consumption of raw fish was needed for the progression of this work. Further data would be sought but there is a need to gather additional reliable and detailed consumption data for the development of such risk assessments.

6.2.3.4 Exposure assessment of *Vibrio cholerae* in shrimp from developing countries for domestic and export consumption

The consultation emphasized that the predominant route of spread of *V. cholerae* O1 is via faecally contaminated water or by food contaminated by such water. The latter may apply to seafood. However, an increasing number of epidemiological studies have shown that food is an equally important route of transmission. In Latin America, undercooked seafood has been associated with a cholera outbreak. Although the latter may be significant in individual cases or outbreaks the risk should not be seen out of context with the other, more significant, routes of spread. The consultation noted the contention in the exposure assessment that the infectious dose of cholera is high and this certainly seems to be the case from the volunteer experiments that have been done. However, other evidence in the working paper suggests that the infectious dose may be as low as 10^2 organisms. It will be important for the development of a model to be clear as to the infective dose that applies in the case of food-associated infection. There is the possibility that seafood containing low concentrations of *V. cholerae* could contaminate other foods in which greater multiplication could occur.

The schematic process models for both domestic consumption in cholera-endemic countries (Figure 12) and for international trade (Figure 6.13) need to include additional steps in order to reflect differences in production, processing, transport and storage around the world. It was noted that while the approach proposed for modelling *V. cholerae* in exported shrimps was valid, there are differences in trade that are not captured. One example is that of cooked peeled shrimp which is a major import into countries such as Australia and the United Kingdom.

As with the consideration of *V. parahaemolyticus* in finfish, the information on seafood consumption should be expanded to include international data sets and this emphasizes the need to take steps to ensure that data appropriate to risk assessments is gathered and maintained in the future.

6.2.4 Conclusions and recommendations

- The format of the *Vibrio* risk assessment draft document should be revised to reflect the nature of the data and the models for the pathogen commodity combinations as indicated in Annex 4.
- The *V. parahaemolyticus* temperature model for oysters should be validated at temperatures other than 26°C.
- Relevant research should be undertaken to determine the proportion of pathogenic strains of *V. parahaemolyticus* in any region to be subjected to the model.
- The methodology used for the determination of pathogenicity of *V. parahaemolyticus* should be molecular identification of the presence of *tdh* and/or *trh*. The potential lack of availability of these techniques in developing countries needs to be recognized.

- With regards to the *V. parahaemolyticus* model, data from the five other identified countries should continue to be accessed by the expert drafting group and incorporated where appropriate.
- Local data should be used where possible to develop an empirical model to predict the density of *Vibrio* spp. in seafood at harvest.
- It is important to consider local conditions when applying a model based on the United States conditions, e.g. in New Zealand higher salinity in oyster leases may be a significant controlling factor.
- The drafting group should suggest ways in which the identified "Gaps in the data" should be addressed in order that any data obtained is appropriate to use in the model. The key data requirements for risk assessment of *Vibrio* spp. in seafood are outlined in Annex 5.
- The drafting group should identify the form of data needed for input to the model.
- In general terms data suppliers should provide information in an easily usable (spreadsheet) format.
- Risk of *V. cholerae* O1 infection through consumption of imported shrimp in developed countries is negligible.
- In developing countries, there is a risk of infection due to cross-contamination of shrimp through water or handling by asymptomatic carriers.
- More research is needed to understand the levels of toxigenic *V. cholerae* in waters and shrimp culture environments.
- Provision should be made for dialogue between risk assessors and risk managers to provide feedback on model creation and model documentation to better serve risk managers.

6.3. ISSUES TO BE BROUGHT TO THE ATTENTION OF FAO AND WHO

- Development of risk assessments requires information on food consumption patterns with the following particular details: demographics, preparation methods (where relevant), frequency of consumption and portion size.
- Applying *Vibrio* risk assessments to particular regions or areas will require the collection of data on the quantitative prevalence and numbers of the pathogenic vibrios at harvest and retail, with a knowledge of associated storage and transport times and temperatures.
    - Such data will only be of the greatest use in such assessments if there is harmonization and appropriate quality assurance of methodologies used to gather the data. It is important to facilitate co-operation between institutes in developing and developed countries.
- In order to enable international use of models based on the temperature and salinity of coastal and estuarine waters, there is a need to collect data on these variables, combined with estimations of *V. parahaemolyticus* and *V. vulnificus* concentrations in seawater.
- Further information is needed on the public health significance of pathogenic vibrios in all regions. This should be obtained by the reporting of human infections and monitoring of food contamination. Illness surveillance systems should be sufficiently thorough to provide useful data and collation of data and surveillance and monitoring reports should be undertaken at both national and regional levels.
- Data on sporadic cases as well as outbreaks would be important in assessing the full public health significance of these organisms.
- Further information is needed on dose-response relationships with respect to ingested food. This may be obtained by intensive investigation of outbreaks combined with analysis of incriminated foods.
- There is an uncertainty associated with the effect of different food matrices on the infective dose of different pathogenic vibrios. Research ought to be undertaken to determine the shift in dose-response curves when vibrios are ingested in different food matrices.

7. CONCLUSIONS OF THE EXPERT CONSULTATION

In addition to their specific conclusions on the risk assessments of *Campylobacter* in broiler chickens and *Vibrio* spp. in seafood (sections 5.2 and 6.2), the expert consultation concluded that considering the inherent difficulties and limitations, the draft risk assessments were comprehensive, of high quality and potentially useful for decision-making. Appreciation was expressed for the magnitude of work carried out by the expert drafting groups. The work represented a substantial advance in the application of scientific knowledge to improve the objective basis for managing microbiological hazards relating to *Campylobacter* in broiler chickens and *Vibrio* spp. in seafood.

The expert consultation agreed to the need to develop risk assessments for *Campylobacter* spp. in broiler chickens and *Vibrio* spp. in seafood and endorsed the approach taken by the drafting groups. The expert consultation furthermore recognized that the frameworks elaborated for risk assessments of *Campylobacter* and *Vibrio* spp. may provide the basis for the development of tools that could be customized and applied in different countries throughout the world. However it constitutes a big challenge, to ensure the flexibility necessary to account for regional and national differences. It was the opinion of the expert consultation that the risk assessments, when finalized, will help to guide and support risk management decisions.

The expert consultation concluded that validation of results was an essential part of any modelling exercise. However, there are currently only very limited data available that allow validation of important elements in the models proposed, which makes the validation of the final public health estimates difficult. There is an urgent need for epidemiological research to fill these data gaps, in particular research that can validate dose-response models.

The consultation concluded that the work in progress by the expert drafting groups should be seen as an exercise aimed at demonstrating:

- The applicability and usefulness of the available methodology.
- The need for the collection of data based on surveys and experimental studies specifically designed to provide information for microbiological risk assessment and the initial priorities in this regard.
- The opportunity for performing sensitivity or importance analysis to advise risk managers on where the risk management options can be implemented with the best use of resources.
- The potential of this work to answer specific risk management questions.

The consultation supported the recommendation of the previous expert consultation on risk assessments of *Salmonella* spp. in eggs and broiler chickens and *Listeria monocytogenes* in ready-to-eat foods[3] to develop guidelines for judging the quality of risk assessments.

The consultation furthermore concluded that frequent interaction between the risk assessors and risk managers in the further preparation of the risk assessments should be undertaken. Presentations by drafting group representatives at meetings of the CCFH would be a productive means to provide better understanding among risk managers of the potential uses and limitations of models and address the specific questions and concerns of the CCFH.

The consultation acknowledged the value of placing the presentation of the expert drafting groups on the FAO and WHO webpage.

8. RECOMMENDATIONS

The consultation recommended that FAO and WHO should:

- Develop guidelines for the collection of data to ensure that the quality of data collected for use in risk assessment is comparable between countries (sampling plans and methods of data analysis).

[3] The report of the joint FAO/WHO expert consultation on risk characterization of *Salmonella* spp. in broiler chickens and *Listeria monocytogenes* in ready-to-eat foods that took place in FAO headquarters Rome on 30 April - 4 May 2001 is available at http://www.fao.org/ES/ESN/pagerisk/reportSL.pdf and http://www.who.int/fsf/mbriskassess/report30April01.pdf.

- Encourage reporting of prevalence and concentration of specified hazards at different steps of the full exposure pathway in all regions of the world recognizing that currently most available control data are useless for the purpose of quantitative microbiological risk assessment.
- Develop a framework document for guiding the establishment of repositories for food safety data and surveillance data critical for effective risk assessment.
- Facilitate the development of surveillance systems with the view to data generation for quantitative microbiological risk assessment and explore ways to further evaluate the importance of [*]food saving systems that have recently been recognized as a useful means of obtaining quantitative data in the case of outbreaks.
- Promote the collection of consumption data appropriate for microbiological risk assessment at the national level.
- Facilitate dialogue between risk assessors and risk managers to provide feedback on model creation and model documentation to better serve risk managers.
- Ensure that requests by risk managers for the development of hazard characterizations or exposure assessments include a clear description of purpose and scope.
- Assist member countries in the preparation of project proposals on microbiological risk assessment activities for presentation to potential donors.
- Encourage member countries to allow time and funding for members of the drafting group to assist in the development of the risk assessment models, since this has been identified as a limitation concerning some drafting group experts.
- Define the terms used in the risk assessment documents in order to ensure consistency in the use of terminology and also to make them clear to all interested parties.
- Facilitate direct technical cooperation between developed and developing countries so that they achieve the technical capability required to carry out microbiological risk assessment. This support should take into consideration the local situation in order for the result to be sustainable.
- Consider, for future risk assessment activities, the involvement of scientific experts to serve as a standing advisory resource group to the risk assessment drafting group, for the duration of a specific work assignment. This would be a significant complementary activity to the more formal in-person experts meetings and would certainly take greater advantage of the intellectual resources of the expert community through their knowledge of and access to relevant data, references and network of colleagues.
- Consider processes to elicit expert judgement in a structured manner, using recognized procedures and protocols to draw out knowledge and opinion and to minimize bias. This will be beneficial in limiting model uncertainty when data are lacking, or when the available data are conflicting, primarily for those parameters which are important determinants for the characterization of risk.

In addition the expert consultation recommended that:

- Risk assessment of microbiological hazards in foods be included in the curricula in relevant university courses.

[*] Food saving systems: A system whereby all large foodservice establishments are advised to keep frozen portions of prepared foods for a specified time period for subsequent testing in the case of illness being associated with the food.

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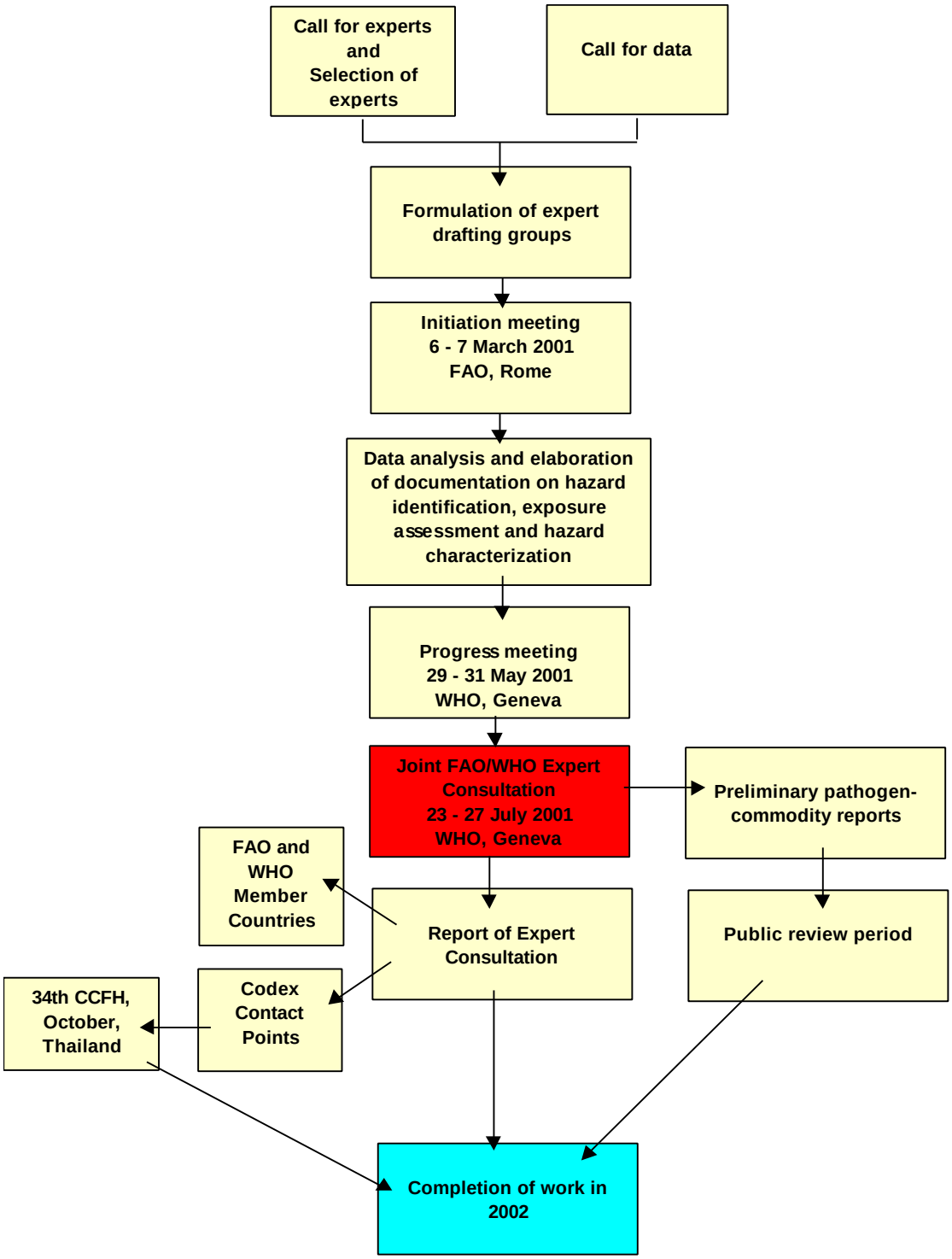
ANNEX 2: JOINT FAO/WHO MICROBIOLOGICAL RISK ASSESSMENT ACTIVITIES

Process of Operation for the Joint FAO/WHO Activities on Risk Assessment of Microbiological Hazards in Foods.



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ANNEX 3: LIST OF WORKING DOCUMENTS

Six working papers were prepared for, and presented during the expert consultation. These served as the basis for the discussions, which led to the development of the report and the recommendations. These documents were prepared for FAO and WHO by a number of expert drafting groups. The full text of these documents will be made available on the FAO and WHO webpages; http://www.fao.org/ES/ESN/pagerisk/riskpage.htm and http://www.who.int/fsf/.

Paper no.	Title	Authors
MRA 01/03	Hazard characterization of Vibrio spp. in seafood	Mark Walderhaug, Food and Drug Administration, United States John Bowers, Food and Drug Administration, United States
MRA 01/04	Exposure assessment of Vibrio spp. in seafood	Angelo Depaola, Food and Drug Administration, United States I. Karunasagar, University of Agriculture Sciences, India Ken Osaka, National Institute of Infectious Diseases, Japan John Sumner, M&S Food Consultants Pty. Ltd., Australia Mark Walderhaug, Food and Drug Administration, United States
MRA 01/05	Hazard identification, hazard characterization and exposure assessment of Campylobacter spp. in broiler chickens	Steve Anderson, Food Safety and Inspection Service, United States, Bjarke Bak Christensen, Veterinary and Food Administration, Denmark Aamir Fazil, Microbial Food Safety Risk Assessment, Health Canada Emma Hartnett, Veterinary Laboratories Agency, United Kingdom Anna Lammerding, Health Canada Maarten Nauta, Rijksinstituut voor Volkgesondheid en Milieu (RIVM), The Netherlands Greg Paoli, Decisionalysis Risk Consultants, Canada Hanne Rosenquist, Veterinary and Food Administration, Denmark

ANNEX 4: SUGGESTED FORMAT OF THE *VIBRIO* RISK ASSESSMENT DOCUMENT

Document 1 Hazard Identification - *Vibrio* spp in Seafood

Document 2 *Vibrio parahaemolyticus* in oysters

- Section 1 Exposure Assessment
- Section 2 Hazard Characterization
- Section 3 Risk Characterization

Document 3 *Vibrio vulnificus* in oysters

- Section 1 Exposure Assessment
- Section 2 Hazard Characterization

Document 4 *Vibrio parahaemolyticus* in finfish

- Section 1 Exposure Assessment
- Section 2 Hazard Characterization

ANNEX 4: KEY DATA REQUIREMENTS FOR RISK ASSESSMENTS ON, AND DATA RELEVANT TO RISK ASSESSMENT OF *VIBRIO* SPP. IN SEAFOOD

The key data requirements identified by the expert drafting group are as follows:

1.Vibrio parahaemolyticus

a. Number of total and pathogenic *V. parahaemolyticus* (*tdh* and /or *trh* positive) in oysters or other seafoods at harvest, that may be consumed raw or used in ready-to-eat seafood products.

b. Temperature and salinity of the harvest waters and where possible the numbers of *V. parahaemolyticus* in seawater.

c. Time, temperature and other relevant information that may affect survival and growth of V. *parahaemolyticus* during storage, handling and processing practices.

d. Survival and growth rates in food matrices applicable to typical industry storage, handling and processing conditions.

e. Numbers of V. *parahaemolyticus* at the point of consumption in above commodities.

f. Amount of consumption for each of the above commodities.

g. Number of illnesses reported for each of the above commodities.

h. Dose-response data of various strains in animal models or humans, and specifically quantitative data from outbreak investigations.

i. Data on effect of mitigation strategies

2. ***Vibrio vulnificus***

a. Total numbers of *V. vulnificus* in oysters and other seafood at harvest.

b. Temperature and salinity of the harvest waters, and where possible the numbers of *V. vulnificus* in seawater.

c. Time, temperature and other relevant information that may affect survival and growth of V. *vulnificus* during storage, handling and processing practices.

d. Survival and growth rates in food matrices applicable to typical industry storage, handling and processing conditions.

e. Numbers of *V. vulnificus* at the point of consumption in raw oysters and other seafood.

f. Amount of consumption of raw oysters and other seafood.

g. Portion of the population with chronic illnesses (i.e. liver disease, immunocompromised, diabetes).

h. Number of illnesses reported for raw oysters and other seafood.

i. Dose-response data for various strains in animal models and specifically quantitative data from outbreak investigations.

j. Data on the effect of mitigation strategies

3. ***Vibrio cholerae***

a. Number of toxigenic *V. cholerae* in shrimp and other seafood (wild or aquaculture) at harvest

b. Temperature and salinity of the harvest waters and where possible the numbers of *V. cholerae* in seawater.

c. Time, temperature and other relevant information that may affect survival and growth of *V. cholerae* on storage, handling and processing practices.

d. Survival and growth rates in food matrices applicable to typical industry storage, handling and processing conditions.

e. Numbers of *V. cholerae* at the point of consumption in cooked and raw shrimp and other seafood.

f. Amount of consumption for each of the above commodities.

g. Number of illnesses reported for each of the above commodities.

h. Dose-response data for various strains in animal models or humans, and specifically quantitative data from outbreak investigations.

i. Data on the effect of mitigation strategies.