MICROBIAL RISK ASSESSMENT GUIDELINE

PATHOGENIC MICROORGANISMS WITH FOCUS ON FOOD AND WATER

Prepared by the Interagency Microbiological Risk Assessment Guideline Workgroup

DRAFT

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PREFACE

This Microbial Risk Assessment (MRA) Guideline is written to be a resource for microbial risk assessors at the U.S. Department of Agriculture's Food Safety and Inspection Service and the U.S. Environmental Protection Agency and their agents, contractors, and the general risk assessment community with which these agencies interact. Other Federal agencies expressed interest in the development of this guideline and provided experts to participate in this interagency effort (see list of participants).

In recognition of the needs and mandates of the participating Agencies and the various statutory authorities that may apply to microbial risk assessment (MRA), this Guideline emphasizes the need for a flexible template for conducting microbial risk assessment. It provides general, broad fundamental risk assessment principles specifically for microbial risks, but is not intended to be prescriptive nor is it intended to supplant the internal practices or policies of any Federal agency. Users have the flexibility to adapt pertinent sections to relevant statutory authorities and purposes if needed. On the individual level, it is written for persons with some knowledge of microbiology and basic understanding of risk assessment principles but some basics may be presented at the introduction to a topic. The intention is that this Guideline can be periodically updated, particularly as more information becomes available.

This Guideline primarily focuses on infectious diseases associated with the gastrointestinal tract and fecal or oral transmission of the causal agents mainly in food and water, but clearly has application to other scenarios.

Many human pathogens found in food, water, and the environment cause acute diseases that have short incubation periods, symptoms typically lasting several days to a week, and usually non-lethal effects with complete recovery from the illness. However, some pathogens associated with the gastrointestinal tract may cause more serious diseases or sequelae such as reactive arthritis, cancer, Guillain-Barré syndrome, and juvenile onset diabetes which may have long-term implications. Further, there are indigenous water-based pathogens, such as *Legionella* spp. and *Mycobacterium* spp., that grow in biofilm environments and their inhalation via aerosols may cause pneumonia, which can be fatal. Applying risk assessment approaches associated with MRA procedures discussed in this guideline should help risk assessors characterize the common exposure sources, causative agents, associated symptoms, contributing immunity factors, and other common threads contributing to chronic illness.

The Guideline does not specifically include biological warfare agents, airborne microbial hazards, or agriculturally or industrially important microorganisms, oligonucleotides, prions, preformed microbial toxins, and other submicrobial entities. These agents have many unknowns associated with their sources, modes of "infection" and disease, transmissibility, and survivability. Nonetheless, information in this guideline may provide information to risk assessors addressing these issues.

1. INTRODUCTION

Microbial risk assessments (MRA) are a valuable tool for Federal Agencies tasked with understanding, reducing, and preventing risks presented by hazardous microorganisms, whether natural or anthropogenic, intentional or unintended. Increasing globalization has compounded these risks, with the broadening and often rapid distribution of illnesses. Clear and credible risk assessment methods are proving ever more necessary for agencies to address both current and future risks associated with contamination of air, water, soil and food by bacteria, fungi, protozoa, viruses, and their toxins.

This guideline is intended to layout an overarching approach to conducting microbial risk assessment and to introduce the users to tools and methods available to them. Additionally, it is expected to promote consistency and improve transparency in the way -microbial risk assessments are conducted. Nonetheless this Guideline is not intended to replace existing guidelines that are in use by Agencies. The decision to apply methods and approaches in this guideline, either totally or in part, is left to the discretion of the individual department or agency. This document is offered to provide information that may be useful for microbial risk assessors.

1.1 What is Human Health Risk Assessment?

Risk assessment is widely recognized as a systematic way to prepare, organize, and analyze information to help make regulatory decisions, establish programs, and prioritize research and development efforts. In *Risk Assessment in the Federal Government; Managing the Process*, published by the National Research Council of the National Academies of Science (NRC, 1983; hereafter referred to as the "NRC 1983 report"), risk assessment is defined as "the qualitative or quantitative characterization of the potential health effects of particular substances on individuals or populations," structured to include a hazard identification (HI), dose-response assessment, exposure assessment, and risk characterization. Similarly, the *Codex Alimentarius* Commission, established by the United Nations Food and Agriculture Organization (FAO) and World Health Organization (WHO) and recognized by the World Trade Organization as the relevant organization for international food safety standards and guidelines, defined risk assessment as "a scientifically based process consisting of the following steps: (i) hazard identification, (ii) hazard characterization, (iii) exposure assessment, and (iv) risk characterization" in *Principles and Guidelines for the Conduct of Microbiological Risk Assessment* (Codex, 1999; hereafter referred to as the Codex framework).

The risk assessment process is used to facilitate the application of science to policy decisions. Risk assessment at the Federal level informs the risk management decision making process and risk communication through organized scientific analyses of data related to a specified hazard. Risk assessment can also evaluate potential or proposed risk management strategies' impact on public health. Essentially, a MRA is the

formal, scientifically based process to estimate the likelihood (probability) of exposure to a microbial hazard and the resulting public health (and/or environmental) impact from this exposure. Risk assessment not only includes the likelihood of exposure and the impact of that exposure, but also includes steps for planning, scoping, and hazard identification and characterization (see Section 1.4). This Guideline will only focus on MRA conducted for public health purposes.

Risk assessment, risk management, and risk communication are the three components of a risk analysis. Figure 1.1 is a representation of how these terms are related. The three activities are shown as partially overlapping because they depend upon each other and draw from each other. This Guideline addresses risk management and risk communication from the perspective of what a risk assessor should know about those disciplines and provides transparency to decision makers and stakeholders regarding how a MRA is conducted. Therefore, the risk communication and risk management chapters in this Guideline provide only a small portion of what those disciplines contribute to risk analysis as a whole.

Figure 1.1 Risk Assessment, Management, and Communication in Risk Analysis

1.2 What Led to the Decision to Create this MRA Guideline?

Government Agencies have conducted formal risk assessments for chemicals in food, water, and the natural environment for decades. These assessments had their origins in support of or in response to the establishment of a number of laws and regulations that required the federal government to control chemical contaminants in food and environmental media. The NRC 1983 report helped unify the risk assessment processes for chemicals in foods and the environment and provided a framework that federal agencies, their clients, and the risk assessment community in general could apply in conducting risk assessments. Since then, the NRC 1983 report has been used by regulatory agencies as guidance in conducting risk assessments.

The need for a common chemical risk assessment approach was established in the 1980s, before there was a similar requirement for a MRA approach. At that time, MRA limitations included lack of data, tools, and methods, such as limited dose-response models, poor quantification of microbial occurrence, limited analytical methods (sensitivity, specificity, precision, and accuracy), and poor understanding of human immunological responses. Since the 1990s, the use of MRAs has gained significant credibility in the federal regulatory community, as new information on the identification of infectious microbial pathogens, their occurrence, potential for human exposure, dose-response, and attributable health effects became increasingly available. A number of credible mathematical models, protocols, and other tools have become available that allow MRAs to be conducted even with substantial variability, uncertainty, and lack of specific data – issues that previously would have impeded preparation of quantitative risk assessments. Indeed methods are now available to specifically capture and report variability and uncertainty associated with data use in MRA. During the 1990s, it also became apparent that the NRC 1983 report had some shortcomings for conducting MRAs since chemicals are different from microorganisms in a number of ways (see Section 1.5). The NRC 1983 report provides a very useful foundation for chemical and microbial risk assessment, but does not provide the level of detail that is required to conduct a quantitative microbial risk assessment. Chemical risk assessors developed more detailed guidelines to expand on the NRC 1983 report. The detailed chemical risk assessment protocols clearly would not work for microorganisms, so it became necessary for MRA to consider microbial aspects that are not considered in the NRC 1983 report. While Agencies conducting MRAs had for a time continued to generally rely on the NRC 1983 report, they have individually made adjustments to adapt it for MRA. For example, the FAO of the United Nations and the WHO use the international Codex framework, which follows the same overall structure as the NRC 1983 report, to conduct a MRA of *Listeria monocytogenes* in ready-to-eat foods (FAO/WHO, 2004) and the Environmental Protection Agency (EPA) used the framework in the NRC 1983 report to evaluate the public health impact of drinking water regulations for *Cryptosporidium* oocysts (EPA, 2006a).

The workgroup that created this Guideline did so with the intent of leveraging limited resources, improving efficiencies, promoting joint interaction, exploiting their individual experiences, and improving consistency in the way the microbial risk assessments are done and thereby fostering greater transparency. This Guideline does not contain every tool that can be used in MRA, but it does include the most widely used tools. This Guideline also is considerably longer and more detailed than the MRA frameworks that precede it (Codex, 1999; ILSI, 2000; Codex, 2007a, 2007b). EPA Office of Water's Protocol for Microbial Risk Assessment and the EPA Science Advisory Board's review of that document were also considered during the development of this document (EPA, 2009a).

1.3 What are the Benefits of Developing a Common MRA Guideline?

Agencies that need to be concerned about pathogens often have similar requirements to protect the health of potentially exposed people. For example, a number of pathogens of concern originate in the gastrointestinal tract of humans and animals and can potentially contaminate food, surface water, or drinking water. The agencies that regulate food and environmental contaminants recognize that the ultimate sources of pathogens are the same for different media. Because the health effects and dose-response relationships are similar among different matrices for many of the pathogens, it is useful to have common principles and approaches to assess risks across media and exposure settings.

This Guideline facilitates systematic and transparent consideration of all relevant factors that impact the risk assessment and facilitates reproducible risk evaluation. Agencies assessing a similar medium or pathogen can more readily compare and contrast the details and assumptions of their assessment to another Agencies' assessment. This is not to say that each risk assessment will be completely cross-comparable with other risk assessments, since there are a number of specific data sources and Agency requirements that require different inputs and applications. The difference in requirements is why this Guideline is designed to be modular and able to provide flexibility for each Agency's specific requirements. On an international scale, there is also a need to have common approaches to MRA to effectively satisfy international trade agreements and public health protection for importation of food and beverage products and assess international risks from emerging pathogens around the world.

This Guideline is the result of the collaborations of microbial risk assessors from a number of Federal Agencies (USDA/FSIS, USDA/APHIS, EPA/OW, EPA/ORD, EPA/OPP, EPA/OPPT, EPA/OSA, FDA/CDER, CDC/NIOSH, CDC/NCEZID, NASA, USACHPPM) who have dedicated their time and expertise to prepare a general guideline that all microbial risk assessors in the Government, their contractors, and the risk assessment community in general can apply for conducting MRAs of infectious agents or disease-causing microorganisms. The workgroup that developed this Guideline agreed that this document must be general enough so that any Agency, that chooses to do so, can use it effectively. By being based on general and modular guidelines, each specific risk assessment component can then be supplemented (or modified) to suit the particular legislative, legal, or exposure factor needs of an agency.

The Office of Management and Budget's (OMB) *Good Guidance Practices* has been followed as this Guideline was developed (OMB, 2007a).

1.4 When Can I Apply this MRA Guideline?

This guideline is applicable to a wide array of scenarios but focuses on microorganisms that are capable of causing infection and disease in the human population. Specifically it is applicable for assessing risk associated with ingestion of foodborne pathogens (in raw and processed foods), and water-based or waterborne

pathogens (in drinking water, recreational water, wastewater, etc.). I could also be applied to assessing risk of human pathogens in soil and solid wastes or transported through the air, and human exposure to biological warfare agents. The Guideline may also apply for other common forms of exposure including inhalation and dermal exposure pathways. At present, oligonucleotides, prions, preformed microbial toxins, and other submicrobial entities are not covered under the Guideline owing to a wide array of unknowns associated with those agents.

This Guideline should help you account for differences between the general population and different life stages and sensitive populations. It is flexible enough that risks to individuals, subpopulations, and the general population can be dealt with using either available static or dynamic susceptibility models. The approaches and tools provided by this Guideline should be appropriate for typical human health related concerns. The Guideline does not include criteria to specifically identify sensitive groups or life stages, because those groupings are specific to each risk assessment, based on the Agencies' unique health protection concern and public health goal.

The intent of the Guideline is to provide the necessary elements for successfully conducting a full risk assessment, quantitative or qualitative, including:

a) Planning and scoping including problem formulation,

b) Hazard identification (HI),

c) Hazard characterization (HC),

d) Dose-response assessment,

e) Exposure assessment, and

f) Risk characterization.

This Guideline incorporates these elements and follows the Codex definition for the risk assessment process (see Section 1.1), except that the qualitative aspects of hazard characterization are presented with hazard identification, instead of with dose-response (Codex, 1999). It is intended that the Guideline will also assist risk assessors to meet the needs and expectations of risk managers, and provide appropriate and effective risk communication with managers, stakeholders, and the public.

1.5 What are Some Fundamental Differences between Microbes and Chemicals?

The differences between chemical hazards and human susceptibility factors considered in chemical risk assessments and microbial-host factors in MRAs can be few or many depending on the microbial antagonist, host resistance factors, and exposure considerations. While these two types of risk assessments are conceptually similar, there are typically enough differences between chemicals and microorganisms that having an

approach specifically detailing unique microbial considerations is very useful. Even though the uniqueness of each chemical is considered individually in chemical risk assessment, some significant differences in MRAs to be considered include:

a) **Microbial growth and death** – Pathogens increase and decrease in number in the environment and host. Different species, and even different strains within a pathogenic species, grow and die in unique patterns. In contrast, although chemicals can bioaccumulate, bioconcentrate and undergo transformations, they do not multiply in the environment or hosts. Both chemicals and pathogens can decrease due to environmental factors; chemicals can be transformed or degraded, and pathogens can die or become unculturable but may remain infectious. Some decrease in chemicals may be reversible (e.g., sequestered toxicants may remobilize). However, microbial toxins can remain after the organism dies and some enterotoxins are heat stable and resistant to degradation.

b) **Host immunity and susceptibility** – Although body weight, age, and metabolic capacity differences are considered in chemical risk assessment, genetic and acquired differences in susceptibility are not considered in chemical risk assessment in the same manner as in MRA. Whereas uncertainty factors or assessment using data from known sensitive populations may be used to consider these host differences in chemical risk assessment, in MRA immune status can be included in a dynamic model. The immune system may be considered in chemical risk assessment if a chemical causes a hypersensitivity reaction. Infection and resulting illness due to pathogens is, in some cases, highly dependent on the immune status of the individual, which can fluctuate based on time since last exposure, presence of concurrent infections, and a number of other factors (e.g., life stage, nutrition, genetics).

c) **Diversity of health endpoints** – The same dose of a pathogen may result in a broad range of health outcomes or endpoints depending on the characteristics of the host and the exposure scenario. Endpoints for the same dose could include: no effect, asymptomatic infection, mild or severe symptoms, or mortality for the most susceptible. The percentage of potential hosts in each category fluctuates over time.

d) **Genetic diversity and evolution of microbial strains** – Microorganisms are genetically diverse and allelic ratios in a population can change significantly within a few generations. In addition, microbial genomes can evolve quickly (within days or weeks) through mutation or vertical gene transfer (within a species) or horizontal gene transfer (between different species, families, and higher taxonomic differences). Strains of the same species (e.g., *Cryptosporidium parvum, E. coli*) can have multiple genotypes, potentially with different virulence for human hosts. Some pathogens (e.g., *Helicobacter pylori*, many viruses) behave like quasi-species, which are fluctuating populations of genetically distinct variants that co-exist within a single host

(Boerlijst et al., 1996; Covacci and Rappuoli, 1998). Microbes represent a "moving target," because the distribution of strains and virulence factors can fluctuate rapidly in a given medium. Although there is great diversity among chemicals, particularly considering mixtures, the mechanism of change is not evolution.

e) **Potential for secondary transmission** – Microbial infections can be transmitted between individuals, and even to some animals. With the exception of the mother-fetus relationship, chemicals in tissues of exposed individuals are not known to transmit to other individuals. Chemicals that are on an exposed individual's clothing or skin can be transferred to household and other inanimate objects (fomites), but that transfer results in dilution of the chemical, as opposed to pathogen secondary transmission, which can amplify the pathogen. Some microbes can remain viable for days, weeks, or months on surfaces, which increases the potential for transmission. For some pathogens humans can become asymptomatic chronic carriers and can infect others and contaminate food and water sources with displaying symptoms themselves for prolonged periods.

f) **Heterogeneous spatial and temporal distribution in the environment** – Pathogens are typically heterogeneously distributed in environmental matrices. Pathogen growth may lead to clustered distributions, and pathogens may physically clump together or may be embedded in or attached to organic and inorganic particulate debris, making concentration determinations difficult. Although concentration in pipe scale and biofilms is also a problem for chemical contaminants, some pathogens can grow and/or be protected in these environments. Also, many types of pathogens occur only episodically and typically can be found only during short-lived disease outbreaks (i.e., epidemics) in a community. Seasonal and event-related (wet weather) spikes are common.

g) **Single exposure health outcome** – Chemical risk assessments are conducted for acute exposures that cause immediate health outcomes and for chronic exposures with long-term health effects. For chronic exposure to chemicals, the risk may be from daily exposure over a 70-year lifespan, whereas for pathogens the risk may be from a single exposure with health effects noticeable within days or weeks. Some pathogens may have sequelae, which are health outcomes that appear much later than the original symptoms. Some sequelae are chronic (long lasting). Unlike the long-term exposures often considered for chemicals, longer-term risks due to pathogen exposure are not typically considered for MRA.

h) **Wide range of microbial response to interventions** – Many risk assessments address risks to human health associated with media that have been subjected to some sort of treatment (such as wastewater treatment for water or strict processing of foods). Microorganisms respond with wide variability to

environmental and treatment factors. For example, response to drinking water treatment needs to be taken into account when comparing microbial levels in ambient water and treated drinking water. However, in the Clean Water Act 304(a) ambient water quality criteria, EPA makes a policy assumption that drinking water treatment has no effect on chemical concentration when they determine what levels to set for ambient water (EPA, 2000c).[1]

i) **Detection method sensitivity** – Microbial detection methods may not be sensitive enough to detect pathogens at a level of regulatory concern. This is not necessarily the case for all pathogens or all media, but does apply to some combinations of organisms and media. Theoretically, a single pathogenic organism can cause infection and lead to illness. Analytical methods for detecting low levels of pathogens (e.g., one organism in 1000 liters of water) are not sufficiently developed to be reliable. In short, the human body is a more sensitive detector of pathogens than many laboratory methods. In addition, the active or viable-but-not-culturable (VBNC) state is not detectable by traditional culture-based laboratory methods (see Section 3.8)

j) **Population, community, and ecosystem-level dynamics** – Microbial pathogens have complex interactions with other members of their species, other species, and the abiotic environment. For example, pathogens compete with non-pathogens for resources, and many non-viral human pathogens have animal hosts that can greatly complicate the ecological dynamics of pathogen occurrence.

k) **Routes of exposure** – Many routes of exposure are similar for chemicals and microorganisms; however, there are some potentially important differences. Dermal exposure may be important for some chemicals but not necessarily with microbial exposure since unbroken skin is a natural barrier for entry. However, dermal contamination with pathogens can lead to oral exposure via transfer to consumed food or water. Other aspects may include consideration of direct person-to-person or person-to-environment-to-person routes.

These factors are significant considerations in MRA, but the approach for dealing with each one may be unique to a particular risk assessment scenario.

1.6 What is the Relationship of Infectious Disease to Human Health as Applied in a MRA?

Understanding the relationships and interactions between a microbial pathogen, its host, and the exposure to the pathogen in the environment is key to determining the potential health impact a pathogen will have on an individual or population. The epidemiological triangle (disease triad) illustrates the inter-relationship between the host, pathogen, and environment components (Figure 1.2). For each of the components, there are several key factors that may be considered. In many cases, a comprehensive

[1] Except for disinfection byproducts

quantitative treatment of all the factors to conduct a MRA is not possible because of data limitations. However, the factors can be at least considered qualitatively. The various points of the disease triad will be discussed throughout this Guideline.

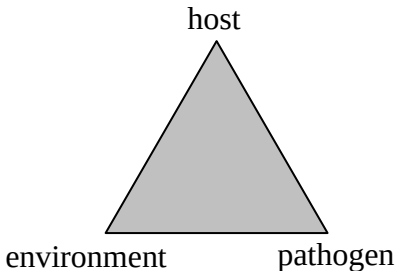

Figure 1.2 The Epidemiological Triangle

1.7 What are the Benefits of Iterative MRA?

There is general recognition by risk assessors that an iterative approach is necessary when conducting a MRA, so that there is opportunity to modify the risk assessment based upon changes in data availability and evolving agency policies (Presidential/Congressional Committee [P/CC], 1997). Often, the lack of data, new data or interpretations, or uncertainty or variability in information will require you to revisit the original charge or premise for conducting a risk assessment. As revisions are conducted, new information can be incorporated, especially in areas found to be the most important to assessing risk. This ensures that the risk manager is provided with the most accurate interpretation of risks, so that appropriate management decisions can be made. Risk communication strategies can also be developed in parallel with risk assessment iterations. You should be prepared to be involved in the risk communication process as needed (see Chapter 8).

1.8 How Does This Guideline Fit in with My Agency's Current MRA Approaches and Practices?

This Guideline is not intended to supplant your Agency's prerogative in establishing the approach that you may follow for preparing Agency-wide or even Office-wide MRA procedures. The goal of this MRA Guideline is to provide an overarching framework and an approach to conducting MRAs that are flexible and broadly defined, so that any Agency can utilize them in preparing more tailored approaches to MRA. This Guideline is not intended to preferentially take the place of your Agency's own risk assessment approach, protocol, or guideline, but should help promote consistency in MRA among agencies. Each Agency has its own unique regulatory requirements, legal requirements, policies, and conventions that must be considered in their risk assessments. Ideally, this interagency Guideline is intended to provide the general framework that can be incorporated into the more specific MRA guidance that your Agency prepares. However, it is up to each Agency to make the determination of how or whether they utilize this guideline, consistent with international and national scientific guidance (e.g., Codex Alimentarius and the United States' National Academies).

1.9 How is this MRA Guidance Related to Other MRA Frameworks/Guidelines that are Currently Available?

There are two major sets of frameworks that influenced the development of this Guideline. The first set of frameworks includes the NRC 1983 report (NRC, 1983) and the NRC *Science and Decisions: Advancing Risk Assessment* (NRC, 2009), which are related because the NRC 2009 framework is an enhancement of the NRC 1983 report. The NRC frameworks are geared to chemical risk assessment, but have applicability to MRA. They are also broadly applicable for many different exposure media. The second set of frameworks is the *Codex Alimentarius* Commission (Codex), *Principles and Guidelines for the Conduct of Microbiological Risk Assessment* (Codex, 1999) and Codex *Proposed Draft Principles and Guidelines for the Conduct of Microbial Risk Management* (MRM) (Codex, 2007a). The Codex framework is specific to the food media, but is tailored to microbial hazards. This Guideline is broadly applicable to many media, but is tailored to microbial hazards to humans, so both sets of frameworks were important for the development of this Guideline. These two sets of frameworks were not the only sources consulted for development of this Guideline.

U.S. government agencies (e.g., EPA, USDA, FDA, DHS, and DOD), as well as government organizations from other countries (e.g., Canada, European Union, New Zealand) and international Agencies (e.g., WHO, FAO/CODEX, and the Organization for Economic Cooperation and Development [OECD] – for example, see FAO/WHO, 2006), have prepared various levels of guidance to support MRA applications. An EPA-sponsored study, *Foundations and Frameworks for Human Microbial Risk Assessment* (Parkin, 2008), presented an extensive search and evaluation of frameworks available for use in conducting MRA. The study determined that there were four general categories of frameworks that have been applied in MRA:

a) The 1983 NRC report

b) A modified NRC 1983 approach without an explicit problem formulation (or planning and scoping) step

c) A modified NRC with a problem formulation (or planning and scoping) step

d) The International Life Science Institute (ILSI) approach (in association with the EPA's Office of Water) developed for water-based media (ILSI, 2000)

While most microbial risk assessors recognize shortcomings with utilization of a uniform, exclusive application of the NRC 1983 approach, most have not explicitly attempted to take a completely fresh look at approaches to conduct MRA except for the ILSI (2000) approach which was loosely based upon EPA's draft Ecological Risk Assessment Guidelines.

EPA, FDA, USDA, DOD, and DHS have utilized their own unique approaches to conducting risk assessments, but it is important to keep track of other federally mandated

requirements that may apply to MRA. For example, when relevant, Executive Orders and OMB memorandums apply to MRA (EPA, 2002b; OMB, 2007b; Presidential Memorandum, 2009; OSTP, 2010).

1.10 What are Major Principles for MRA?

The principles in Text Box 1.1 are adaptable to microbial hazards in media other than food. However, different agencies may have different practices regarding how risk management and risk assessment activities are separated and the principles are framed in reference to food safety risks. Some agencies have more integrated management and assessment. In cases where the risk management and risk assessment functions are more integrated, transparency about how and where risk management is integrated in risk assessment is crucial. The NRC report *Science and Decisions: Advancing Risk Assessment* proposed that EPA should adopt:

> "… a framework for risk-based decision making. The framework consists of three phases: I, enhanced problem formulation and scoping, in which the available risk-management options are identified; II, planning and assessment, in which risk-assessment tools are used to determine risks under existing conditions and under potential risk-management options; and III, risk management, in which risk and nonrisk information is integrated to inform choices among options." (NRC, 2009)

The 2009 NRC framework also has a functional integration of risk assessment and risk management within a risk-based decision framework.

Text Box 1.1 General Principles of MRA

The ten general principles of MRA provided by the international Codex framework for microbiological hazards in food are below, as stated in Codex (2007b). The original numbering from the document is maintained. Principles 1-19 are general risk analysis principles and policy principles and are not included in this excerpt.

20. Each risk assessment should be fit for its intended purpose.

21. The scope and purpose of the risk assessment being carried out should be clearly stated and in accordance with risk assessment policy. The output form and possible alternative outputs of the risk assessment should be defined.

22. Experts involved in risk assessment including government officials and experts from outside government should be objective in their scientific work and not be subject to any conflict of interest that may compromise the integrity of the assessment. Information on the identities of these experts, their individual expertise and their professional experience should be publicly available, subject to national considerations. These experts should be selected in a transparent manner on the basis of their expertise and their independence with regard to the interests involved, including disclosure of conflicts of interest in

connection with risk assessment.

23. Risk assessment should incorporate the four steps of risk assessment, i.e. hazard identification, hazard characterization, exposure assessment and risk characterization.

24. Risk assessment should be based on scientific data most relevant to the national context. It should use available quantitative information to the greatest extent possible. Risk assessment may also take into account qualitative information.

25. Risk assessment should take into account relevant production, storage and handling practices used throughout the food chain including traditional practices, methods of analysis, sampling and inspection and the prevalence of specific adverse health effects.

26. Constraints, uncertainties and assumptions having an impact on the risk assessment should be explicitly considered at each step in the risk assessment and documented in a transparent manner. Expression of uncertainty or variability in risk estimates may be qualitative or quantitative, but should be quantified to the extent that is scientifically achievable.

27. Risk assessments should be based on realistic exposure scenarios, with consideration of different situations being defined by risk assessment policy. They should include consideration of susceptible and high-risk population groups. Acute, chronic (including long-term), cumulative and/or combined adverse health effects should be taken into account in carrying out risk assessment, where relevant.

28. The report of the risk assessment should indicate any constraints, uncertainties, assumptions and their impact on the risk assessment. Minority opinions should also be recorded. The responsibility for resolving the impact of uncertainty on the risk management decision lies with the risk manager, not the risk assessors.

29. The conclusion of the risk assessment including a risk estimate, if available, should be presented in a readily understandable and useful form to risk managers and made available to other risk assessors and interested parties so that they can review the assessment.

An overarching principle for MRA in this Guideline is to provide a systematic approach to the consideration of all information, models, and other tools that allow a suitable examination of the relationship of a microbial pathogen and human exposure to specific health endpoints and levels of effect. The process may be iterative, with increasing quality of data to reduce uncertainties with each iteration (see section 2.5.1). Details of this systematic approach will be covered in later chapters. The other major principles you can consider in conducting risk assessments are "transparency, clarity, consistency, and reasonableness" (TCCR) (EPA, 2000a).

a) **Transparency** is the foremost principle of the four because when followed it should help satisfy the other principles, in that it will lead to clarity and allow the evaluation of consistency and reasonableness. To provide transparency, the methods used and assumptions should be clear and understandable to the intended recipients of the assessment. From this, the audience for the risk assessment

should be able to assess the adequacy of the data and methods used to provide the information. It also means that conclusions drawn from the risk assessment and associated research that are science-based are separate from subsequent policy judgments and risk management decisions, and the use of methods, models, assumptions and defaults are clear.

b) **Clarity** means the manner in which the risk assessment is presented, such as how it is written and the appropriate use of illustrations or graphics to portray the findings. A main goal of this Guideline is to enable production of documents available for public scrutiny that are easily comprehendible and in simple language.

c) **Consistency** provides a context for the reader that can be readily understood for comparison to other similar documents and whether it provides harmony with an Agency's policy, procedural guidance, and scientific rationale. Where there are differences or inconsistencies, an explanation of why different approaches or interpretations were used and why the conclusions differ can be provided.

d) **Reasonableness** provides the audience with knowledge about the extent that professional judgments and assumptions are well founded. This can also address plausibility and comparisons against other similar types of risk assessment findings.

While these are the major underlying principles, other factors are also important, such as the assessment of data quality, data analysis, and peer review. Most Agencies have addressed their needs for these in separate Agency guidelines and all have adopted OMB Information Quality guidelines relevant to these issues (OMB, 2002).

The Office of Science and Technology Policy (OSTP), directs that, "When scientific or technological information is considered in policy decisions, the information should be subject to well established scientific processes, including peer review where appropriate, and each agency should appropriately and accurately reflect that information in complying with and applying relevant statutory standards" (Presidential Memorandum, 2009; OSTP, 2010).

In 2007, OSTP and OMB reissued the 1995 Principles for Risk Analysis (OMB, 2007b). The six principles pertaining to risk assessment include:

1. Agencies should employ the best reasonably obtainable scientific information to assess risks to health, safety, and the environment.
2. Characterizations of risks and of changes in the nature or magnitude of risks should be both qualitative and quantitative, consistent with available data. The characterizations should be broad enough to inform the range of policies to reduce risks.

3. Judgments used in developing a risk assessment, such as assumptions, defaults, and uncertainties, should be stated explicitly. The rationale for these judgments and their influence on the risk assessment should be articulated.

4. Risk assessments should encompass all appropriate hazards (e.g., acute and chronic risks, including cancer and non-cancer risks, to human health and the environment). In addition to considering the full population at risk, attention should be directed to subpopulations that may be particularly susceptible to such risks and/or may be more highly exposed.

5. Peer review of risk assessments can ensure that the highest professional standards are maintained. Therefore, agencies should develop policies to maximize its use.

6. Agencies should strive to adopt consistent approaches to evaluating the risks posed by hazardous agents or events.

These principles have been incorporated throughout this Guideline.

2. PLANNING AND SCOPING

Planning and scoping will help ensure that a risk assessment is relevant and well done. The NRC 2009 framework recommended "increased attention to the design of risk assessment in its formative stages [and] that planning and scoping and problem formulation, as articulated in EPA guidance documents (EPA, 1998a, 2003), should be formalized and implemented in EPA risk assessments" (NRC, 2009). Rigorous preparation is needed at the start of the risk assessment process, to facilitate communication during and following the risk assessment and to ensure that all issues are sufficiently vetted, all participants are clear on the objectives and goals, and managers are clear about the commitment of personnel and other resources (EPA, 1992; NRC, 1996).

For more detailed descriptions of the usefulness and implementation of the planning and scoping process, the FDA, USDA, and EPA sources provide general information on how to proceed (FDA, 2002; USDA, 2003; EPA, 2000a, 2002a; NRC 2009). This chapter provides an overview of this first step in the risk assessment process. Several interesting case studies from ecological risk assessment are presented in EPA's *Lessons Learned on Planning and Scoping for Environmental Risk Assessments* (EPA, 2002a).

2.1 What is Planning and Scoping?

Planning and scoping can be viewed as a process that defines the purpose and scope of a risk assessment and focuses the issues and approach(es) involved in performing the assessment. A clearly articulated purpose and scope provides a sound foundation for later judging the success of the risk assessment and for an effective risk characterization. In a sense, the planning and scoping process lays out a "road map" for how the risk assessment will be accomplished.

EPA considers Planning and Scoping steps to be as follows (based on EPA, 2003a, 2004b):

a) Defining the purpose of the assessment

b) Defining the scope of analysis and products needed

c) Agreeing on participants, roles and responsibilities

d) Agreeing on depth of assessment and analytical approach (for example, will the risk assessment include static or dynamic modeling)

e) Agreeing on resources available and schedule

f) Formulating the problem (see Section 2.1.1)

g) Developing the conceptual model (see Section 2.6.3)

h) Constructing the analysis plan (see Section 2.6.4)

See Section 2.3 for an overview of how CFSAN does planning and scoping for major risk assessments.

2.1.1 What is Problem Formulation?

The problem formulation exercise is a discussion and analysis activity that involves all relevant parties including the risk manager, risk assessment team, risk communication specialist and, when appropriate, relevant stakeholders and interested parties. The outputs of the problem formulation are:

a) A definition of the valued entity and endpoint: what is the entity that should be protected and what are the undesirable effects that you are trying to avoid. For MRA this may involve a policy determination of what the "valued entity" is (e.g., general population, young children, pregnant women, immunologically compromised) and what is considered to be an appropriate level of protection (ALOP) against infection or disease.

b) A Conceptual Model that lays out the anticipated exposure scenarios of the microorganisms from the source to the receptor. With MRA this may be the movement of enteric pathogens from some source (e.g., treatment works, manure application to a field, critical point in the food processing system) to the subpopulation of concern and highlights various hazardous events that may lead to increased risk and where risk management may be most effective.

c) An Analysis Plan which provides a road map for addressing the problem. In short, it is analogous to an experimental design. In the analysis plan risk hypotheses generated earlier are examined and discussed; the relationships between pathways and valued entities are further examined; and the level of precision and data quality are considered in light of available information.

2.2 What are the Benefits of Planning and Scoping?

The planning and scoping process helps everyone involved in the risk assessment understand how the risk assessment fits into the overall decision making process. Planning and scoping promotes:

a) initial planning to identify appropriate timelines and necessary resources, thereby improving efficiency;

b) agreement among principle parties regarding the goals, commitment, time-frame and resources, by setting realistic expectations;

c) the prospect of less unanticipated controversy, since all interested parties contribute, areas of disagreement can be dealt with upfront, not left as a surprise at the end;

d) identification of and participation by those from many disciplines (e.g., microbiologists, toxicologists, economists, lawyers) to help in the process thereby ensuring that each risk assessment and characterization is useful for the intended audience(s);

e) an understanding of the degree of complexity needed in the risk assessment to adequately inform the decision at hand; and

f) better informed decisions.

2.3 What do I Consider When Deciding to Initiate a MRA?

Unless your agency is required to conduct a particular risk assessment, managers need to decide whether a risk assessment is appropriate, feasible and will actually be performed. This decision is commonly made during planning and scoping. Deciding not to initiate a risk assessment may be an acceptable outcome of planning and scoping. The agency may decide that a decision can be reached without conducting a risk assessment or other priorities or risk assessments may take precedence. There are several criteria you can consider for identifying a candidate risk assessment:

a) Characteristics and importance of the hazard(s) of concern

b) Magnitude (presence, prevalence, concentration of hazards) and severity (impact on public health) of the risk

c) Urgency of the situation

d) Subpopulations of concern

e) Other factors associated with specific hazards (such as water treatment processes, food processing, cooking, cross contamination)

f) Availability of resources (time, money, staff)

A risk assessment may be required to comply with regulatory analysis requirements (OMB, 2003). The Office of Information and Regulatory Affairs has provided a checklist to assist agencies in producing regulatory impact analyses (RIAs), as required for economically significant rules by Executive Order 12866 and OMB Circular A-4 (E.O. 12866, 1993; OMB, 2010; OMB, 2003). Appropriate assessments of risk may be necessary to address international trade agreements (e.g., World Trade Organization Sanitary and Phytosanitary Agreement). Ensuring that each assessment of risk is fit for its intended purpose and is based on scientific data most relevant to the national context ensures that the effort and scope of the assessment of risk are appropriate for the risk management questions being raised so that practical risk management options can be formulated.

Other specific examples of when a risk assessment may be appropriate include:

a) Review of the reliability or utility of a standard

b) Cases where the current standard is inconsistent with other government policies, guidelines, or thresholds

c) Cases where an agency has been petitioned for a regulatory action

d) Establishment of standards for regulatory action

e) Evaluation of the public health implications of different tolerable risk levels

f) Cases where a data gap analysis is desired

g) Cases where the hazard is a serious health issue, emerging pathogen and/or public health concern

h) The exposure system is complex.

FDA's Center for Food Safety and Applied Nutrition's (CFSAN) *Initiation and Conduct of All "Major" Risk Assessments within a Risk Analysis Framework* outlines how CFSAN selects, conducts, and communicates food safety risk assessments (FDA, 2002). CFSAN's process for identifying and selecting major risk assessments is divided into four phases (FDA, 2002):

a) Phase 1: Concept Generation – Collect ideas and maintain a list of potential risk management questions for which a risk assessment would assist with policy decisions. Develop justification for candidate risk assessments, including, purpose of assessment, scope of problem, importance to the Center, and how the result will be used by the Center.

b) Phase 2: Problem Identification – The candidate risk assessment and supporting information (justification) are reviewed to determine whether the assessment meets the Center's needs. This phase results in one of three recommended actions, conduct data feasibility study, not required for regulatory decision, or more information needed to make decision.

c) Phase 3: Data Feasibility (Evaluation and Recommendation) – Information is collected and reviewed to determine availability of data needed to answer risk assessment question(s). This phase results in one of four recommended actions, conduct quantitative risk assessment, conduct qualitative risk assessment, more research needed, or modify question and conduct alternative assessment.

d) Phase 4: Disposition (Selection) – Using the results from the data feasibility determination as an aid, risk assessment(s) to be conducted are selected. Decision is based on technical merit, resource availability, the Center's priority needs, and other legitimate factors.

2.4 Who Can be Involved with Planning and Scoping?

All interested parties could be involved with planning and scoping, but, from a practical standpoint, bringing everyone together at one time and discussing all aspects of the effort at once may be difficult and therefore would probably include more than a single session. Major participants include relevant risk managers, risk assessors, and other members of the "team" working on the decision that needs to be made. In addition to the appropriate microbiologists, infectious disease experts and/or individuals trained in infection control and other members such as economists, lawyers, engineers, policy makers, and communicators should be included. A risk assessment dialogue among the risk managers, risk assessors, risk communicators, economists, and other technical experts should develop the broad dimensions and elements of the risk assessment, the management questions and goals for the assessment, a tentative budget and schedule, and an approach for conducting the risk assessment. The risk assessor, risk manager, and planning team interactions may vary depending on the Agency. Generally, they meet to evaluate and select the kind of risk information, exposure scenarios, and assessment issues to be covered. Risk assessors should be very involved in planning and scoping at every stage, because risk managers will have technical and data related questions that may require your input. As a risk assessor, you will want to weigh in on whether the tools and data available can answer the questions being posed. Planning and scoping would not be successful without the technical input of risk assessors and the management perspectives from risk managers. In addition, the risk manager should consider stating explicitly any reasons to limit the technical scope of the assessment, and you should consider including details on resource limitations, data availability and quality, and methods availability. In other words, you can be very clear and transparent about what you plan to include in the risk assessment and what you plan to exclude from the risk assessment and why. Further elaboration on the role of risk assessors is found in Section 2.8.

Agencies with a hierarchy of risk managers will have a hierarchy of risk management activities. Planning and scoping can outline the management hierarchy with a degree of detail that is appropriate for planning deliverables and milestones. The roles are not exclusive to each level and in practice would likely overlap considerably. Risk managers at all levels are responsible for ensuring appropriate communication with managers above and below their level. It is important to identify the team leader and his/her basic responsibilities at the outset to avoid misunderstandings as the process develops. Risk managers are generally the decision makers in their organization. The top manager is the ultimate decision maker for his/her organization and is accountable for both the risk characterization process and products in his/her office. However, every risk manager may make decisions appropriate for their level in the management hierarchy.

You should be prepared to communicate with many levels of managers particularly if you are working on a high profile risk assessment.

The importance of stakeholder involvement during the planning and scoping process depends upon the nature of the problem, their interest, and ability to contribute. Stakeholders can be identified early in the planning and scoping of the risk assessment. How stakeholders can be involved most effectively needs to be decided on a case-by-case basis. Public involvement, early and often, leads to a much clearer risk assessment product. This involvement also allows for flexibility and buy-in for future decision making if the need arises to deviate from the original plan.

Affected parties can share their points of view about the risk and how it could be managed. Their input is particularly helpful in determining what should be included in the assessment, how they might be affected or exposed to the risk, and what additional data or exposure scenarios should be developed.

You should determine if your agency has any stakeholder involvement guidance and integrate those practices into planning and scoping. Stakeholder involvement practices may be well developed and formalized or haphazard and inconsistent. Building the necessary relationships with stakeholders to maintain dialogue takes considerable effort, but this should not deter you and risk managers from engaging in this important activity. Risk communication specialists should be engaged in developing stakeholder involvement plans. The National Academies of Science report, *Public Participation in Environmental Assessment and Decision Making*, is a resource for considerations when engaging stakeholders (NRC, 2008).

2.5 How Can the MRA be Used?

Although risk assessments conducted by different agencies are not used for the same purposes, all agencies perform risk assessments with one or more of the following goals in mind (adapted from U.S. Army Center for Health Promotion and Preventive Medicine [USACHPPM], 2009):

a) To mitigate, e.g., to mitigate adverse effects or risk from a specific event

b) To confirm, e.g., to determine if regulations, policies, standards, criteria, and/or goals are adequate

c) To decide whether and/or how to regulate, e.g., as needed to establish regulations, policies, standards, criteria, and/or goals

d) To investigate, e.g., to determine research or other requirements that would enhance predictive and/or risk ranking capabilities, or facilitate completion of screening or feasibility assessments

Depending on the risk assessment's purpose, a particular assessment approach may be employed. The appropriate risk assessment approach for a specific risk management problem or decision depends on the question(s) that need to be answered and the availability of data. For example, if adequate data are available, a quantitative risk assessment is possible; if fewer data are available, a qualitative assessment or a data gap analysis (discussed in Sections 2.6.5 and 6.4) may be more appropriate.

2.5.1 What "Depth" Can I go into in the Risk Assessment?

A major consideration for the risk assessment approach is how much detail or "depth" you need to incorporate to address the risk management question(s) or decision. Due to various management needs, the risk assessment approach is not necessarily a one-size-fits-all approach. This guidance is intended to provide flexible methods for supporting different types of assessments (screening, safety) and outputs (qualitative or quantitative), as described in the American Society for Microbiology (ASM) Press book (Schaffner, 2008). Guidance from Codex Alimentarius (1999) and World Organization for Animal Health (OIE, 1999) described qualitative and quantitative outputs as equally valid (Wooldridge, 2008). Woodridge (2008) provides detailed discussion of qualitative and quantitative assessments, and other risk analysts (Dennis et al., 2008) discuss both estimates of risk and safety, mortality for listeriosis and allowable ('safe') levels of *Vibrio* in seafood, respectively.

Be aware that the terms referring to the different types of assessments are specifically defined in different contexts; care should be used when "naming" or referring to types of assessments. For example, this Guideline is specific to the selection and conduct of risk assessment, not a safety assessment. One difference is that risk assessment estimates the likelihood and/or frequency of adverse health outcomes resulting from an exposure and, in some cases, sources of risk and quantitative reductions in risk based on various interventions. While much of this guideline is relevant to a safety assessment, a safety assessment may estimate the likelihood and/or frequency of exceeding a specified threshold of concern (e.g., predetermined regulatory limits or standards), or provide a determination of what is "safe" based on the conventions of the standard-setting procedure. For example, Codex standards traditionally specify maximum limits for additives and residues in foods based on the concept of "no appreciable risk" and for contaminants in foods based on the concept of "as low as reasonably achievable" (FAO/WHO, 1997). Risk-related terms in some statutes have formal definitions.

There are many cases where you will likely need to perform a screening risk assessment versus a fully developed one. This is usually done when a very time critical decision is needed (e.g., quick mitigation action is required after an event; imminent exposure to a microbial hazard is identified). Screening risk assessments often provide a conservative, health-protective, estimate of possible risk that is based on the more readily available data. It is likely that you will resort to default assumptions to bridge data gaps that you cannot wait for research to fill. It is important to be transparent about the amount of uncertainty in your screening estimate and discuss whether the uncertainty

causes underestimates or overestimates of risk based on the assumptions applied. This may be followed up by a more detailed assessment that will need to be conducted or risk managers will need to take action(s). Thus, you also may be asked to return to the assessment and refine and recalculate your estimates based on further gathering of data with more time allotted (e.g., quick sampling assays, use of surrogate data, expert elicitation). Alternatively, there may be a risk management decision to conduct a full risk assessment.

A comprehensive ("major") risk assessment requires a substantial commitment of resources. Thus, this depth of risk assessment is not appropriate when risk managers do not need this level of sophistication to make a decision. Circumstances that may not warrant a quantitative risk assessment would include, for example, a risk that is well described by definitive data, a problem that is relatively simple, or an issue that is not of regulatory concern. However, a comprehensive risk assessment is a powerful tool to help risk managers evaluate and interpret information when the data describing a hazard are incomplete, the exposure system is complex, or the issue is of high regulatory or stakeholder concern. Details for a major risk assessment are presented in this guideline.

Risk assessments can be either qualitative or quantitative in their description of the likelihood of adverse health effects, depending on the extent of the data and knowledge available, the existence of models or other tools for quantitative predictions, the complexity of the problem, the scope and nature of the question(s) posed by the risk managers, and the time available to conduct the assessment. In quantitative assessments, the risk is expressed as a mathematical statement of the probability of illness or death after exposure to a specific hazard, and it represents the cumulative probabilities of certain events happening and the uncertainty associated with those events. Conversely, qualitative risk assessments use verbal descriptors of risk and severity as well as uncertainty, and often involve the aggregation of assumptions.

2.5.2 What Are Examples of Types of MRA?

Risk assessments can take various forms depending on the agencies' needs. Examples of risk assessment types that you are likely to use include risk ranking, product pathway, risk-risk, geographic, and for sustainability assessments:

a) **Risk ranking** – Risk ranking assessments compare the relative risk among several hazards. For example, this type of assessment might involve a single pathogen associated with multiple foods, a single food that has multiple pathogens, or multiple pathogens and multiple foods. Risk ranking assessments can help establish regulatory program priorities and identify critical research needs. The Food and Drug Administration/U.S. Department of Agriculture (FDA/USDA) *Listeria monocytogenes* risk assessment is an example of a risk ranking assessment (FDA/USDA/CDC, 2003).

b) **Product pathway analyses** – In product pathway assessments, the factors that influence the risk associated with specific vehicle/hazard pairs are examined. For

food, it ideally starts at the farm and ends with consumption. This type of assessment technique helps identify the key factors that affect exposure including the impact of potential mitigation or intervention strategies on the predicted risk. The FDA *Vibrio parahaemolyticus* risk assessment is an example of a product pathway analysis (FDA, 2005).

c) **Risk-risk** – In risk-risk assessments, a trade off of one risk for another is considered, i.e., reducing the risk of one hazard increases the risk of another. An example of this would be a determination of the impact on public health by treating drinking water with a chemical (risk to chlorine exposure) versus the impact of exposure to pathogenic organisms in water that is not treated.

d) **Geographic** – In a geographic risk assessment, the factors that either limit or allow the risk to occur in a given region are examined. The risk of introduction of disease agents through water, air, food animals, or animal products in the U.S. (e.g., intentionally as in a bioterrorism act or unintentionally) can be examined. For example, the risk of introduction of bovine spongiform encephalopathy (BSE) into the U.S cattle herds and the subsequent risk of variant Creutzfeldt-Jacob Disease (vCJD) in humans by the transmission from cattle through meats and animal product pathways might be examined using a geographical approach.

e) **MRA within sustainability assessments** – Using a systems-level assessment over the life-time of its technical components, sustainability assessments attempt to account for human health, ecosystem health, and economic considerations. The human health aspects include chemical and MRAs. The difference here is to include the MRA over the expected life-time of the technical system and via all exposure pathways of pathogens to humans (e.g., drinking water, reuse waters, aerosols pathways, recreational exposures, and contaminated soils/foods).

2.5.3 What are Threat and Vulnerability Assessments for High-Consequence Biological Events?

This Guidance is primarily for risk assessment and does not go into detail regarding the related disciplines of threat and vulnerability assessments. Nevertheless, it is important to understand these specialized tools for evaluating the susceptibility of systems and facilities to potential threats, such as adversarial actions (e.g., vandalism, insider sabotage, or terrorist attack), natural disasters, and other emergencies. Therefore, threat and vulnerability assessments are described briefly below.

Similar to a risk assessment, threat and vulnerability assessments identify threats and characterize the nature, probability, and magnitude of adverse effects. However, vulnerability assessments not only consider the risk to the infrastructure itself, but also consequences to the surrounding community and environment, such as economic losses and human health impacts from consuming contaminated food. Vulnerability assessments also usually identify corrective actions that can reduce the risk or lessen the severity of potential consequences. Threat and vulnerability assessments are often used

as tools to prioritize which threats or vulnerable operations need to be addressed first. They are not actually a "risk assessment" per se, although sometimes the results can be similar to those produced in a classic microbial risk assessment, and the results can help to inform risk management decisions.

Since the anthrax attacks of 2001, there has been growing awareness of the potential for the deliberate use by terrorists of biological agents that pose a threat to the health and welfare of the exposed subpopulation and to the economy. It may be necessary to assess the risks associated with intentional contamination of the food or water supply with biological agents, or release of biological agents as an aerosol into highly populated indoor or outdoor public areas.

Threats and vulnerabilities can be characterized sequentially or as part of a single assessment. Initially, the nature and probability of potential threats are determined. Threat assessments of intentional contamination include information on the availability and ease of production of agents, as well as the sophistication and capabilities of the likely perpetrator. Because of a lack of data and the typically random nature of terrorist events, the probability of threats can usually not be quantified statistically.

A vulnerability assessment helps you consider the potential for impact of significant loss of operational and/or functional capability from a deliberate attack or an anticipated natural disaster, as well as the vulnerability of the facility/location itself to the identified threats. For example, projected impact of loss can be viewed as the degree to which the operation and critical infrastructure of a metropolitan drinking water distribution system may be impaired by a successful attack involving a biological threat or a natural disaster resulting in extensive biological contamination of primary source waters feeding the distribution system. Consequences might also include adverse health impacts to customers of the water system and economic losses to the community.

Each scenario is unique and represents a different combination of factors affecting the probability and the consequences of an intentional release of a biological threat agent. The health-related factors might include the toxicity of the agent; the concentration of the agent; fate of the agent in during processing, storage, distribution, and preparation of a food product; quantity of contaminated food or water consumed; contagiousness; and the availability and effectiveness of countermeasures, such as recalls and medical treatment. The economic and public health impacts from a biological attack could be devastating and catastrophic. Contaminated facilities may be out of commission for extended periods of time for decontamination, and may never be able to return to operation or profitability. The public may also lose confidence in the safety of the food and water supply.

An effective vulnerability assessment provides a prioritized plan for mitigation measures, such as security upgrades, modifications of operational procedures, and/or policy changes, and provides a framework for developing risk reduction options and associated costs.

A preemptive targeting prioritization tool you can use for a vulnerability assessment is the CARVER plus Shock method (FDA, 2007), which has been adapted for use in the food sector. This tool can be used to assess the vulnerabilities within a system or infrastructure to an attack. It allows you to think like an attacker by identifying the most attractive targets for attack. By conducting such a vulnerability assessment and determining the most vulnerable points in your infrastructure, you can then focus your resources on protecting your most vulnerable points.

CARVER is an acronym for the following six attributes used to evaluate the attractiveness of a target for attack (FDA, 2007):

a) **C**riticality – measure of public health and economic impacts of an attack

b) **A**ccessibility – ability to physically access and egress from target

c) **R**ecuperability – ability of system to recover from an attack

d) **V**ulnerability – ease of accomplishing attack

e) **E**ffect – amount of direct loss from an attack as measured by loss in production

f) **R**ecognizability – ease of identifying target

In addition, the modified CARVER tool evaluates a seventh attribute, the combined health, economic, and psychological impacts of an attack, also called the Shock attributes of a target (FDA, 2007).

2.6 What is Discussed During Planning and Scoping and What Products Emerge?

Principal outputs from planning and scoping can include various products that are appropriate to the management objectives and the plan for analysis of the risk. Not all of these examples are necessary for every occasion and certain ones will be more appropriate for your particular problem. Further, some outputs can be contained in other products (e.g., the risk profile document can contain many of the listed products). You need to discuss what outputs will be generated during the planning and scoping discussions. In general, many of these products are good candidates for peer review. Peer review early in the risk

Example Products of Planning and Scoping

Statement of Concern
Statement of Purpose and Objectives
Background Section
Scope
Scenarios
Literature Review
Data Inventory
Tools and Methods Inventory
Risk management questions or Charge
Risk Profile
Conceptual Model
Value-of-information analysis
Communication Plan
Analysis Plan
Work Plan
Data quality objectives

assessment process can provide timely insights, corrections to assumptions, and directions on proper ways to proceed during the risk assessment.

During planning and scoping, participants can engage in a dialogue to answer the following questions and commit the outcome of those discussions to paper to facilitate mutual understanding (paper products underlined):

a) **What is the motivation for the risk assessment?** Drafting a Statement of Concern is a good way to reach a common understanding of what broad issue the risk assessment will address. Describe in simple terms what hazard is being addressed and how it is thought to relate to human health for an exposure scenario. Include any other driving factors for the risk assessment, such as a food safety issues, regulatory requirements, public concern, or new scientific findings.

b) **What are the management goals, issues, questions, and policies that need to be addressed?** A Statement of Purpose and Objectives is a concise paragraph that addresses the management goals. The management questions are framed as questions and are written down. The management questions should be designed so that if answered, the risk manager has the information needed to inform the decisions that need to be made.

c) **What is the context of the risk assessment?** Risk assessments are done in historical and social contexts. It is helpful to summarize in a Background Section any previous risk assessments that addressed the same or similar hazards. In particular, if your agency has conducted a previous risk assessment on the hazard, then you can summarize the relationship between the current and previous risk assessment. The context of a risk assessment may include new mandates, regulatory requirements, policy developments, technical advancements, risk assessment method and tool advancements, and new or enhanced data sets.

d) **What is the scope and coverage of the risk assessment?** The Scope outlines the scenarios the risk assessment will cover. Answering the scoping questions below can help ensure that you have the information regarding scope that is necessary to conduct the MRA.

    1) Which infectious disease hazard is being addressed (pathogen strain[s], or indicator[s], taxon [genus, species, strain/biovar])? Define the hazard.

    2) Which human subpopulations will be included in the risk assessment (e.g., general population, life stages, or geographically defined subpopulations)? Describe which subpopulations are explicitly included in the risk assessment model, which will be accounted for implicitly, and which subpopulations may be excluded by the risk assessment model (e.g., most extreme behaviors).

    3) What health outcomes or endpoints are addressed by the risk assessment, and how is the health outcome measured? Clearly defining the health

endpoint is important for transparency and also focuses the scope of the risk assessment (e.g., infection, disease symptom/s, mortality).

4) What unit and routes of exposure are relevant and why? Determine the time-span of exposure relevant to the decision.

5) For risk assessments designed to derive nominally or presumptive "safe" levels of microorganisms (i.e. levels below a threshold of regulatory concern), what level of protection will be provided, and what is the technical or policy justification for that level? Transparency in public health objectives is important.

6) What specific exposure scenarios should be modeled? List specific scenarios the risk managers would like to model (varying the inputs), including desired spatial and temporal features.

e) **What type of risk assessment is needed to address the risk management question(s)?** Section 2.5.2 describes different types of risk assessments, including quantitative, qualitative, risk ranking, product pathway, risk-risk, geographic, and systems-level.

f) **What is the state of the current knowledge?** The planning and scoping can include an overview of current knowledge and can be used to outline the topics that will be reviewed in more depth in the rest of the risk assessment. A Literature Review will likely help with understanding the current state of the science.

g) **What and where are the available data?** In addition to identifying the available data, a Data Inventory can address data relevance, how the data will be used, data accessibility considerations, and initial data quality evaluations. An inventory may also provide notice to the public of the data currently available to the agency in a call for data. See Section 2.6.6 for further discussion of data quality.

h) **How do I know what questions the risk assessment needs to answer?** The Risk management Questions or Charge are usually written down and discussed iteratively between the risk assessment team and risk managers so there is a common understanding of the questions.

i) **What are the information/data needs of other members of the "team?"** The risk assessment may be part of a larger project, such as an economic analysis. There may be economic, social, or legal analyses that need to be coordinated with the risk assessment.

j) **How will you model the risk?** A Tools and Methods Inventory should include statistical methods for estimating model inputs and tools for addressing uncertainty and variability and should make an initial determination of which methods and tools are likely to be most useful.

k) **What are possible risk assessment or risk management options?** Particularly for risk assessments that are needed to evaluate intervention strategies or needed to support regulatory determinations, different options should be presented as different scenarios for the risk assessment. Those Scenarios can be clearly stated during problem formulation and may evolve during risk assessment iterations.

l) **What are the logistical considerations for conducting the risk assessment?** Logistical considerations can go into the Analysis Plan (see Section 2.6.4) or a Work Plan document. These include:

1) resources available to do the assessment, including funding and staff time;

2) participants in the process and their roles;

3) plans for coordinating across offices, with other agencies and with stakeholders; and

4) scheduling (e.g., milestones, deliverable due dates, quality audits, meetings), including provisions for timely and adequate internal, independent external peer review, and if required, interagency review as per EO 12866.

m) **How will planning activities and results be communicated to senior managers and to the public?** Chapter 8 discusses risk communication within the context of risk assessment.

n) **What are the legal considerations and constraints that may shape the ultimate decision and supporting risk assessment?** The technical office within the agency that is conducting the risk assessment can engage the agency's legal department, according to normal practices within that agency. Legal context can be discussed in the Background Section.

o) **How and at what iterations will the risk assessment be peer reviewed?** OMB has published *Information Quality Bulletin for Peer Review*, which provides general peer review guidance and sets minimal expectations for the review of scientific information (OMB, 2004). Most agencies have agency specific peer review guidance that complies with the OMB guidance. You should follow your agency's peer review guidance. For example, EPA's *Peer Review Handbook* (EPA, 2000d, 2006c) provides guidance on selection of peer reviewers that includes where to find peer reviewers, what mix of expertise may be important, representing diversity of disciplines, and limiting conflicts of interest. According to the National Committee on Radiation Programs (NCRP, 1996), an expert has the following characteristics:

1) training and experience in the subject area resulting in superior knowledge in the field;

2) access to relevant information;

3) an ability to process and effectively use the information; and

4) is recognized by his or her peers or those conducting the study as qualified to provide judgments about assumptions, models, and model parameters at the level of detail required.

In summary, the planning and scoping discussion may include a preliminary characterization of exposure and effects, as well as examination of scientific data and data needs, policy and regulatory issues, and scenario-specific factors to define the feasibility, scope, and objectives for the risk assessment. The level of detail and the information that will be needed to complete the assessment also are determined. Just as important, planning and scoping helps set the boundaries of the problem(s) addressed and the scope of the MRA.

For perspective, risk managers (decision makers) naturally desire more information, less uncertainty, and more in depth interpretation, when the impact of their decisions increases. They also want to know the financial and social implications of possible decisions. The risk assessment may not be the appropriate support analysis to address these issues, but if done well, risk assessment can be a critical input. These are aspects that may be discussed during planning and scoping.

2.6.1 What are Risk Management Questions and What is the Charge?

A brief description of management questions is presented in Section 2.4. Generating specific risk management questions for a risk assessment helps formulate a clear, focused charge that identifies the technical and scientific issues on which you need to address and suggestions for conducting the risk assessment. Formulating these questions usually requires significant interaction between risk assessors and risk managers, as well as dialogue with appropriate other parties (e.g., those with relevant information about the potential hazard). You want to be sure that the questions you generate will focus the risk assessment to provide the appropriate analyses that can properly inform the risk management decision at a level of detail appropriate for the issue. The resulting risk assessment can be designed to address and answer as best as possible the risk management questions posed.

The charge focuses the assessment by presenting specific questions and concerns surrounding such issues as the comprehensiveness of the data, information, and literature, the soundness of the methods proposed, the scientific support for the assumptions that may be employed, and the sensitivity of the results to possible alternative assumptions. As a general rule, time is well-spent preparing a good set of questions or a charge, as they are crucial for an effective risk assessment and ultimate decision. In this context the charge is the set of questions and does not imply a formal charge.

2.6.2 What is a Risk Profile?

Codex defines a risk profile for food safety as: a description of a food safety problem and its context that presents in a concise form, the current state of knowledge related to a food safety issue, describes potential microbiological risk management options that have been identified to date, when any, and the food safety policy context that will influence further possible actions… Consideration of the information given in the risk profile may result in a range of initial decisions, such as commissioning a microbiological risk assessment, gathering more information or developing risk knowledge at the level of the risk manager, implementing an immediate and/or temporary decision (Codex Alimentarius Commission, 2007).[2] A typical risk profile includes a description of the situation, product, or commodity involved; information on pathways by which consumers are exposed to the microorganism; possible risks associated with that exposure; consumer perceptions of the risks; and the distribution of possible risks among different segments of the population. For a list of information that Codex recommends including in a risk profile see Codex (2007).

A risk profile assists in identifying the risk management questions that need to be addressed. The risk profile should be clearly and thoroughly documented, so that risk managers can use it to decide on further action in relation to a specific health issue. If links are made between risk profiles for other risk assessments, risk profiles can provide the basis for qualitative ranking of problems for subsequent risk management.

Notably, risk profiles can also be used as decision tools that do not lead to risk assessment. For examples see the New Zealand Food Safety Authority.[3]

2.6.3 What is a Conceptual Model?

A conceptual model is a written or visual representation you construct of predicted relationships between the hazard and exposed subpopulations.[4] It is based on your problem formulation and working hypotheses, supported by preliminary data and information, and used to organize the conduct of a MRA.

The conceptual model depicts the movement of the hazardous agent to the host. Other tiers of conceptual models may identify variables and data needed to conduct the MRA. A conceptual model (e.g., a source-pathway-receptor model; "farm-to-fork" model) can be developed early in the planning and scoping process, to the level necessary to address the risk assessment's purpose. For example, in some cases, a pathogen may be available in multiple media and cause different diseases depending on the route of

[2] "Risk profile" is sometimes used to refer to summary information at the end of each chapter in frameworks based on the ecological risk assessment framework.
[3] http://www.foodsafety.govt.nz/science/risk-profiles/
[4] The conceptual model in planning and scoping differs from conceptual models that are used to map how parameters are related in modeling software. The term conceptual model is used in both contexts in this Guideline.

exposure. In those cases, during planning and scoping, you can clearly identify the medium, pathway of exposure, and route of exposure to be assessed.

Developing a sound and useful conceptual model may require several iterations. With the conceptual model, you can describe or visualize the relationships among the assessment and measurement endpoints, the data required, and the methodologies that will be used to analyze the data. An overall high-level conceptual model as well as more detailed conceptual models that cover just dose-response or exposure assessment components may be useful (EPA, 1998a).

2.6.4 What is an Analysis Plan?

The analysis plan lays out the approach to be taken by the risk assessment team. It shows how data sources and information will be used and integrated in the assessment and how measurement endpoints (e.g., fecal shedding) and uncertainties are related to the assessment endpoints (e.g., morbidity and mortality). As a product of planning and scoping, the analysis plan can act as a bridge to the risk assessment. The analysis plan is the implementation strategy for performing the risk assessment and addressing the decision needs. It documents the agreements made during the planning and scoping process and provides details on how the risk assessment will proceed. This provides transparency to the whole process. In addition, the analysis plan provides measures against which the final risk assessment and its risk characterization can be evaluated. As the risk assessment proceeds, the analysis plan may need to be revised to ensure that the risk assessment still meets the decision needs (EPA, 1998a). If a separate work plan for logistics is not developed, then staffing, scheduling, and resource details can also go in the analysis plan.

2.6.5 How are Data Gaps Identified and Addressed in the Context of Planning and Scoping?

Subsequent chapters in this Guideline address many of the tools used to address data gaps. Incomplete information and data gaps are a significant challenge throughout the risk assessment discipline. Much of the "art" in risk assessment involves the judgments regarding incomplete data and data gaps. In addition to missing data, there are commonly questions about the degree to which available data are representative of the actual conditions being assessed.

The extent to which a data gap exists is ultimately a matter of scientific judgment within the context of what is an acceptable level of confidence. Different assessors and managers may have different comfort levels for making decisions based on the same data. In some cases there may be differing options about how representative the data are and whether the data adequately fit in the risk assessment scenario. The quality of existing data is also considered when determining if a data gap exists (Section 2.6.6). To reduce the heterogeneity of comfort levels, many different systematic schemes for evaluating data quality, completeness, and applicability have been developed. Statistical approaches are standard for evaluating data quantity, but still require a judgment about the

appropriate confidence level for decision making. If the risk assessors and risk managers on your team cannot agree on what constitutes a significant data gap, the team may need to take a step back and first agree on criteria for evaluating data. With some data gaps, risk managers look to risk assessors to tell them whether the data are sufficient, while risk assessors may claim that a policy decision needs to be made by the risk managers regarding setting the threshold for sufficiency. These types of situations can stall a risk assessment, but ultimately the group has to reach agreement on the judgment call or policy decision. If the existence of a data gap is not obvious and agreed upon easily, the evidence used to support the identification of the data gap should be clearly documented.

Stakeholders may have strong opinions about data gaps. Some may demand a decision in the absence of data, while others may interpret the goal of science-based decision making to mean that more complete data must be available to make a decision. Robust planning and scoping should be able to predict which data gaps have the potential to cause the most debate.

Every parameter in the risk assessment will have some level of incomplete information. Ranking the importance of the data gaps can help you focus resources on the most critical data gaps that, if filled, could influence the risk assessment results the most. Whether a data gap "matters" to the risk assessment results can be evaluated by conducting sensitivity analysis (Sections 5.3.3 and 6.7) or value-of-information (VOI) analysis (Section 2.6.7). Once a data gap is identified and determined to be important it can be a matter of scientific judgment or a policy decision that determines how the data gap will be addressed in the risk assessment. You can fill the information needs in the near term using existing data, in the midterm by conducting tests with currently available test methods to provide data on the topic of interest, or over the long term to develop better, more realistic understandings of exposure and effects and to construct more realistic test methods to evaluate pathogens of concern. In cases where an aspect of risk is likely to be important but insufficient data are available, highlight the deficiency or use judgment or assumed values to approximate the missing data (see Section 6.4 for further discussion). Such judgments and approximations should be clearly described and the implications explained in the risk characterization.

OSTP states in their scientific integrity memo that, "The accurate presentation of scientific and technological information is critical to informed decision making by the public and policymakers. Agencies should communicate scientific and technological findings by including a clear explication of underlying assumptions; accurate contextualization of uncertainties; and a description of the probabilities associated with both optimistic and pessimistic projections, including best-case and worst-case scenarios where appropriate" (OSTP, 2010).

Depending upon the circumstances, the utility of a risk assessment may be compromised if important policy decisions are put on hold while waiting for more research results. Risk assessors and risk managers need to balance the need to obtain more data/information against the need to make a timely decision. The data gaps

identified in planning and scoping may be very useful to establish a research program and/or agenda to address current data gaps.

Expert opinion or judgment is a common source of information used in exposure assessments. If no other empiric evidence is available, expert judgment may offer the best available science to inform a model. Alternatively, when data are completely absent and the availability of expert opinion or judgment is questionable, it is possible to avoid the need for such data by model simplification (Cox, 2006; Vose, 2008). Such an approach is particularly worthwhile when empiric evidence is available to inform the probability distributions of process outputs subsequent to the process that is missing data. Such 'downstream' data (i.e., between the missing element and the ultimate exposure distribution) actually reflect the likelihood of microbe levels given the processes (modeled or missing) that occurred prior to the process. Therefore, processes for which data are missing may effectively be skipped over if data are available downstream.

Methods for eliciting expert judgments have been suggested (Kaplan, 2000). Techniques for resolving conflicting opinions among experts focus on having experts cite the experiences that inform them. In general, a diverse group of experts is preferred when eliciting input to the exposure assessment.

Scrutiny and analysis of large amounts of complex data is another use of technical experts in exposure assessments. Convening an expert panel to review and assess available evidence about a model's variables and parameters is often an effective technique for endowing the exposure assessment with credible depictions of inputs and their attendant uncertainties (ECSCC, 2003).

Ouchi (2004) and Morgan and Henrion (1990) provide summaries of methods and citations for primary literature in the field of expert elicitation. Some of the methods summarized by Ouchi (2004) and Morgan and Henrion (1990) include the following:

a) Behavioral approaches:
    1) Face-to-face interaction
    2) Delphi method
    3) Nominal group technique
b) Mathematical approaches (for probabilistic risk analysis):
    1) Non-Bayesian axiomatic models (opinion pools, performance-based weight model)
    2) Bayesian models (additive error and multiplicative error models, stochastic dependence)
    3) Psychological scaling models (Thurstone model, Bradley-Terry model, negative exponential lifetime model)

It should be noted that Morgan and Henrion (1990) observed that "because the public decision maker must often informally factor in a number of other considerations, it

is rarely of great practical consequence that a more formal treatment in the combining or weighting of alternative expert views is not possible."

An example of how EPA has used expert judgment is the Office of Air Quality Planning and Standards (OAQPS) report *An Expert Judgment Assessment of the Concentration-Response Relationship Between $PM_{2.5}$ Exposure and Mortality* (EPA, 2004a). Based on an NRC report recommendation, OAQPS used expert judgment to develop probability distributions for key sources of uncertainty regarding the mortality effects of ambient fine (>2.5 microns) particulate matter ($PM_{2.5}$) exposure. EPA's Environmental Benefits Mapping and Analysis program (BenMAP)[5] includes the exposure-response functions derived through expert judgment assessment as options for risk modeling.

EPA's Science Policy Council's Draft Expert Elicitation Task Force findings include (EPA 2009c):

a) Past experience with expert elicitation at EPA indicates that it can provide useful, credible results. NAS has highlighted these past efforts as exemplary and recommended that EPA continue in the direction established by these precedents.

b) The use of expert elicitation is appropriate for some situations; but, not for others. Factors favoring the use of expert elicitation include: inadequate information to inform a decision, lack of scientific consensus, and the need to characterize uncertainty. Factors favoring alternatives to expert elicitation include theoretical and practical limitations. Typically, an expert elicitation requires a significant investment of resources and time to provide credible results.

c) Expert elicitation can work well when a scientific problem has a body of knowledge; but, lacks a consensus interpretation. For this case, expert beliefs about the value and meaning of data can provide valuable assessments and insights. This may be the case for an emerging scientific challenge or one that depends on uncertain future events. However, when a problem has abundant relevant empirical data and relative consensus exists in the scientific community, there is probably little need to conduct an expert elicitation. At the other end of the spectrum, if data are inadequate for the experts to develop judgments, an expert elicitation may not be worthwhile.

d) Given that EPA uses other more familiar approaches to characterize uncertainty, the application and acceptance of expert elicitation at EPA will likely grow with experience. If early expert elicitation efforts are well designed and implemented, this will promote the credibility and endorsement of expert elicitation within the Agency and by external stakeholders.

e) The nature of the regulatory process (i.e., legal, political, financial, technical, and procedural considerations) will influence whether and how to conduct an expert elicitation and how to communicate and use results. Within the regulatory process, EPA can use expert elicitation to encourage transparency, credibility,

[5] http://www.epa.gov/ttnecas1/benmodels.html

objectivity (unbiased and balanced), rigor (control of heuristics and biases), and relevance to the problem of concern.

Hoffmann et al. (2007) used expert elicitation to attribute illnesses associated with one of eleven major foodborne pathogens to the consumption of one of eleven categories of food. They used responses from a large panel to create and analyze four uncertainty measures: (1) agreement among experts; (2) expert agreement with prior estimate; (3) mean individual expert uncertainty; and (4) variability in experts' individual uncertainty. Hoffman and colleague's framework shows how these measures when viewed together can provide greater insight into the state of knowledge available to support decisions, than could individual measures. They used statistical analysis to assess the quality of both expert judgment data and external data. Hoffmann and colleague's suggest that analysis of multiple uncertainty measures is likely to be particularly useful to decision makers when external validity checks that rely on conventional scientific methods or further data collection is infeasible or costly.

2.6.6 How do I Consider Information Quality Including Data Quality?

Section 515 of the Treasury and General Government Appropriations Act for Fiscal Year 2001 (Public Law 106-554, also known as the "Data Quality Act" or "Information Quality Act") directed OMB to issue government-wide guidelines that "provide policy and procedural guidance to Federal agencies for ensuring and maximizing the quality, objectivity, utility, and integrity of information (including statistical information) disseminated by Federal agencies." Federal agencies responded to the OMB guidelines (OMB, 2002) by developing Agency specific guidelines. For example, EPA published *Guidelines for Ensuring and Maximizing the Quality, Objectivity, Utility, and Integrity, of Information Disseminated by the Environmental Protection Agency* (hereafter known as EPA Information Quality (IQ) Guidelines; EPA, 2002b). EPA's IQ Guidelines[6] include the following adaptation of the quality principles found in the Safe Drinking Water Act (SDWA) Amendments of 1996 (EPA, 2002b):

a) The substance of the information is accurate, reliable and unbiased. This involves the use of:

    1) the best available science and supporting studies conducted in accordance with sound and objective scientific practices, including, when available, peer reviewed science and supporting studies; and

    2) data collected by accepted methods or best available methods (if the reliability of the method and the nature of the decision justify the use of the data).

b) The presentation of information on human health, safety, or environmental risks, consistent with the purpose of the information, is comprehensive, informative, and understandable. In a document made available to the public, EPA specifies:

[6] These principles should be adopted or adapted in all Federal agency IQ Guidelines for assessments related to evaluations and public heath (OMB, 2002).

1) each subpopulation addressed by any estimate of applicable human health risk or each risk assessment endpoint, including subpopulations if applicable, addressed by any estimate of applicable ecological risk;

2) the expected risk or central estimate of human health risk for the specific subpopulations affected or the ecological assessment endpoints, including subpopulations if applicable;

3) each appropriate upper-bound or lower-bound estimate of risk;

4) each significant uncertainty identified in the process of the assessment of risk and studies that would assist in resolving the uncertainty; and

5) peer-reviewed studies known to the administrator that support, are directly relevant to, or fail to support any estimate of risk and the methodology used to reconcile inconsistencies in the scientific data.

Discussion of data quality is an important part of planning and scoping. You can evaluate data quality within the context of your agency's data quality guidelines and work with managers to make decisions about what types of data should or should not be included based on data quality and scope. Data quality requirements may differ depending on the planned use of the risk assessment. If data are excluded, it is always important to note the exclusion and reason for the exclusion. Methods for evaluating data quality are important tools for producing a risk assessment that has both scientific value and credibility with stakeholders.

In some cases development of Data Quality Objectives may be required by Agency policy. Data Quality Objectives facilitate transparent documentation of the justification of why data were included or excluded (IRAC, 2000). The following list describes several important characteristics that may be helpful to evaluate the usefulness of data sets for risk assessment, such as submitter information, data source, methods, and confidentiality (adapted from IRAC, 2000):

a) General information: Complete name and correspondence address of principal investigator, purpose of study, and availability of raw data

b) Source of data: Funding source/affiliation of principal investigator or data collectors, who collected/produced the data, and for numerical data, provide numerator and denominator study design: type of study, sample size, sampling frame/sample selection, and how sample relates to the population (is the sample from a particular country, region or producer?)

c) Data collection: Method of data collection/compilation, age of data, country/region of origin, time frame for collection (seasonality), and conditions of collection (field versus laboratory data)

d) Microbial methods: Testing methods (which tests were run), sensitivity and specificity of test(s), techniques used, precision of measurement, definition of units being used, species of animals used, if any, and specific organism tested or studied

e) Evaluation of information: Consistency with regard to findings of other researchers, publications that cite the data, peer review of the data, investigator's evaluation of data, and investigator's recommended limitations of data

f) Protections for sharing raw data: Confidentiality for human subjects (blinded data)

There are general criteria for evaluating data to decide if it should be included in a risk assessment. Basic questions to evaluate data include the following (adapted from EPA, 1998a):

a) Are the study's objectives relevant to the risk assessment? The most relevant data for risk assessment are those that focus on the (1) organism of interest; (2) population at risk; and (3) circumstances of exposure (e.g., vehicle, level, timescale, and route).

b) Are the variables and conditions the study represents comparable with those important for the risk assessment?

c) Is the study design adequate to meet its objectives?

d) Was the study conducted properly?

e) Were there associations between observable data and the outcomes (health or otherwise) of interest?

f) Are factors that could increase or attenuate risk (risk factors) controlled for in the data?

g) How are variability and uncertainty treated in the study report?

h) Are the data sufficiently robust to be used to support a causal effect between exposure and infection or illness?

i) Does the study meet agency requirements regarding ethics, such as having passed internal review boards or complying with Agency regulations regarding research? For example, EPA's Protections for Subjects in Human Research Rule[7] requires that all pregnant women, all nursing women, and all children are excluded from all studies involving intentional exposure that are intended for submission under pesticide laws. Additional information on the conduct and use of observational studies in EPA's risk assessments are addressed in *Scientific and Ethical Approaches for Observational Exposure Studies* (EPA, 2008).

EPA also has general assessment factors, including soundness, applicability and utility, clarity and completeness, uncertainty and variability, and evaluation and review,

[7] http://www.epa.gov/oppfead1/guidance/human-test.htm

which are covered in more detail in EPA's Science Policy Council *Assessment Factors* (EPA, 2003b).

Since data are never complete and are rarely collected specifically for risk assessment; many types of data are considered for inclusion in risk assessment. Good quality data include complete datasets, relevant data, and peer-reviewed data that are considered high quality by experts in the field and agree with other data sets in terms of comparison of methods and development of tests. Complete datasets would include information on all the characteristics listed above. Relevant data may depend on the risk question that is being addressed. Some characteristics of relevant data include age of data, region or country of origin, purpose of study, and species involved.

2.6.7 What is a Value-of-Information Analysis?

During the planning and scoping discussions, a question you might ask is whether to wait for additional research, or is there enough confidence in readily available information to make a decision (Section 2.4.1). The aim of a value-of-information (VOI) analysis for the decision maker will be in its ability to determine when no more information about the risk of microbial pathogens is economically beneficial to making a decision (Disney and Peters, 2003). For example, VOI is the amount a decision maker would be willing to pay for information prior to making a decision. The maximum VOI is for complete information. Consequently, the VOI analysis can contribute to the identification of cost-effective strategies for reducing a risk to appropriate levels. VOI analysis provides a way to quantify the value of actions taken to reduce the risk associated with a decision (Hirshliefer and Riley, 1992).

Furthermore, decision makers could take a variety of actions to increase their confidence in the estimates of the probability of human illness, before making a final decision. VOI analysis could be used to identify optimal "confidence-increasing strategies" for reducing uncertainty in quantitative risk analyses estimates of the probability of an occurrence (Disney and Peters, 2003).

VOI analysis provides a set of methods for optimizing efforts and resources to gather, to process, and to apply information to help decision-makers achieve their objectives (NRC, p). NRC provides a schematic of the application of VOI analysis to assess the impacts of additional studies in a specific decision context. Information opportunities that address uncertainties in the baseline model are considered with respect to the changes they would have on the decision-maker's preferred decision option and the associated change in net benefits. The analysis may also consider any direct costs (for example, financial) and indirect costs (for example, the health or economic impacts of delayed decision-making) associated with the information opportunity. The valuation of information is ultimately driven by the decision-maker's values with respect to the distribution of risks and costs, including any costs associated with delayed decisions (NRC, 2009).

2.6.8 What is a Communications Plan?

Risk communication (Chapter 8) can be initiated as soon as the risk assessment process begins and can be incorporated throughout the process; risk communication should not be an afterthought. During the planning and scoping discussions, the risk communication specialists need to work with risk assessors, risk managers, and appropriate stakeholders to develop a communication plan or strategy to be used in all dealings with the public (essentially all stakeholders). In the plan you should consider:

a) Clearly identify stakeholders and collaborators (e.g., in multiple partnering projects).

b) Set forth specific communication goals (e.g., what will be your proposed message(s)).

c) Establish who will be the audiences for whom you will communicate throughout the risk assessment process. The audience can range from technically sophisticated risk assessors and knowledgeable risk managers to an educated lay audience with limited knowledge of risk assessment.

d) Provide for the development of the content of any communication tailored toward your projected audiences.

e) Develop proposed outreach avenues to your audiences (e.g., fact sheets, press releases, newsletters, notices, open meetings, briefings, emails, websites).

You will most likely rely upon communication specialists, including your agency's public affairs office, to get the message across. It is advisable that these specialists be involved with the risk assessment effort from planning and scoping onward. A principal spokesperson can be identified (possibly the risk assessor), and you can decide which information channel(s) should be used. Your agency may have guidance for risk communication plans or stakeholder involvement. For further information regarding risk communication from an assessor's point of view, refer to Chapter 8.

2.7 What Types of Decisions within Risk Assessment are Science Policy?

This section discusses decisions made within the context of design and data usage for risk assessment. It does not address the types of policy or regulatory decision making that occurs after the results of the risk assessment have been considered. Science policy decisions are usually differentiated from scientific judgment calls. Whereas risk assessors can make decisions based on scientific judgment, if a decision goes beyond what would reasonably be considered firmly supported by science, then policy comes into play. Once policy is involved, then risk managers need to become engaged in the decision-making. Failing to distinguish between policy decisions and scientific judgment in a risk assessment is a serious threat to the scientific credibility of the assessment. It is important to note that:

a) The utilization of science policy in the risk assessment process is not meant to "bury" or "hide" risk management decisions within the risk assessment (i.e., any science policy position or choice used in the risk assessment process does not direct the risk assessment itself toward a specific risk management decision).

b) To be transparent, policy choices need to be stated explicitly in the risk assessment.

c) Although science policy is utilized in the risk assessment process, it is important to recognize that the policy positions themselves are developed outside the risk assessment.

d) Scientific data should support science policy positions and risk assessors and risk managers should ensure that the risk assessment proceeds in a way that best serves the informational needs for subsequent decision(s) based on the risk assessment.

Science policy positions and choices are by necessity utilized during the risk assessment process. You will likely use science policy positions in two major ways in the risk assessment.

First, there are some basic, fundamental science policy positions that frame the risk assessment process to ensure that the risk assessments produced are appropriate for a particular decision. These scoping "boundaries" for the risk assessment are articulated during the planning and scoping process and ultimately explained clearly in the risk characterization (i.e., what will be addressed in the risk assessment for decision purposes, but also just as importantly, what will not be addressed and why [e.g., not pertinent to the decision needed]). These science policy positions not only shape the risk assessment process, but are usually a factor in the decision making process outside the risk assessment.

Second, the use of default assumptions in a risk assessment is a science policy choice often invoked when there is a lack of data. These science policy choices are more specific than the framing science policies mentioned above. Given the nature of uncertainty and data gaps, default assumptions (sometimes simply called defaults) are often used to address these uncertainties when data are unavailable or otherwise not suitable for use. A default assumption is the option chosen on the basis of a science policy choice that appears to be the best choice in the absence of data to the contrary (NRC, 1983). The NRC, in its review of risk assessment practices in *Science and Judgment in Risk Assessment* (NRC, 1994), acknowledged that default assumptions are needed to address uncertainty arising from a lack of data and other information. The report also stated that Agencies should have principles for choosing default options.

When pathogen-specific data are unavailable (i.e., when there are data gaps) or insufficient to estimate parameters or resolve paradigms, a default can be used in order

to continue with the risk assessment. This is a science policy choice, generally agreed upon during the planning and scoping discussions, when data gaps are identified (see Section 2.4.1 for information on data gaps). During the risk assessment itself, a default is used only when essential data are lacking. Point estimates can also be considered defaults, when the distribution of the parameter only adds unnecessary complexity given the needs of the risk assessment. For example, drinking water consumption is often modeled probabilistically for MRAs with a median of 1L per day. The consumption value of 2 L/day per person is often used for chemicals and represents the 90th percentile of the 1994 to 1996 and 1998 Continuing Survey of Food Intake by Individuals community drinking water consumption data. As illustrated in this example, the choices you make need to be well within the range of plausible outcomes and often at specific percentiles (for variability) within that range of observation. The use of 1L versus 2L/day is not related to differences in microbial versus chemical risk assessment.

The default assumptions are not pathogen-specific *per se*, but are relevant to the data gap in the risk assessment. Defaults are based on published studies, empirical observations, extrapolation from related observations, and/or scientific theory. A representative list of the areas where assumptions are commonly made in MRAs is presented in Appendix A.

2.8 What is My Role as a Risk Assessor in Risk Assessment?

People who perform the risk assessment, in whole or for the most part, are the risk assessors. One often overlooked responsibility as a risk assessor is to communicate your key risk findings and conclusions and your confidence in them. Most of these responsibilities are noted in other sections of this Guideline. Specific responsibilities often include:

a) Collect, compile, and document appropriate data

b) Let the risk manager know whether the key data used for the assessment are considered experimental, state-of-the art or generally accepted scientific knowledge

c) Describe quantitative risk estimates in plain English; the use of tables and graphics may be helpful as a supplement

d) Describe the uncertainties inherent in the risk assessment and the default positions used to address these uncertainties or gaps in the assessment

e) Refer the reader to an Agency risk assessment guideline or other easily obtainable reference that explains terminology (e.g., how a microbial criterion was developed)

f) Determine what types and models are appropriate for the particular risk assessment

g) Input data and models into software

h) Run modules

i) Conduct sensitivity analyses

j) Identify the subpopulations or systems addressed and provide an explanation for the selection of these as the "valued entities."

k) Describe the level of confidence associated with the conclusions and assumptions and defaults

l) Summarize and identify the key pieces of information critical to your evaluation

m) Put this risk assessment into a context with other similar risks that are available to you and describe how the risk estimated for this pathogen or agent compares to others regulated by your agency

n) Describe how the strengths and weaknesses of the assessment compare with other assessments prepared in the past

o) Describe the rationale and basis for the conclusions drawn by those outside your agency about this pathogen or agent

p) If their conclusions differ from yours, let the manager know whether theirs is a reasonable alternative; can their conclusions reasonably be derived from the data set

q) Inform the risk manager of the strengths and weaknesses of their evaluations compared to yours

r) If you have developed specific assessments for one or more risk management options, let the risk manager know what changes in risk would occur under these various candidate risk management options

s) Highlight key or critical issues and, if needed, identify a hierarchy of issues based on importance to the assessment or those that could influence decisions makers.

t) Oversee and manage contractor support on the risk assessment, including review of documentation and calculations for mathematical errors

u) Keep the decision maker informed of the status of your risk assessment and risk characterization

v) Archive the risk assessment and associated materials in a manner consistent with your organization's archiving procedures

3. HAZARD IDENTIFICATION AND HAZARD CHARACTERIZATION

Hazard identification and hazard characterization (HI/HC) are key components of risk assessment.[8] In HI, the suspect hazard (i.e., the microorganism/adverse effect) is identified and defined in the context of epidemiological, surveillance, clinical, microbial (agent specific), and environmental (including meteorological and geographical) information available to the risk assessor. The HC focuses on a particular microorganism(s) and potential or known mechanisms of host-pathogen interaction, virulence, and pathogenicity. As discussed in Section 1.6, the epidemiological triangle is a useful framework for conceptualizing HC. Meteorological and geographic conditions impact the persistence and transmissibility of microbial agents in the environment and influence the level of potential exposure. This chapter presents information that is basic to HI/HC. A list of questions that may be posed during HI/HC is presented in Appendix B.

3.1 How do I Define the Hazard?

The term hazard broadly refers to the subject of an assessment. It can be interpreted in a number of ways. It may be defined as the stressor agent capable of causing an adverse effect on the exposed individual or be defined as the adverse effect itself. The subject of the risk assessment (hazard of interest) is a policy decision driven by the existing statutes, regulations, or consistency with agency processes. You should be aware that the term hazard may refer to an agent that is causal or associated with adverse outcome, the mechanism and metabolic products leading to the outcome, or simply the subject of an assessment wherein the likelihood for adverse outcomes is being explored. The adverse effects in humans that result from exposure to microbial agents or metabolites under favorable host (healthy or susceptible due to certain life-stages/pre-existing conditions) and environmental conditions may occur soon after exposure or arise at a substantially later time (sequelae). This guideline focuses on pathogenic infectious disease hazards.

Terms such as "agent" and "stressor" may sometimes be used synonymously with hazard.[9] The microbial Thesaurus developed by EPA (EPA, 2007b) differentiates the terms as follows: The term "'stressor' is used in ecological risk assessment and includes but is not limited to the connotation that the adverse response can be the result of a lack of something – such as a habitat – which would be called a 'stressor'. The term 'agent' does not have this connotation. 'Agent' is used to denote a causative entity that actually

[8] As noted previously, in the 1983 NRC Report, Hazard Characterization is the qualitative part of dose-response assessment. In this Guideline, because of the qualitative nature of Hazard Characterization it is considered with Hazard Identification, which is also qualitative. Risk assessors can still organize the risk assessment documentation under the headings provided by NRC or WHO.

[9] Epidemiologists may use the term "agent" slightly differently. Agent - a factor (e.g., a microorganism or chemical substance) or form of energy whose presence, excessive presence, or in the case of deficiency diseases, relative absence is essential for the occurrence of a disease or other adverse health outcome. http://www.cdc.gov/excite/library/glossary.htm

physically exists as part of the environment and can be used in either ecological or human health risk assessment. 'Hazard' is used primarily in human health risk assessment, although 'hazards' are not limited to 'agents.' For example, the number of days spent in a hospital may be a hazard that correlates with risk of nosocomial infection.

In the context of MRA for frank pathogens, the term hazard represents the pathogen's potential to generally cause adverse effects in normally healthy humans, while in the case of opportunistic microorganisms, the term hazard refers to potential to cause adverse human outcomes under certain environmental and host conditions, most often when the host is compromised. Thus, hazard may be used to denote the subject of the assessment that needs further identification and characterization.

If the hazard is a pathogenic microorganism, then identification of the microorganism is an important aspect of the hazard description. Microorganisms can be categorized (defined) based on the methods used to detect them. Nucleic acid-based assays may result in a different categorization than culture- based assays. As mentioned previously, quasi-species are particularly hard to define (Section 1.5). It is important to note that in some situations even small deoxyribonucleic acid (DNA) sequence changes may elicit significantly different adverse outcomes in humans (Battista and Earl, 2004). But in the case of "zero tolerance" organisms, this distinction does not matter. For examples of the nuances of microbial nomenclature, refer to Section 3.8.

In general, when performing assessments on individual strains or isolates, identification rather than taxonomy or nomenclature becomes the issue. In this case, identification refers to the placement of an isolate within an existing taxon, or determining that it does not match any existing taxon, (i.e., proper labeling of the isolate), as opposed to the creation of a new group or groups to which the isolate is related, that share attributes with the isolate (i.e., classification of the isolate). The purpose of identification is to ensure that you know which taxon or subtaxon you are evaluating.

Microbial identification is often anything but a trivial exercise. Unless an isolate has been the subject of a taxonomic study, or is one of the cultures used to establish a commercial identification method database that is kept current, it is often difficult to ensure that an isolate belongs unequivocally to a specific taxon. In MRA, to develop accurate exposure assessment and resulting risk characterization, you should carefully evaluate the definition of the hazard and pay close attention to how different data sets used in the risk assessment define the hazard.

3.2 What are Hazard Identification and Hazard Characterization?

Hazard identification and hazard characterization provide a *qualitative* examination of the hazard identified. The *quantitative* relationship between hazard and effect is examined in the dose response assessment (see Chapter 4). Hazard identification provides the framework for gathering relevant information to construct a realistic scenario focusing on the likely microbial hazards present, so that they can be appropriately considered for the MRA. In HI, information related to the epidemiological,

surveillance, clinical, and microbial aspects of the hazard is reviewed as a critical part of the assessment. Hazard characterization helps you describe the mechanisms involved in causing harm and the microorganism's ability or potential to cause harmful effects.

You should be aware that the extent of available data for HI, HC, and exposure assessment vary greatly in MRAs. Coverage may vary depending on the type of the assessment (e.g., qualitative, quantitative, retrospective, prospective) and agency policy or convention. You should consider the extent of coverage and potential overlaps in the problem formulation step during planning and scoping. In this Guideline, HC and some elements of exposure assessment are included in the HI/HC chapter.

If your Agency uses the WHO/FAO framework (Codex 1999) the preferred chapter heading for topics discussed in this Guideline (Chapter 3) is Hazard Identification and the qualitative descriptions related to dose-response and quantitative aspects of dose-response are in a chapter titled Hazard Characterization.

3.3 What Hazard Characteristics Can I Consider?

There are several hazard characteristics that you can consider when assessing a microorganism or its by-products (adapted from USACHPPM, 2009 and EPA 2009a):

a) **Infectivity** – the ability of a pathogen to enter, survive, and multiply (infect) in a host

b) **Invasiveness** – the ability to degrade and migrate through the extracellular matrix

c) **Virulence** – the ability of the pathogen to defeat the host defenses, increase the severity and longevity of the symptoms

d) **Pathogenicity** – the ability to cause a disease state. It is the cumulative effect of virulence and invasiveness (see the glossary for a discussion of pathogenicity versus virulence)

e) **Host range** – which hosts a pathogen can infect. Some pathogens have very specific host ranges; therefore the disease is limited to one host. Other pathogens have wide host ranges, and they can cause disease in many species

f) **Horizontal gene transfer** - the movement (transfer) of genetic material (e.g., DNA) from one organism taxon to another that, with maintenance and expression of that genetic material, may lead to antibiotic resistance traits or acquisition of other virulence factors

g) **Genetic drift –** changes in the frequency of alleles in a population due to random sampling

h) **Replication** – the ability for a microorganism to multiply within the environment or the host

i) **Persistence –** the ability of the microorganism to survive in

the environment or the host

j) **Transmissibility –** the ability of a microorganism to survive, replicate, and pass through animate or inanimate matrices and stay infective

k) **Opportunistic Pathogens** – the ability of a usually innocuous microorganism to cause an adverse health effect in a susceptible host

l) **Taxonomy and Strain –** definition of the hazard with respect to traditional biological classification. Taxonomy and strain variation have a potentially large impact on risk assessment. The difference in dose-response range between isolates (and strains) can be orders of magnitude. Some strains may not be infective for humans. In addition, the ratio of different strains in the environment can fluctuate

m) **Resistance to control or treatment processes** – the ability of a pathogen to survive treatments, such as chlorination. If the risk assessment is for a performance target then the treatment and control processes may be of central importance

Genomics, proteomics, and metabonomics may all be important for hazard characterization as well as host characterization (EPA, 2006b).

3.4 How do Microbial Hazards Cause Adverse Outcomes?

The relationship between a host and a microorganism is complex with dynamic interplay; there are a number of mechanisms by which a pathogen can induce an illness. In general, these include either direct invasion of the host cells and colonization of a specific tissue or organ, causing necrosis or other direct damage or triggering host responses that are self damaging, or through the production of toxic by-products that can elicit an adverse effect through toxicological modes of action. Note that toxins may be produced by microbes in the environment or directly in the host. The conditions under which microbes produce toxins and how host exposure occurs is important to consider. For example hazardous algae blooms may result in oral or dermal exposure to toxins produced by algae in the environment, whereas, pathogenic *E. coli* produces toxins during infection which can continue to cause damage to the host even after the microbes die.

Among pathogenic organisms, there are several common patterns or themes in the cascade of events that dictate the progression of disease process. The first essential step in the establishment of a disease is the ability of the pathogen to adhere to a tissue. This step, while prompted by the pathogen, is often the result of a host-microorganism interaction that is host specific. The second step is the invasion/penetration of the host's epithelium, whether the skin, lining of the lungs, or the lining of the gastrointestinal tract. The pathogens are able to invade the host cell and establish a niche where they can multiply. The success of a pathogen to initiate and cause disease is limited by its ability to counteract effectively the host defense mechanism and be able to multiply to a level that elicits a symptomatic response. The ability of the microorganism to effectively

defeat or evade the host defense response determines the latency period, intensity, and persistence of the disease state.

Modeling mechanisms of infection (e.g. how molecular and cellular host and pathogen factors interact) may someday be applicable to MRA; however, currently the science is not developed enough for the pathogens of concern and it is unclear how much value this feature would add, given the large uncertainties in other areas of MRA.

3.4.1 What does Virulence and/or Pathogenicity Mean in the Context of Causing an Adverse Outcome?

Pathogenic microorganisms have virulence factors with specific modes of action for entry, colonization, and adverse health effects.[10] The first step in assessing pathogenicity is to collect the microbial evidence for the adverse health effects associated with the agent of concern. To cause disease, pathogens must overcome various host defense systems, and their ability to do this is indicative of the virulence of the microorganism. Examples of some types of microbial virulence factors include:

a) Factors that help the microorganism persist in the environment

b) Factors that help the microorganism evade the host immune system

c) Expression of surface proteins or polysaccharides that help bind the organism to a specific site in the host

d) Production of toxins

In causing a disease, not only do the pathogenicity and virulence potentials of the microbial agent play a role, but also the degree of susceptibility of the host and the influence of environmental factors on exposure influence the final outcome. Understanding the interactions between a microbial pathogen, the host, and the environment is key in determining the potential health impact a pathogen will have on an individual (or a population). The classic epidemiological triangle (disease triad) illustrates the inter-relationship between the host, pathogen, and environment (See Figure 1.2).

3.5 What are the Mechanisms that May Lead to the Development of New Pathogens or Pathogens with New Traits?

In the microbial world, there exist mechanisms that consistently produce newer strains of pathogens or to have existing pathogens acquire more virulent traits from other microorganisms. Such mechanisms can result in the horizontal transfer of genes within and between viral and bacterial strains. While horizontal transfer of genes often results in

[10] Pathogenicity is the quality or state of being pathogenic, the potential ability to produce disease. Virulence is the disease producing power of an organism, the degree of pathogenicity within a group or species.

reductions in fitness, on some occasions the transfer results in more potent viruses and other pathogens. Recent advances in whole genome nucleotide sequence analysis demonstrate that viral, bacterial and protozoan pathogen evolution includes horizontal gene transfer of virulence factors between different species and high taxa. Thus an understanding of the role of horizontal gene transfer between different pathogens is essential for the evaluation of the possible introduction of new microbial hazards. This may be as a result of an unintentional or deliberate environmental release of natural or genetically modified microorganisms.[11]

Acquisition of new traits comes about by the transfer of genetic traits vertically or horizontally among microbial species. All living organisms have at least one natural mechanism for genetic transfer. Techniques of biotechnology take advantage of these mechanisms to precisely transfer desired characteristics or remove undesirable ones in genetically modified bacteria. In prokaryotes, these mechanisms include: 1) conjugation, through which portions of genetic materials are exchanged between two related cells in physical contact; 2) transduction which occurs through infection by a virus intermediate, a bacteriophage; and 3) transformation in which there is direct uptake and incorporation of extracellular DNA. Facilitating these transfers are genes for mobilization of DNA from one genomic compartment to another, as from a large replicon (chromosome) to a smaller one (plasmid). These are often found in insertion elements and transposons.

It is commonly recognized that mobile genetic elements have contributed to rapid changes in virulence potential by the acquisition of new traits that increase their survival and adaptation in human hosts and in adverse environmental conditions. When mobilizing genomic elements (phages, plasmids, insertion elements, or transposons) acquire such functional gene segments, selection can segregate these into self-transmissible units, called 'genomic islands'. When specific traits or virulence factors that contribute to pathogenicity are included within these units, they are called pathogenicity islands (Knapp et al., 1986; Schmidt et al., 2004). The advent of whole genome sequencing and other advances in molecular biology has allowed development of criteria for recognizing pathogenicity islands in microorganisms of interest (Guzman et al., 2008; Yoon et al., 2007; Dobrindt et al., 2004).[12] In some cases, where the suspect microorganism is known to be related to ones that have been sequenced, the use of sequence analyses can be employed to look for components of pathogenicity islands or virulence factors.[13] As indicated in section 3.4.1, bacterial pathogenicity determinants are generally grouped as virulence factors or mechanisms which includes antibiotic resistance, pore-forming toxins, superantigens (Schmidt et al., 2004)[14] and even quorum sensing (Lerat and Moran, 2004). Thus the knowledge of mechanisms that have the potential to result in microorganisms with new pathogenic traits may be of critical importance in conducting certain types of risk assessments.

[11] Genetically modified microorganisms are a potential topic for a future volume of this Guideline.
[12] http://www.gem.re.kr/paidb/about_paidb.php
[13] http://www.ncbi.nlm.nih.gov/PMGifs/Genomes/micr.html; http://www.genomesonline.org/; http://www.tigr.org; http://www.sanger.ac.uk/
[14] http://www.gem.re.kr/paidb/about_paidb.php

3.6 What are the Major Categories of Microorganisms?

The major microbial categories that cause adverse outcomes to humans include bacteria, fungi, viruses, protozoan parasites, algae, and indeterminate types (Table 3.1). Helminthes (tapeworms, roundworms) are also considered to be hazardous organisms, particularly if direct exposure to feces is possible. Although helminthes are multicellular parasites and not microorganisms, they are sometimes considered in conjunction with pathogens because infectious stages are too small to be easily detected by the naked eye.

There is a vast array of microorganisms and associated literature on pathogenic genera, species, subspecies, strain and subtypes, which is outside the scope of this document. There is vast literature available on taxonomic characterization of microbial agents. However, depending on the specific requirement of an assessment, it is recommended that an assessor consult relevant literature and subject matter experts as needed. Under some circumstances the hazard may not be identifiable however the human health effects may be distinct. These are hazardous agents of indeterminate type but may still be clinically well defined enough to facilitate risk assessment approaches.

Table 3.1 presents some of the major categories of microbial hazards in the context of MRAs (Labbe and Garcia, 2001; Peter, 1998; Alexopoulos et al., 1996). The broad categorization of microbial organisms should help you understand how an agent in a given category causes disease in humans. The placement of hazardous organisms into broad categories is particularly important in retrospective assessments to narrow the focus of investigation based on documented history for the category in question.

Batz et al. (2004) constructed a comprehensive list of pathogens for the Foodborne Illness Risk Ranking Model (FIRRM) analytical software tool using data generated by various federal agencies. This includes estimates of the incidence of foodborne illness by CDC as reported by Mead et al. (1999) and cost-of-illness studies by the USDA's Economic Research Service, with additional data reported by FDA, USDA FSIS, and Center for Science in the Public Interest. An additional list of foodborne pathogenic organisms and toxins compiled by FDA commonly called Bad Bug Book may also be useful (CFSAN, 2006). A review of waterborne pathogens is provided by Craun et al. (2010).

EPA's Water Quality Criteria program,[15] which addresses microbial contamination of the nation's waters under the Safe Drinking Water Act (SDWA) and the Clean Water Acts (CWA), provides information on microbial methods, Health Advisories, Regulatory Support and Criteria Documents. Health Advisories serve as informal technical guidance to assist federal, state, and local officials responsible for protecting public health when emergency spills or contamination situations occur. Criteria documents and guidance for drinking water contaminants provide information so preliminary decisions can be made as to whether the contaminant is a significant health threat via drinking water exposure and whether sufficient data exists to perform quantitative risk assessments.

[15] http://www.epa.gov/waterscience/criteria/humanhealth/microbial/

EPA's Candidate Contaminant List of microbial organisms (CCL3)[16] is a list of twelve microbial agents currently not subject to any regulation based on a contaminant's potential to occur in public water systems and the potential for public health concern. The list of chemical contaminants includes cyanotoxins produced and released by cyanobacteria ("blue-green algae").

[16] http://www.epa.gov/ogwdw000/ccl/ccl3.html#microbial

1 **Table 3.1 Major Categories of Foodborne and Waterborne Microorganisms**

Category	Features				
	Examples	**Morphological**	**Physiological**	**Genetic**	**Pathogenicity Adaptation Mechanisms**
Bacteria	*Francisella tularensis, Brucella suis, Escherichia coli* O157:H7, *Salmonella, Campylobacter, Listeria, Legionella* Cyanobacteria, *Vibrio*	Single-celled prokaryotes	Metabolically diverse, invasive, produce intra/extracellular toxins	No nucleus, double stranded DNA, presence of extra chromosomal DNA/Plasmids, mutate frequently, horizontal gene transfer mechanisms	Some species form spores to withstand adverse conditions. Mutation and gene transfer, pathogenicity islands and other genetic traits/mechanisms lead to frequent strain variation, acquisition of enhanced virulence traits and adaptation to new environments , toxin production
Viruses	Noroviruses, Adenoviruses, Hepatitis A	Acellular, most are enveloped with geometric structures	Metabolically inactive, obligatory parasitic, host dependent	Single or double stranded RNA or DNA, mutate rapidly in host	Frequent genetic drift, shift and other genetic mechanisms may lead to changes in antigenic properties, host survival/adaptation and result in more virulent variants/strains
Protozoa	*Toxoplasma gondii, Giardia spp., Cryptosporidium hominis, Cryptosporidium parvum, Naegleria spp.*	Single celled Eukaryotes of the Protista, display different morphologic structures and stages of infectivity	Host dependent parasites	Nucleus present, not known to mutate as frequently as bacteria and viruses	Cyst and spore formed to withstand adverse conditions. Relatively stable genome, however, mutation and gene transfer may lead to strain variation, enhanced virulence and adaptation to new environment
Fungi	*Aspergillus fumigatus, Penicillium, Candida, Aspergillus flavus*	Eukaryote Mostly multicellular and filamentous, pathogenic fungi are mostly unicellular (e.g., yeasts)	Metabolically diverse, invasive, produce mycotoxins	Nucleus present, sometimes presence of extra chromosomal DNA/Plasmids	Spores
Algae Chlorophyta, Rhodophyta Dinoflagellata	*Pfiesteria piscicid*, "red tide" *Gambierdiscus toxicus (Ciguatera)*	Single-celled photosynthetic organisms, often dinoflagellates, Eukaryotes	Metabolically diverse highly complex life cycle, a few toxin producing	Nucleus present	Three typical forms are classified as amoeboid, flagellated and encysted varieties
Indeterminate agent*	Can vary, unknown	Can vary, unknown	Can vary	Can vary, unknown	Can vary, unknown

2 * The vehicle exposure/pathway may be important as the agent is indeterminate

3.7 What Methodological Approaches can be Used to Identify and Quantify Microorganisms?

You should be familiar with laboratory approaches for identifying and quantifying the microorganism(s) of concern. For any datasets that required laboratory methods that were used in risk assessment, carefully review issues related to sensitivity, specificity, limit of detection, sampling method, and sample size. Extensive reviews of microbial methods are available (CFSAN, 2001; USDA 2008; AOAC International 2007).

Identifying an unknown microorganism is a two-step process, requiring methods to characterize the traits of an organism and approaches to interpret the characterization data. Methods to generate characterization data range from traditional culture-based phenotypic and biochemical tests to the more recently developed molecular techniques. Approaches can reflect the evolutionary inheritance of traits (e.g., lineal decent), the intrinsic properties of the organism regardless of how they were acquired, or a combination of both.

Methods used for identification and quantification are often related to similar ones used for classification of microbes. However, the different purposes of classification and identification require separate considerations, even when the same technology is utilized for both. Under the controlled conditions of a classification study a particular methodology may be exquisite in its ability to distinguish among selected related isolates, but that same method may be only able to provide an approximate identification when one encounters an isolate outside the lab without the benefit of closely related isolates available for comparison.

While traditional culturing methods for identification and quantification of microbes are still the mainstay for fecal indicators, it is clear that this approach by itself does not allow for complete evaluation of microbial organisms beyond the genus and species level, and does not detect strains that may be active/infectious but non-culturable. The classical culture-based approach relies on culture of the organism in question, isolation in pure culture, and a study of the morphological, biochemical, physiological and other traits. However, not all microorganisms are culturable (see viable-but-not-culturable discussion in Section 3.8).

More sophisticated methods have the ability to discriminate subtypes and also capture information on pathogenicity determinants of interest at the genotypic and phenotypic levels, either with isolated cultures or even in mixed enrichments. Some molecular methods can be used even in the screening steps early on, to capture the virulence potential of a specific microorganism or multiple microorganisms (profiles) without the isolation steps that are generally less sensitive and more time-consuming.

3.8 Are there any Special Concerns Regarding Microbial Detection Methods?

Purpose of risk assessment and choice of the method(s)

Discrimination down to the smallest organizational level (i.e., strain) is not necessarily the objective of all identifications done for risk assessments. You should determine what information is critical for completing the assessment before pursuing a specific level of identification. If the pathogenic potential is associated with all members of some higher order taxa, then identification to that higher taxonomic level may be all that is required for broad-based assessments. For example, knowledge that an isolate is a member of a species complex with many shared characteristics may be sufficient to permit assessment of potential for pathogenic effects, such as the *Mycobacterium avium* Complex (MAC). However, if evaluation of a specific incident is the purpose of an assessment, it is important to note that even the lowest levels of taxon (SNP variant) are known to elicit significant adverse outcomes. For examples, an enterohemorrhagic type of *Escherichia coli* known as serotype O157:H7, can cause serious hemolytic uremic syndrome and even death. It is known that the degree of adverse outcomes seen with *E. coli* O157:H7 infections vary distinctly among different clades (group of SNP subtypes) (Manning et al., 2008). Thus, in retrospective assessments you should pay attention to detailed information within a taxonomic subunit.

When doing assessments for broad categories of microorganisms, genus-level identifications may be all that is needed. Culturing techniques that involve phenotypic analyses may suffice. However, for this level of identification, 16S rDNA analyses have become commonplace and are usually deemed adequate for prokaryotes. Morphological features are traditionally used for fungi and protozoa, but biochemical and molecular methods are beginning to be essential to avoid miss-identifications.

Culture related issues

Many non-spore-forming bacteria exposed to environmental stress conditions may decline in number and not be detectable by traditional culture based laboratory methods, depending on the level of detection for each method and the number of replicate cultures employed. To ensure that microorganisms in the viable-but-nonculturable (VBNC) state are not missed, you should be aware of relevant methods of direct identification such as molecular methodologies[17] for detection and direct microscopy. For microorganisms present at low levels, specific enrichment cultures can be employed that allow growth of very low population numbers to levels high enough for traditional culture techniques. A VNBC state has been described where metabolic characteristics are quantifiable though cells cannot be grown on traditional culture media, most notably in the genus *Vibrio*, under conditions when water temperatures drop below 10°C (Smith and Oliver, 2006;

[17] There are methods to isolate the DNA from environmental samples that do not require a pre-enrichment culture, such as length heterogeneity-polymerase chain reaction (LH-PCR). Methods such as LH-PCR require as little as 10 ng of DNA in the PCR reaction to amplify 16S rRNA sequences (Bisson, et al., 2007). In addition, PCR has been used to detect antibiotic resistance genes in bacteria found in deli-meats (Li and Wang, 2010).

Fischer-Le Saux et al., 2002; Rowman, 2004; Huq et al., 2000). Others have shown that under such stress conditions very small numbers of surviving cells are able to grow to higher levels using dead cell materials remaining in the stressed culture, therefore giving the appearance that the cell state had changed from nonculturable back to culturable (Bogosian et al., 1998). Molecular methods generally detect living and non-living pathogens, unless specific methods have been developed that target aspects of their growth viability (Sen and Ashbolt, 2011). However, some important pathogens, such as Norovirus, cannot be reliably grown in cell culture, so there is currently no way to check on the infectivity of environmentally recovered virions of Norovirus.

Similarly in the case of parasites and viruses that are nonculturable and do not replicate outside the host, serological or advanced molecular methods have been routinely utilized in identification and reporting. Other methods of detection may also be needed where pre-formed metabolites/toxins may be present but the suspect pathogen cannot be recovered, for example, emetic toxin for *Bacillus cereus* or endotoxins in aerosols.

Amplification of organisms by growing in culture can also exert selective pressure, which can skew identification of microbes from environmental or clinical samples. You should be aware of any methods related considerations that might impact your judgment of the relevance or quality of a particular study.

Level of discriminating power

The two major subtyping approaches commonly used are based on our ability to discriminate phenotypic or genotypic traits. The phenotypic approach includes serotyping, phage typing, multilocus enzyme electrophoresis, and esterase typing. The genetic subtyping approach encompasses pulsed field gel electrophoresis (PFGE), ribotyping, PCR-based subtyping techniques (e.g., random amplification of polymorphic DNA [RAPD]), amplified fragment length polymorphism (AFLP), PCR-restriction fragment length polymorphism (PCR-RFLP), repetitive element PCR (REP-PCR), DNA sequencing-based subtyping techniques (e.g., multilocus sequence typing [MLST]) (Liu, 2006), multilocus genotype typing (MLGT), and single nucleotide polymorphisms (SNPs). Some of these molecular based subtyping techniques provide not only powerful discriminating capabilities to further identify a unit of potential hazard in question, but also to relate microbial traits to public health outcomes and perhaps most importantly, enables subtype-based surveillance to detect outbreaks.

Depending on the pathogen/unit of hazard of interest, pay close attention to the discriminating power of the methods used and level of details needed in risk assessment based on the risk management needs. For example, there are about 3000 serotypes within the species *Salmonella enterica* with differing levels of human pathogenicity. Subtyping strategies include serotyping and further determination of the virulence traits important to public health. For example, subtyping *S. enterica* serotype Typhimurium definitive type 104 (DT104) involves identifying isolates through serotyping, followed by antimicrobial susceptibility testing to identify the R-type ACSSuT (ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline) antibiotic resistance pattern. *S. enterica*

serotype Typhimurium isolates with R-type ACSSuT are subsequently phage typed to specifically identify DT104 (Akkina et al., 1999).

In the case of the bacterial human pathogen *Listeria monocytogenes,* which is made up of a range of strains/genotypes with varying degree of pathogenicity, some strains may be highly pathogenic and sometimes deadly; others may be relatively less virulent and cause little harm in the host. The ability to differentiate strains of *L. monocytogenes* is particularly important for microbial hazard identification and for tracking transmission of pathogenic strains within a given environment via food processing environments. The purpose of subtyping is to link human infections that may be related, detecting unusual clusters of human disease, and determining the source of exposure thorough epidemiological investigation, as well as finding and controlling the source of contamination. The application of molecular techniques has facilitated the identification and characterization of major virulence-associated genes and proteins in *L. monocytogenes*. Various DNA fragment-based typing methods have been used to differentiate *L. monocytogenes* strains at the subspecies level to include epidemic clones, genotypes, lineage types, and serotypes.

Differences in methodological approaches and choice of method(s)

Phenotypic methods include techniques that directly or indirectly detect, measure, or characterize features of a microorganism resulting from the observable expression of its (total) genetic constitution. Phenotypic characteristics of bacteria include morphological, physiological, and biochemical features. Methods for characterizing phenotype require growth of the microorganism in pure culture under appropriate conditions. Chemotaxonomic methods examine phenotype by using quantitative analysis of the organism's chemical constituents. Genotypic methods directly compare nucleic acid sequences, rather than rely on gene expression.

Approaches to data interpretation can be determinative, numeric taxonomic, or phylogenetic. Sometimes neither genotypic nor phenotypic methods alone suffice for either classification or identification of some bacteria, but it may be possible to combine these methods using polyphasic taxonomy in which data from phenotypic, chemotaxonomic (e.g., cell-envelope lipids, electrophoresis patterns), and molecular methods (e.g., ribosomal deoxyribonucleic acid [rDNA], DNA gyrase, subunit B [gyrB]) are combined. When results from these methods all agree, you can usually rely on the outcome. However, conclusive data may not be always available for a microorganism of interest.

Other considerations for identification

The proper use of methodology needed to identify (classify) the subject of an assessment depends on the level of detail required, whether prospective or retrospective, and cannot be over emphasized. When the hazard units are whole taxa, you need to consider the historical perspective and the current state of taxonomy and nomenclature for that unit to ensure that relevant data and information are compiled. Scientific and

technological advances make the field of microbial classification dynamic in that the taxonomic and other ways of classification are continually improving our understanding of the unit of infectious structure (e.g., genus, species, subtypes, SNPs), their risk potential, and mechanisms of pathogenicity. Phylogenetic analyses of microorganisms have resulted in frequent re-assignment of microorganisms into different genera or species, so synonyms for the microorganism may need to be tracked. Additionally, the evolving nature of microorganisms enables them to acquire newer traits for pathogenicity, host range, specificity, adaptability, and survivability outside of and within the host that add to the complexity of HI.

You should consider defining the unit of hazard in the context of the evolving taxonomic information and risk management needs. For example, the term *Burkholderia cepacia* had been applied both to a single species and groups of strains, termed genomovars (Mahenthiralingam, 2000; Vandamme et al., 1997, 2000). These groups of strains have subsequently acquired species status, with independent names being established. The latter have been called, and remain, the *Burkholderia cepacia complex* (Bcc). *Burkholderia cepacia* is now construed as a single species within the Bcc, but isolates of other species in the complex have, at earlier times, been called *B. cepacia*, with attendant literature using only that epithet. However, at times the unit of hazard may not be identifiable as a structural taxonomic unit. You should become aware of such issues to be able to refine the scope of the unit of hazard in question.

When performing assessments with a hazard unit that is an individual strain or isolate, identification rather than taxonomy or nomenclature becomes the issue. In this case identification refers to the verification of the labeling of an isolate, as well as the placement of an isolate within an existing taxon, or determining that it does not match any existing taxon. The purpose of identification is to be sure that you know which organism/unit of hazard applies to a data set. For example, if occurrence data are for a set of isolates and dose-response data are for only one of the isolates, then the uncertainty this introduces needs to be discussed.

In rare situations, the unit of hazard (e.g., genus, species, subtypes) may not be identifiable. However, defined host symptomatology may lead to an underlying suspect agent (Soller et al., 2010). Under such circumstances, the vehicle of transmission of the suspect agent may be considered as the unit of hazard. Overall, the level of details in microbial HI/HC depends on the risk assessment characteristics and risk management needs (e.g., issues, goals).

3.9 What Host Factors Can I Take into Consideration?

You can consider the following factors when evaluating potential health effects due to exposure to a pathogen. Following each factor is a short description of how the factor may influence the health outcome. There may be little or no data for these factors, so their consideration may be limited to a qualitative discussion (adapted from USACHPPM, 2009; EPA 2009a).

a) **Age/Life Stage** – Life stage refers to a distinguishable time frame in an individual's life characterized by unique and relatively stable behavioral and/or physiological characteristics that are associated with development and growth. Children and elderly are usually considered more susceptible due to immaturity or other potential weaknesses in their immune systems. Behaviors that affect pathogen exposure patterns may also be related to age. For example, children may also experience greater exposure (therefore larger doses) due to their behaviors (e.g., close-proximity playing, hand-to-mouth tendencies).

b) **Pregnancy** – Women who are pregnant and fetuses are generally considered to be a sensitive and perhaps more susceptible life stage. For example, Hepatitis E, which is a self-limiting disease for most people, can cause up to 20% mortality in women in the third trimester of pregnancy (Jameel, 1999). The underlying reason for increased susceptibility is due to the influence of pregnancy on the immune system. Pregnancy can also change exposure patterns, for example, water consumption in pregnant women is higher than the in the general population.

c) **Immune Status** – The immune system plays an important role in clearing pathogens from the human body, which influences potential health effects. Previous exposure may confer limited and/or short term protective immunity (Frost et al., 2005) or long lasting immunity (especially to viruses). The converse of this may also be true, that is, when individuals or subpopulations that have not previously been exposed to particular pathogens, infection and illness rates can be higher than would otherwise be anticipated. "Traveler's diarrhea" is an observed phenomenon that exemplifies this type of situation. In addition, individuals with compromised immune systems who come into contact with pathogens may react very differently from individuals with intact immune systems. Definitions of subpopulations or life stages included in the risk assessment should include the criteria used to classify individuals as immunocompromised and may need to be limited to specific identifiable types of immune defects. Note that children, newborns, the elderly, and pregnant women are also immunologically different from healthy adults.

d) **Natural Microbiota** – The competition that is provided by the presence of natural microbiota influences the impact a pathogenic organism will exert on a host. Prior treatment with antimicrobials that alter gut flora is a recognized risk factor for infection with *C. difficile* and *Salmonella*.

e) **Nutrition** – The nutritional state of the host affects the immune system. Malnourished individuals tend to have weaker immune defenses than well nourished individuals.

f) **Clearance Mechanisms**[18] – The human body has clearance systems to remove foreign particles from tissues. For example, nasal and oral clearance systems can remove airborne hazards and intestinal clearance mechanisms can help remove

[18] These may also be referred to as innate immunity.

gastrointestinal pathogens. Intestinal hypermotility may represent a host defense mechanism against *Giardia* (Anderson et al., 2006). Gastric acidity is a barrier that is a primary factor affecting the outcome of infections from food and waterborne pathogens. Certain behaviors, for example smoking, may affect the functionality of clearance mechanisms (e.g., mucociliary escalator); therefore, the functionality of clearance mechanisms of various host subpopulations should be considered.

g) **Genetic Factors** – The expression of certain genetic factors may increase an individual's sensitivity to particular pathogens. Therefore, if genetic factors are known for the organism being assessed, expression level differences could be considered when characterizing the pathogen and performing dose-response modeling. Some genetic factors that influence pathogen dynamics may be linked to race, which could be considered in the characterization of a hazard and dose-response modeling.

h) **Preexisting Conditions** – Preexisting conditions may affect a host's response to a pathogen, therefore, preexisting conditions should be considered, if possible. Physical and emotional stressors may also influence host susceptibility.

i) **Carrier Status (Persistence in Population)** – The possibility of some humans to serve as "carriers" for pathogens needs to be considered when estimating the potential spread of pathogens, especially when the a carrier may interact with hosts who are considered susceptible.

j) **Treatment Efficacy** – Whether or not effective treatment is available may be important to risk assessment. Treatment efficacy can be a major determinant of mortality.

k) **Social and Behavioral Traits** – Social and behavioral traits primarily affect exposure patterns. For example, a relatively small proportion of the population is responsible for consuming the majority of raw and partially cooked shellfish (FDA, 2005; see age and behavior above). Social and behavioral traits may also be associated with cultural and racial identities and may be important for specific consideration in a risk assessment.

3.10 How does Life Stage Affect Sensitivity to Infection and Disease Manifestation?

Sensitivity to infection is based on both exposure to a pathogen and the integrity of the immune system. Early life stages have a combination of factors that increase both the possibility of infection and intensity and duration of the disease. Young children spend the first two years or more close to the ground whether crawling or playing. Reliance on hands to move around, hand-to-mouth activity, and eating with hands as opposed to using utensils all raise the possibility for exposure and ingestion of pathogenic microorganisms compared with more mature individuals.

A second issue with children is the immature development of their immune systems. Infants have not had the exposure to the wide range of microbial stressors needed to afford protection. Newborn infants have passively acquired immunity from their mothers, which dissipates over the ensuing months. This provides some protection for newborn children, allowing them to develop the array of acquired immunity needed for full protection. Nonetheless, young children tend to be more susceptible than older individuals.

A significant consideration is the intensity and persistence of the disease in newborn infants. In the absence of prior experience with a specific pathogen, the body requires more time to develop and process its immune response. Therefore, a pathogen can elicit more pronounced and longer lasting effects from infection resulting in more adverse outcomes in children compared with adults. In developing countries, where treatments are limited, children succumb to gastrointestinal infections at a higher rate than in countries where timely medical intervention is widely available.

EPA's Risk Assessment Forum published *Guidance on Selecting Age Groups for Monitoring and Assessing Childhood Exposures to Environmental Contaminants* (EPA, 2005). Based on physiological and behavioral milestones EPA recommends that children be grouped by the following ages:

a) Less than 12 months old: birth to <1 month, 1 to <3 months, 3 to <6 months, and 6 to <12 months.

b) Greater than 12 months old: 1 to <2 years, 2 to <3 years, 3 to <6 years, 6 to <11 years, 11 to <16 years, and 16 to <21 years.

In addition to children, the elderly may also be more susceptible to infections than healthy adults. Risk assessments designed to include the whole population should include discussion of the elderly, even if data specific to this subgroup are not available.

3.11 What Environmental Factors Can I Take into Consideration?

From the point of origin until it reaches the host, a given microorganism interacts with the environment at various levels that influence its survivability and virulence. Hence, at the point when the microorganism finally encounters the host, the ability to cause adverse outcomes depends on the complex interactions (microorganism-matrix, matrix-matrix, microorganism-carriers/vectors) that the microorganism experienced during transport. For example, the survivability, virulence, transmission and successful development of the diseases/adverse outcome for a given microorganism may be influenced by the run-off from animal husbandry or from wild animal habitats into water bodies that can reach the target host directly through exposure to water or indirectly through zoonotic transmission routes or contaminated foods. Thus, you should consider the potential role of the non-host environmental conditions in influencing changes in the

properties of a microorganism to render it potentially hazardous. These may include matrices (air, water, soil, food), fomites, vectors, and carriers. As the physical, chemical, and biological traits of the matrix can influence the microorganism's virulence and survivability, a thorough characterization of the matrix becomes critical in a given risk assessment. Environmental factors can be considered with the epidemiological triad in mind (Figure 1.2), which also provides context for segueing into exposure assessment (see Chapter 5).

4. DOSE-RESPONSE ASSESSMENT

The goal of the "dose-response assessment" in MRA is to establish the relationship between the dose of a pathogen that individuals or subpopulations are exposed to and the probability of adverse health effects (e.g., infection, illness, death) to individuals or subpopulations. Qualitative evaluation (hazard characterization) of a pathogen is also included in the conclusions drawn with regard to potential health impacts, particularly if data for a quantitative MRA are not available. From the estimated quantitative relationship (dose-response model), the probability of potential adverse health effects of a given severity can be estimated from a given exposure to a pathogen. The exposure assessment and dose-response assessment are combined in the risk characterization step where risk due to a particular exposure for a defined subpopulation and a defined hazard is described. It is important to note that because the dose-response assessment is used in the context of an exposure assessment, it is critical that the dose units used for quantitative estimates are comparable.[19]

It should be noted up front that there is no general accepted guidance regarding when the available microbial dose response data are sufficiently representative for a particular scenario, as the factors associated with each scenario are likely to be unique to each case.

The information in this chapter is organized into two sections: 1) general considerations for dose-response modeling (Section 4.1); and 2) current practice in dose-response modeling methods (Section 4.2). The section on "General Considerations for Dose-Response Modeling" discusses the technical application of using statistical models to estimate health effects due to exposure. The second section focuses on the currently available quantitative dose-response models and on the development of new alternative models.

4.1 What is Dose-Response Modeling and What are Some General Considerations for Dose-Response Modeling?

Dose-response modeling is the process of using mathematical relationships to describe the probability of an adverse health effect (e.g., infection, illness) occurring in an individual or the frequency of an adverse health effect in a subpopulation when that individual or subpopulation is exposed to a specific dose of pathogenic microorganisms. The dose level may be measured in terms of a discrete number of organisms (e.g., oocysts), colony forming units (cfu), plaque forming units (pfu), or by molecular methods that may estimate gene copies or cell equivalents. A cfu represents one to several viable bacteria cells while a pfu represents one to several infectious viral particles. Alternatively, a dose level may be specified by an average administered or ingested dose or represented by some other measure (e.g., median infectious dose [ID_{50}] units). The most common practice of dose-response modeling has been the fitting of limited data sets

[19] For example, the number of organisms ingested in a serving for a human volunteer trial might be converted to number of organisms in a daily exposure. The units might go from event based in the dose-response assessment to a series of daily exposures over a lifetime for the exposure assessment.

that have been derived from experimental trials to statistical models that are often, but not always, biologically based. More recently, researchers have also been using outbreak data for dynamic dose-response modeling of disease incidence in subpopulations, and there is also a movement towards the development of models that incorporate the inherent complexities found in any biological system.

4.1.1 How do I Choose Between Modeling a Discrete Dose Versus an Average Dose?

Pathogen doses are inherently discrete. While chemical doses are expressed in mass units (e.g., mg/kg), pathogen doses are usually expressed as counts of organisms (e.g., oocysts/liter) or an average dose (e.g., mean cfu per serving). Although a group exposure may be described in terms of average pathogen concentration, pathogens are distributed in a particular medium (water, air, food), such that each individual may not receive exactly the same number of organisms. If the concentration of pathogens is very low, some individuals could be exposed to zero pathogens, while others could be exposed to one or more pathogens. In this type of situation, the heterogeneity of the pathogen distribution in the matrix can be very important.[20]

While the average dose of pathogens is continuous and can take any value, the actual number of organisms that an individual may consume is a discrete quantity. In the context of a clinical feeding trial, the distribution of pathogens or cfu in the delivery matrix generally is assumed to be random but homogenous (i.e., with the same mean), with the probability of exposure to a discrete quantity of organisms or cfu (0, 1, 2, etc.) given by the Poisson distribution. Because distributional data is preferred in MRA over point estimates (OMB, 2007b), if you only have data on the average dose, then you can incorporate some assumption about the distribution (e.g., Poisson, extra-Poisson, exponential, mixture). Using *Cryptosporidium parvum* as an example, for an average dose of 0.5 oocysts, most (60 percent) individuals would consume no oocysts, while 30 percent would consume a single oocyst and 10 percent would consume 2 or more oocysts. Drinking water exposures are usually low, often below an average dose of 1×10^{-4} organisms per liter, which essentially means that 1 out of 10,000 individuals would ingest a single organism after drinking one liter of water at that concentration.[21] In modeling feeding trial data where each dose is not directly counted, it is reasonable to assume a Poisson distribution, but this may not be a reasonable assumption in modeling outbreak data.

For low exposures, the discrete nature of the exposure constrains the maximum risk of infection to the probability of exposure. That is, in the previous example, if the pathogen were 100 percent infective, only one individual would become infected. You

[20] Pathogens may clump together, particularly if the matrix has other components that aid clumping.

[21] In all practicality, doses less than 0.01 represent dilutions of single organisms, with an insignificant probability (though still not zero) of exposure to more than one organism. As a result the response is a virtually-linear function of dose at very low doses. Any function used for pathogen dose-response must follow the same "rule." Otherwise, a probability of infection greater than the probability of exposure could be predicted.

should consider using discrete dose-response models in these cases, as the risk is usually expressed as a unit-pathogen infectivity, which is greatly influenced by the high Poisson variability in the dose. In contrast, while exposure to pathogens in food is typically very low, due to the potential for pathogen growth in some foods prior to consumption, the exposure distribution may be extremely skewed with some frequency of exposures in the range of millions to billions of organisms per exposure. In these cases, the Poisson variability around the mean dose is trivial, such that doses in this range can be considered to be continuous and strict adherence to discrete models is not required. In these cases, you can use quantitative dose-response models based on the *average* pathogen dose to estimate the probability of infection or illness (Haas et al., 1999).

The Beta-Binomial dose-response model assumes that the exact number of organisms ingested is known, which is suitable for a feeding trial in which each administered dose has been enumerated. The exponential and beta-Poisson models assume that the number of organisms between subjects in a dose group is Poisson distributed with a fixed mean (FAO/WHO, 2009).

4.1.2 What is the One-Hit Model and Why is it the Preferred Model?

One-hit (or no-threshold) dose-response models are generally the most relevant for microbial dose-response assessment. However, these models may not apply to all pathogens that cause illness by producing pre-formed toxins in food and they may be inappropriate for modeling illness and mortality.

FAO/WHO (2003) observed that despite the traditional concept of a "minimal infectious dose" in the microbial literature, attempts to define the numerical value of a threshold level of pathogens that must be ingested in order for the microorganism to produce infection or disease have typically been unsuccessful. "An alternative hypothesis is that, due to the potential for microorganisms to multiply within the host, infection may result from the survival of a single, viable, infectious pathogenic organism ("single-hit concept"). This implies that, no matter how low the dose, there is always, at least in a mathematical sense, and possibly very small, a non-zero probability of infection and illness" (FAO/WHO, 2003). Similarly, NRC (2003) noted that unlike chemical risk assessment, "microbial dose-response assessment for infectious pathogens does not produce any concept analogous to the no observed adverse effect level (NOAEL), since a single microbial cell may (under the right circumstances) produce illness." Finally, NRC (2005) observed: "In assessing risks attributable to exposure to microorganisms, it has frequently been asserted that there exists a threshold (minimum infectious dose) below which there is no risk to the population. Such a concept is not consistent with the current understanding of microbiological risk assessment.... The no-risk concept originated from the fact that in trials (either animal or human), low doses of microorganisms often produced no adverse effects in exposed subjects.... [However,] all animal and human exposure data that have been subjected to dose-response analysis are consistent with models in which the dose intercept is zero and the value of the conditional dose-response relationship for one organism is non-zero." Empirically, it is impossible to distinguish between a very low non-zero risk and a true threshold. Risk assessors must rely on

concept and theory to establish the most relevant dose-response modeling approaches. Therefore, in the ensuing discussion, the one-hit dose-response models are given preference over alternative models typically used for chemical dose-response modeling (e.g., log-normal, log-logistic, Weibull).

The one-hit model assumes that one infectious organism has the potential to cause infection. When an individual organism is ingested, the probability that the organism defeats the host barriers and initiates an infection may be represented by a unit-infectivity measure. This measure is termed "*r*" by convention, as *r* is the rate constant in the most-commonly used exponential dose-response model. The exponential distribution is based on the assumption that each organism is capable of initiating an infection and behaves independently from other organisms within the host, leading to a binomial probability of infection. The assumptions of a Poisson pathogen distribution and a binomial probability of infection lead to a family of models referred to as one-hit models, where the name relates to the concept that only a single organism is necessary to cause infection. Different pathogen distributions can be assumed in addition to the Poisson. If there is reason to believe that the pathogens are not uniformly distributed in the exposure medium (e.g., clumped), a negative binomial or other skewed distribution can be used (Haas et al., 1995; Teunis et al., 2008b), although the numerical algorithms become more complicated. From a modeling perspective, the one-hit model means that there is a non-zero probability that any given organism will survive the host defenses to initiate an infection, no matter how low the probability may be. Unit-infectivity probabilities can range from 1×10^{-10} to greater than 0.1 (Teunis et al., 1996; Haas et al., 1999). Although, conceptually based on biologically plausible mechanisms, most of these models do not rely on independently validated parameters describing the mechanisms model the underlying physiological processes explicitly, but rely on fitting curves to empirically observed data. The potential for applying physiologically-based models is discussed in section 4.2.8.

The one-hit concept also applies to the production of illness in that, once an individual is infected, progression to illness can proceed without additional exposure. Replication within the host of that single pathogen from the initial exposure would eventually result in enough pathogens to produce illness symptoms. However, the standard one-hit dose-response models are typically used for assessing the risk of infection rather than illness (or mortality). Morbidity and mortality may be expressed in only a small fraction of infected individuals. Since morbidity is currently most commonly assumed to be independent of dose (but conditional on infection) it is best to use a morbidity or mortality ratio applied to the risk of infection (Haas et al., 1995). As a special case ("quorum sensing"), illness may not occur until the number of organisms reaches a critical mass at which time they release the toxins resulting in illness. However, the increase in the number of organisms is a result of growth within the host rather than an increase in the exposure dose. The concept of density dependent quorum sensing is distinct from a threshold for administered dose, because of the possibility, however small, that a single ingested organism may survive the multiple barriers in the gut to become established and reproduce (FAO/WHO, 2003). Threshold models have not been demonstrated to provide significant improvements in fit over the exponential and

beta-Poisson models, but their use has been advocated on the basis of analysis of the infection process and interpretation of epidemiological data.

4.1.3 What Important Factors Can I Consider in Dose-Response Assessment?

Exposure Route

The route of exposure can have a significant bearing on the dose-response curve. As noted above, for infection to occur and result in a disease state, the organism must penetrate the host's defenses whose capability varies with tissue type. You need to be aware that route of exposures can influence both the slope of the dose-response curve as well as the manifestation. For example, Adenovirus can be highly infectious through an inhalation route of exposure, and appears to be less infectious through ingestion. Therefore, matching the route of exposure, with appropriate dose-response information is important. Discussion of the implications of using the available dose-response dataset for broader exposure scenarios is helpful.

In chemical risk assessment, route-to-route extrapolation of internal dose for systemic effects can be performed if adequate toxicokinetic data exist.[22] For pathogens, although systemic involvement can be an important factor, some pathogens can only infect specific tissue types. Other pathogens can infect many different tissue types and have a different dose-response and health endpoint for those different tissues. For example, *Cryptosporidium* usually infects the gastrointestinal tissues, but in immunocompromised patients, it can infect other tissues including lungs and result in a different set of symptoms (O'Donoghue, 1995). Usually the most obvious concern is where the route of exposure leads directly to the most susceptible tissues, such as diarrhea from ingestion, pneumonia from inhalation, or dermal lesions from skin contact. However for pathogens that cause illness through release of toxins, the host tissue that is most susceptible to the toxin may be remote from the physical location of pathogen replication.

Exposure Medium

The nature of the exposure medium can influence the probability that an individual pathogen will survive host defenses and initiate an infection. For oral exposure, pathogens can be ingested in food, water, or from direct contact with fomites or infected individuals. The primary factor in this process is the initial line of defense against the pathogen—stomach acid and digestive enzymes. If the ingestion medium serves to raise the pH in the stomach content, the probability of pathogen survival is enhanced for many organisms, perhaps by orders of magnitude, depending on the extent of acid buffering. The nature of the food matrix, for example, with respect to food structure and fat content, can vary considerably in enhancing or limiting survival of pathogens in the food matrix and in the host gastrointestinal ecosystem (Ross, 2008). In

[22] Chemical risk assessors have defined many different aspects of dose, such as potential dose, applied dose, absorbed dose, internal dose, and delivered or biologically effective dose.

addition, taking stomach-acid reducers for acid-reflux disease can provide protection for the pathogen. Ingestion of drinking water, on the other hand, offers no protection from stomach acid.

In the dose-response documentation, you should discuss possible effects of different matrices relevant to the assessment. You could compare the delivery matrix used in generating dose-response data from a human feeding study to the matrix being considered in the exposure scenario. For example, if the dose-response data is from feeding trials using a water matrix and the exposure scenario in the risk assessment is juice, then you should clearly describe the difference and elaborate on what the potential implications of those differences might mean for the risk assessment.

Pathogen

During hazard identification the pathogen of concern is defined. In the dose-response assessment, discussion of whether or not the pathogen of concern matches the pathogen for which dose-response data is critical. This issue is important because substantial variability in virulence and infectivity has been shown for closely related pathogens. For example, the relative infectivity of the *Salmonella enterica* serotypes (Coleman and Marks, 2000; Soller et al., 2007) and *C. parvum* isolates (Haas et al, 1999; Messner et al, 2001; Teunis et al., 2002; Englehardt and Swartout, 2004) used during human dose-response challenge studies varied by several orders of magnitude. Although you may only have dose-response data for one or a few isolates of a species, risk managers may be interested in the species as a whole (e.g., the *C. parvum* example above) or the genus (given that a number of *Cryptosporidium* spp. may infect humans)(Xiao, 2010). Isolates only represent a small fraction of the genetic diversity of the species that is likely to occur in nature. Discussion of the implications of using the available dose-response dataset for broader exposure scenarios is helpful. There is no general guidance regarding when the available dose response data are sufficiently representative for a particular scenario, as the factors associated with each scenario are likely to be unique to each case.

Data on the actual pathogen of concern are preferred over data on surrogate organisms.[23] In cases where data on the pathogen of concern are not available, data on surrogate organisms can be used if solid biological evidence, such as common virulence factors, can support that choice. The biological basis for the use of the surrogate must be clear (FAO/WHO, 2003).

Haas et al. (1999) provides a method for testing whether differences among strains are statistically significant. It is also important to determine whether the strains can be considered a representative sample from the pathogen of concern or an extreme case. In cases where the strain is representative, you may want to use a mixed model with random strain effects. In cases where the strain is an extreme example, a bounding approach can be used (Vose, 2008).

[23] "Surrogate organism" synonyms include "index pathogen" and "reference pathogen."

Host

During hazard identification you should discuss host factors that are relevant for the risk assessment scenario. Given the current state of knowledge, quantitative microbial dose-response assessment for humans requires some human pathogenicity data. When using dose-response data from human trials, you should discuss the characteristics of the subpopulation in the trials compared to the subpopulation defined in the scope of the risk assessment. You can specifically discuss the types of individuals that are explicitly represented in the human trials (e.g., healthy adults) and individuals that were excluded from the trials (e.g., children, elderly, immune compromised, and pregnant women). Describe the potential implications to the different subpopulations or life stages, and point out any tools that you used to compensate for the differences, such as information from epidemiological data from outbreak situations. Also discuss the likelihood that individuals in the trials may have had some immunity to the pathogen being evaluated.

Although human data are preferable, animal studies and *in vitro* studies may provide useful information on host-pathogen interactions (FAO/WHO, 2003). The strengths and limitations of those data, within the context of your risk assessment scenario, should be clearly explained. Some pathogens have evolved a narrow host range, while others have evolved a broad host range. Many important pathogens are host-specific, but over 60 percent of human pathogens have multiple domestic mammal hosts (Cleaveland et al., 2001). Among pathogens of wild primates, only 10 percent of bacteria, 13 percent of viruses, and 28 percent of protozoa are host-specific, and more than 100 pathogens (including 19 bacteria and 30 viruses) infect both wild primates and humans (Pedersen et al., 2005). Viruses mutate rapidly and can "jump" to new host species more easily than bacteria or protozoa. On the other hand, protozoa can be much less discriminating in their choice of host species, often infecting humans after passing through wild and domesticated animal species. However, the disease manifestations can be quite different, implying that different pathogenic mechanisms are in operation. As an example, *C. parvum* typically causes diarrhea in humans with no systemic involvement but kills (immunocompromised) mice from generalized systemic distribution with no diarrheal symptoms. One host cannot be used as a surrogate for another in all respects, and extrapolation from surrogate host data may require adjustments. Therefore, when surrogate animal models are used, the biological basis for and limitations of the use of the surrogate must be clear (FAO/WHO 2003).

Endpoint

The endpoints that are typically modeled in MRA are infection, illness, and death. Infection is the most immediate health effect for direct modeling, as it is the first manifestation of exposure to a pathogen. However, infection is difficult to assess in humans if there are no clinical symptoms. Infection is generally equated with colonization of some tissue, either externally or internally. Animal studies can include specific tissue analysis of sacrificed groups at intervals following the original inoculation to analyze the clearance time and/or the level of colonization (EPA, 2007a). However,

colonization *per se* is difficult to measure in humans, so typical markers (measurement endpoints) of infection such as shedding of the pathogen in feces or urine, presence of antibodies in the blood (seroconversion) or clinical symptoms are often used to determine if infection has occurred. Although (asymptomatic) infection has no direct adverse health impact, it can be crucial in determining the risk of illness and plays an important role when estimating the impact of secondary transmission. Illness and mortality are the primary endpoints of health concern. Illness endpoints could range from gastrointestinal distress to long-term sequelae. The risks of illness and death generally are estimated from the risk of infection by applying morbidity or mortality ratios, but can be modeled directly in certain circumstances. For example, the most commonly employed dose response relationship for the ingestion of *Salmonella* through a waterborne route of exposure is for an endpoint of illness, not infection. Define the health endpoint of concern and specifically relate that definition to the clinical case definition used in any utilized dose-response studies. If there are any differences between the endpoint definition for the risk assessment and the case definition for data from trials or outbreak studies, you should describe the implications of those differences to the interpretation of the risk assessment results.

Inconsistencies in illness definitions can introduce uncertainties. For example, the definitions of diarrhea may differ slightly between different clinical trials or epidemiological studies and may be based on moisture context of stools. Illness definition endpoints can be quite complex. For example, EPA's bacterial ambient water quality criteria for recreational waters are based on highly credible gastrointestinal illness as defined as "any one of the following unmistakable or combinations of symptoms (within 8 to 10 days of swimming): (1) vomiting; (2) diarrhea with fever or a disabling condition (remained home, remained in bed or sought medical advice because of symptoms); (3) stomachache or nausea accompanied by a fever" (Dufour, 1984). Note that individuals with only diarrhea are not "counted" as a case with the above definition. Other studies may include diarrhea alone, without fever as a valid case of gastrointestinal illness. For example, norovirus does not typically cause fever. You should pay close attention to the nuances of how endpoints are defined and discuss the impact any differences may have on the risk assessment.

How treatment can impact the health outcome should be discussed. Some infections are treatable and others are not. Treatment efficacy could influence determining the most relevant health outcome.

Sources of Data

Clearly document what sources of data were considered, utilized, and omitted, and provide justification for those decisions. The most precise human data are the direct pathogen-challenge studies (clinical feeding trials), in which human volunteer subjects are fed known doses of a specific "enteric" pathogen and observed for gastro-intestinal symptoms over a period of time. These data can be modeled directly to obtain dose-response relationships and parameters for application to specific human exposure scenarios. For ethical reasons, direct pathogen-challenge studies are typically limited to

healthy adult subjects challenged with a pathogen that has well-characterized health outcomes that are no more serious than temporary diarrhea that is self-clearing. Human pathogen-challenge data has been used in EPA regulations to predict the risk of giardiasis (EPA, 1989, 1998b, 1999) and cryptosporidiosis (EPA, 2006a). Although data from clinical feeding trials are carefully collected and documented, there may be limitations to extrapolating from experimental conditions that need to be addressed (e.g., subjects limited to healthy adults, use of non-wild-type pathogen strains, dose delivery matrix buffered to increase the likelihood of pathogen survival).

Human dose-response information can also be obtained from epidemiological data (primarily retrospective outbreak analyses). Although such data may be more representative of the actual host-pathogen-matrix combination of concern, there is usually considerable uncertainty in the dose estimate, number of exposed or number of responders, or combinations of those variables. One significant advantage that epidemiological data have over clinical feeding trials is the potential for evaluating the relative risk of sensitive populations, as was done for *Escherichia coli* O157:H7 (Teunis et al., 2004, 2008a).

Animal dose-response data have been used to estimate the human dose-response curve and to estimate the innate pathogen variability across strains. However, these data are difficult to translate directly to human dose-response and require critical evaluation prior to use due to all of the uncertainties associated with interspecies extrapolation. Epidemiological information also can be used to calibrate ("anchor") dose-response curves derived from animal data with respect to the relevant human response range; this was done in the FDA/USDA/CDC *Listeria monocytogenes* risk assessment by shifting the mouse mortality dose-response curve based on human mortality rates attributed to *L. monocytogenes* (FDA/USDA/CDC, 2003). In addition, the variability of median lethal doses (LD_{50}s) in mice across *Listeria* strains was used as an estimate of the pathogen virulence variability. The assumption was made that the pathogen virulence variability was similar to the level of variability likely to be encountered in human exposures and was useful for dose-response uncertainty analysis. Note that this use of animal data is not in lieu of human data, but is used to support assumptions about characteristics of human data, such as potential range of variability. For more information on the use of animal data in MRA see EPA (2009a).

Outbreak investigations can often provide valuable information about the etiologic agent. A pathogen's ability to produce an outbreak depends on specific characteristics such as ability to survive in the environment and rate of growth or die off, potential to cause disease at a given dose, most likely transmission route, and capacity to spread through person-to-person contact. Therefore, studying the details of outbreaks can help risk assessors develop exposure scenarios for specific pathogens, and data from outbreaks can be an important comparison for dose-response models based on human feeding trials or animal models. The major limitation to outbreak investigation data is that investigators often collect a narrow range of information. Their main objective is to rapidly identify the vehicle and prevent additional infections. Occasionally, data from concurrent drinking water testing are available, or frozen or unopened suspect food

remains for sample collection and accurate identification and enumeration of pathogens. If actual levels of food or water contamination can be measured or estimated, it may be possible to estimate the dose-response relationship from outbreak data. An outbreak that is characterized by a low attack rate in a very large population may provide an opportunity to define the host-response to very low doses of a pathogen (Teunis et al., 2004).

Like outbreak data, annual surveillance statistics provide a way to evaluate MRA models. The initial results of a MRA model can sometimes be cross-checked by comparison with public health surveillance data. The accuracy of dose-response models may be assessed by combining them with exposure estimates known to be realistic and determining if the results approximate the incidence of illness estimated from surveillance data, taking into account the uncertainty due to under-reporting. Using annual disease statistics in modeling dose-response and exposure estimates implicitly includes the entire population and the wide variety of factors that can influence the response. Another benefit is that surveillance databases may have sufficient detail to analyze special subpopulations or life stages such as the elderly or the immunocompromised. If information is available, the surveillance summaries or a series of reported foodborne or waterborne outbreaks can also identify the etiologic agents causing disease outbreaks and often the sources of the contamination. Consultation with public health surveillance experts can provide a "reality check" on the preliminary results of risk ranking and other quantitative MRAs.

4.1.4 How Can I Model the Spread of Disease in the Population?

In addition to primary transmission via contaminated media (e.g., water or food), many microbial pathogens can also be transmitted via person-to-person contact and cause infection and disease.[24] This route of microbial exposure, occurring from an infected person rather than from contaminated media is also referred to as "secondary transmission." You can discuss any known features of secondary transmission when characterizing the microbial hazard, regardless of whether the scope of the risk assessment includes modeling secondary transmission. The ability with which an organism can be transmitted from an infected individual to an individual that is susceptible to infection is an important consideration and should be evaluated during the exposure assessment if possible.

Secondary cases (often represented in epidemiological studies by a secondary attack rate) generally refer to cases that occur among contacts, within the incubation period of the pathogen, and following exposure to a primary case. In some cases, direct person-to-person transmission cannot be distinguished from contamination of the immediate environment (e.g., toddlers sharing toys versus direct physical contact during play). Depending on the purpose of the assessment, it may be appropriate that the

[24] Some situations blur the line between secondary transmission and environmental transmission. For example, infections due to a primary case causing an outbreak of cases in a daycare setting is usually classified as secondary transmission even though transmission may be from both direct human contacts and contaminated objects, food, or water exposures.

definition of secondary transmission include infections that result from propagation of the specific exposure of interest, but not encompass distant transmissions (separated by time and/or space) that may be more appropriately considered to result as a function of person-to-environment-to-person transmission. Temporal and spatial limitations can be specifically noted in the definition of secondary transmission. You can discuss the full range of scenarios that qualify as both primary and secondary transmission. The above definition of secondary transmission is limited to avoid overlap with pathogen occurrence in the environment (person-environment-person), although people are, of course, part of the environment. However, the potential for reintroduction of the pathogen into the exposure media could also be within the definition of secondary transmission.

The degree to which the exposed population is susceptible to infection and illness is an important factor in deciding whether to explicitly model secondary transmission in a risk assessment. Population susceptibility and immunity change dynamically as a population is exposed to a pathogen. Moreover, since many infections are asymptomatic, it is common for pathogens to be transmitted from person to person during asymptomatic infections or during asymptomatic periods of an infection (before or after symptoms), when infected individuals interact normally with susceptible individuals. Previous infection by the same organism often confers temporary or permanent immunity, although individual immunity may be partial (e.g., resistance to infection may be overcome when challenged by the same pathogen at a high dose or by a genetically different strain). If a significant proportion of the population is immune, such as from an immunization program, secondary spread of disease can be virtually prevented.

MRA models can be configured to account for secondary transmission and immunity in a population through the use of a dynamic model (Anderson and May, 1991). These models, which can take several forms (deterministic or stochastic), characterize the dynamic epidemiological status of the population (e.g. susceptible to infection, symptomatic infection, immune). Static MRA models do not, by their nature consider secondary transmission, although dose-response parameters derived from static models may be incorporated into dynamic models. You can indicate if and how secondary transmission is included in the assessment. Inclusion of secondary transmission in MRAs often provides non-intuitive results; therefore, if secondary transmission and other innate characteristics of infectious disease transmission are not included in the assessment, provide a sound justification for this decision.

4.1.5 What Can I Address for Each Model to Improve Transparency?

To promote transparency in presenting the dose-response assessment, you can address the following points for each model presented:

a) Assumptions

1) Clearly state models' key assumptions

2) Discuss assumptions inherent when extrapolating to doses lower than those used in studies.

3) Discuss flexibility in approaches to the dose-response relationship depending upon the pathogen being considered and the assumption about a no-threshold effect (i.e., can it be assumed that one organism is sufficient to produce infection?).

b) Applicability of Models

1) Discuss the biological rationale for the model
2) Discuss the applicability of each model to various exposure situations
3) Discuss limitations of models
4) Articulate strengths/weaknesses and advantages/disadvantages of the models

c) Results

1) Discuss the type of information that the various models are expected to provide.
2) Discuss the use of likelihood methods to compare how well dose-response models fit the data

4.2 What is Current Practice in Quantitative Dose-Response Modeling for Microbial Illness?

This section briefly summarizes some common dose-response models and how those models have been used in previous MRAs. It also discusses the output of dose-response modeling and evaluating uncertainty and accounting for life stages and subpopulations.

4.2.1 What Models Can I Use for Microbial Dose-Response Assessment?

Dose-response models are mathematical functions that yield a probability of an adverse health effect as a function of dose. Numerous dose-response relationships for microbial endpoints have been published in the peer-reviewed literature. No comprehensive summary of dose-response models is available for all human pathogens. However, a thorough summary of dose response models for waterborne pathogens can be found in Table 4.1. The two most commonly used dose-response models are the exponential and beta-Poisson. Several alternative models have also been proposed as alternatives for MRA, including two-parameter models such as log-normal, log-logistic, and extreme value models (Pinsky, 2000), and three-parameter models, such as Weibull gamma (Farber et al., 1996), exponential gamma, Weibull exponential, and the shifted Weibull model (Kodell, 2002). However it should be noted that there is no single selection criteria for dose-response models that is universally used. Several criteria that could be used are provided in Section 4.2.3. You should explain the logic behind your model selection and strengths and limitations of the model selection.

The models discussed in this section estimate risks for exposed individuals; thus, they are known as individual risk models.[25] Population risks (the incidence of disease among a group of exposed individuals) are generally constructed by combining the results of individual risk models with estimates of the distribution of doses in the exposed population (EPA, 2009a).

[25] Note that "individual" risk models may have as their outputs probability distributions of risk that can be interpreted to reflect (1) uncertainty in infectivity of the agent tested, and/or (2) variability in individual susceptibility among the experimental subjects.

Table 4.1 Overview of Dose-Response Relationships for Waterborne Pathogens[a] (Source: EPA 2009a; Adapted from McBride et al., 2002)

Microorganism	Model	Parameters[b]	Reference(s)
Adenovirus 4	Exponential	$r = 0.4172$[c]	Crabtree et al., 1997 Haas et al., 1999
Campylobacter jejuni[h,i]	Beta-Poisson	$\alpha = 0.145$ $\beta = 7.59$	Haas et al., 1999 Medema et al., 1996 Teunis et al., 1996
	Infection: Hypergeometric beta-Poisson Illness: Conditional on infection[g]	$\alpha = 0.024$ $\beta = 0.011$ $\eta = 3.63 \times 10^{-9}$ $r = 2.44 \times 10^{8}$	Teunis et al., 2005
Coxsackievirus	Exponential	$r = 0.0145$	Haas et al., 1999
Cryptosporidium	Exponential	$r = 0.0042$	Haas et al., 1996, 1999
		$r = 0.077$[d]	Okhuysen et al., 1999
		$r =$ in the range 0.04 to 0.16	EPA, 2006a
	Generalized beta-Poisson for Illness	$\alpha = 0.060$ $\beta = 0.095$	Englehardt and Swartout, 2006
Echovirus 12	Exponential	$r = 0.0128$	Haas et al., 1999
	Beta-Poisson	$\alpha = 0.401$ $\beta = 227.2$	Teunis et al., 1996
		$\alpha = 0.374$ $\beta = 186.69$	Regli et al., 1991 Rose and Sobsey, 1993
		$\alpha = 1.3$ $\beta = 75$	Rose and Gerba, 1991
Entamoeba coli	Beta-Poisson	$\alpha = 0.1008$ $\beta = 0.3522$	Haas et al., 1999
Escherichia coli (pathogenic strains)	Beta-Poisson	$\alpha = 0.1778$ $\beta = 1.78 \times 10^{6}$	Haas et al., 1999
E. coli O157:H7	Beta-Poisson[e]	$\alpha = 0.248$ $\beta = 48.80$	Teunis et al., 2008a
	Hypergeometric beta-Poisson	$\alpha = 0.084$ $\beta = 1.44$ (children) $\alpha = 0.050$ $\beta = 1.001$ (adults)	Teunis et al., 2004
Giardia lamblia	Exponential	$r = 0.0199$	Haas et al., 1999 Regli et al., 1991 Rose and Gerba, 1991 Rose et al., 1991 Teunis et al., 1996
Hepatitis A virus	Exponential	$r = 0.5486$[f]	Haas et al., 1999
Legionella	Exponential	$r = 0.06$	Armstrong and Haas, 2008
Norovirus	Infection (with aggregation): Hypergeometric function ${}_1F_1$ Illness: Conditional on Infection[g]	$\alpha = 0.040$ $\beta = 0.055$ $\eta = 2.55 \times 10^{-3}$ $r = 0.086$	Teunis et al., 2008b
Poliovirus I	Beta-Poisson	$\alpha = 0.1097$ $\beta = 1524$	Regli et al., 1991 Rose and Sobsey, 1993
		$\alpha = 15$ $\beta = 1000$	Rose and Gerba, 1991

Microorganism	Model	Parameters[b]	Reference(s)
	Exponential	r = 0.009102	Haas et al., 1999 Regli et al., 1991 Rose and Sobsey, 1993
Poliovirus III	Beta-Poisson	$\alpha = 0.409$ $\beta = 0.788$	Rose and Sobsey, 1993
		$\alpha = 0.409$ $\beta = 0.788$	Regli et al., 1991
		$\alpha = 0.5$ $\beta = 1.14$	Rose and Gerba, 1991
Rotavirus	Beta-Poisson	$\alpha = 0.26$ $\beta = 0.42$	Gerba et al., 1996
		$\alpha = 0.2531$ $\beta = 0.4265$	Haas et al., 1999 Regli et al., 1991 Rose and Sobsey, 1993
		$\alpha = 0.232$ $\beta = 0.247$	Rose and Gerba, 1991
	Hypergeometric beta-Poisson	$\alpha = 0.167$ $\beta = 0.191$	Teunis and Havelaar, 2000
Salmonella	Beta-Poisson	$\alpha = 0.33$ $\beta = 139.9$	Rose and Gerba, 1991
	Gompertz log	ln(a) in the range 29 to 50 b = 2.148	Coleman and Marks, 2000 Coleman et al., 2004 Soller et al., 2007
	Generalized linear mixed models and fractional polynomials of dose	$\beta_0 = 0.323$ $\beta_1 = 5.616$ $\beta_2 = -8.462$ $\beta_3 = -7.782$ $d^2 = 0.780$	Bollaerts et al., 2008
Salmonella (non-typhoid)	Beta-Poisson	$\alpha = 0.3126$ $\beta = 2884$	Haas et al., 1999
Salmonella Typhi	Fractional polynomials	$\beta_1 = -18.1425$ $\beta_2 = 22.5300 \times 10^{-5}$	Namata et al., 2008
	Beta-Poisson	$\alpha = 0.1086$ $\beta = 6{,}097$	Haas et al., 1999
		$\alpha = 0.21$ $\beta = 5{,}531$	Rose and Gerba, 1991
Shigella	Beta-Poisson	$\alpha = 0.21$ $\beta = 42.86$	Haas et al., 1999
Vibrio cholerae	Beta-Poisson	$\alpha = 0.25$ $\beta = 16.2$	Haas et al., 1999

[a] These calculations are based on available data that have used particular pathogen strains processed in particular ways. Where more than one strain of an organism has been studied in clinical trials, a wide range of infectivities can be discovered. Therefore it must be recognized that these calculations can carry a substantial degree of uncertainty.

[b] For the exponential distribution $N_{50} = 0.693/r$; for the beta-Poisson distribution $N_{50} = \beta * (2^{1/\alpha} - 1)$. Values are unitless.

[c] Developed for inhalation exposure to adenovirus 4 aerosols.

[d] Estimated based on ID_{50} reported for the Texas A&M University (TAMU) isolate.

[e] Represents a meta-analysis of seven outbreaks and adjusting for heterogeneity. Alpha/beta pairs derived via MCMC analyses are available from Dr. Teunis. Use of those pairs is preferred to the use of the values shown in this table

[f] Corresponding dose units are grams of feces.

[g] Dose-response relation for the conditional probability of illness in infected subjects = $1 - (1 + \eta CV)^{-r}$, where η and r are shown in the Table; CV is the dose (concentration × volume).

[h] An alternate dose-response model is proposed by Brynestad et al. (2008).

[i] Many of these models have been critiqued in the literature. For example, Coleman et al. (2004) suggest the dose-response models for *Campylobacter* identified in this table do not account for strain variability sufficiently and suggest the need for development of more detailed mechanistic models.

An overview of Exponential, beta-Poisson, deterministic, and Bayesian hierarchical models is provided below and a summary of peer reviewed dose-response models is presented in Table 4.1.

The Exponential Model

The single-hit family of dose-response models was described previously (Section 4.2.1) as the most relevant for microbial dose-response assessment. The simplest of the single-hit family of models is the exponential model (Equation 1).

$$p = 1 - e^{-r\mathrm{d}} \qquad \text{(Eq. 4.1)}$$

Where:

p is the cumulative probability of infection in the exposed population
d is the average pathogen dose in infectious units (organisms)
r is the probability of infection given ingestion of one organism

For the exponential model, *r* is a constant for the interaction of any given pathogen and host species. That is, each infectious particle within each host is assumed to have the same probability of survival. There is no inter-individual variability in the host-pathogen interaction: the exponential model assumes that the same probability of infection applies to every individual in the population. Despite this unrealistic-sounding assumption, the exponential model provides a good fit for a number of the human pathogen-challenge data sets (Teunis et al., 1996; Haas et al., 1999). The primary advantage of the exponential model is its computational simplicity. The primary disadvantage is that is does not account for inter-individual variability in the population.

The beta-Poisson Model

The primary limitation of the exponential model (no variability in *r*) is partially[26] overcome using the beta-Poisson model, by assigning a distribution to *r* to represent the variability in the pathogen-host interaction. The most common distribution applied to *r* is the beta, giving rise to the beta-Poisson model (more strictly, the beta-exponential). As *r* is a probability itself, the assigned distribution must have a range of 0 to 1. The beta distribution is the most flexible of such distributions, including shapes similar to Gaussian (normal), triangular, exponential, power law, uniform, and bimodal. The unit infectivity *r*-value is the mean of the distribution, which is readily calculated from the parameters. The two-parameter beta-Poisson does not have a fixed slope and is more flexible than the exponential. The beta-Poisson still follows the rules (does not exceed the probability of exposure at low dose), but is more biologically plausible than the exponential and will fit better to data with higher variances. The beta-Poisson is a function of the confluent hypergeometric function, which is a sum of an infinite number of terms and has no simple closed-form mathematical representation (Abromowitz and Stegun, 1964). The beta-Poisson cumulative probability is given by Equation 2.

$$1 - \mathrm{M}(\alpha,\ \alpha+\beta,\ -\mathrm{d}) \qquad \text{(Eq. 4.2)}$$

[26] The distribution on *r* does not distinguish between pathogen and host variability.

where M is the confluent hypergeometric function (the ${}_1F_1$ form), α and β are the beta distribution parameters, and d is the mean dose in pathogen infectious units. The solution is estimated numerically.

The Pareto II distribution, commonly called the beta-Poisson in the literature, was shown by Furumoto and Mickey (1967) to approximate the exact theoretical beta-Poisson model, and has found wide usage. The Pareto II distribution function for dose response is given by Equation 3.

$$1 - (1 + d/\beta)^{-\alpha} \qquad \text{(Eq. 4.3)}$$

where d is the dose, and α and β are parameters corresponding to the beta distribution parameters for specific ranges, the approximate form being valid for parameter values $\beta >> 1$ and $\alpha << \beta$ (Teunis and Havelaar, 2000). Outside of this range, the Pareto II can substantially overestimate the risk, sometimes predicting a probability of infection greater than the probability of exposure at low doses (Teunis and Havelaar, 2000). The term "beta-Poisson" will be used, henceforth, with reference to the exact form, while the analytic approximation will be referred to as the Pareto II. Both forms have been used for the gastro-enteric infection endpoint (Haas et al., 1996; Teunis and Havelaar, 1999; Englehardt and Swartout, 2004). Thus, the beta-Poisson is more flexible than the exponential model while retaining simplicity.

Accounting for Immunity

Whatever model you use, the risk of infection applies only to the susceptible population. In fitting the model to a particular human pathogen-challenge study data set, it may be possible using available information to take into account the fraction of immune individuals, sometimes even if the participants were prescreened for presence of antibodies to the organism. The general fitting algorithm for assessing the fraction of immune individuals is given in Equation 4.

$$(1 - fr) * F(d, \boldsymbol{\theta}) \qquad \text{(Eq. 4.4)}$$

where fr is the fraction of resistant (immune) individuals[27], F is the dose-response function (e.g., exponential, beta-Poisson), d is the dose, and $\boldsymbol{\theta}$ is the parameter vector associated with F. If the subjects were screened for prior exposure, you should offer an explanation of the (unexpected) finding of a resistant fraction when using this model. Explanations could be based on theoretical considerations or experimental conditions specific to the case.

Deterministic Models

The foregoing hit-theory models are stochastic in nature, primarily in that each host may or may not become infected at any given pathogen dose. Models that are

[27] Alternatively, 1 – fr can be replaced by fs, the fraction of susceptibles.

deterministic in nature also have been proposed for microbial dose-response evaluation (Moon et al., 2005). These models are deterministic because they assume that each host has a unique tolerance, or threshold dose, above which infection is 100 percent certain, similar to chemical dose-response assessment. An advantage of these models is that in general, these models are more flexible than the hit-theory models and will tend to fit better to many data sets. A disadvantage is that they can result in over prediction of risk at low doses (dilutions of single organisms), because they do not recognize the discrete nature of pathogen distribution. Over prediction is likely to happen for dose-response data sets characterized by high variability, high response at lower doses, or slowly increasing responses across large dose ranges. Over prediction is particularly prevalent in modeling uncertainty in bootstrap simulations, for example. In addition, the biological plausibility of individual (deterministic) host thresholds has not been established for pathogens, as it has for chemicals. Therefore, at least for low-dose extrapolation (e.g., determination of unit infectivity), deterministic models are not recommended. However, for high-dose risk estimation, deterministic models can be useful because of their ability to fit the response data better than the one-hit models.

Bayesian Hierarchical Models

Bayesian methods to estimate dose-response model parameters and evaluate their uncertainty are also being used (Englehardt, 2004; Englehardt and Swartout, 2004; Messner et al., 2001). These methods are particularly useful in cases where data are available from multiple studies. One-stage or hierarchical models can be fit to the data using methods that include Markov Chain Monte Carlo Simulation (MCMC) (Gilks et al., 1996; FAO/WHO, 2003; EPA 1997b). A Markov chain is a stochastic model having discrete states in which the probability of being in any state at any time depends only on the state at the previous time and on the probability transition matrix. MCMC simulations can be used to generate samples from the joint posterior distribution (Messner et al., 2001). An advantage to these models, in contrast with traditional statistical techniques, is that they are able to exploit subjective and related information in addition to numeric data.

A predictive Bayesian dose-response function can be developed as follows. First, the parametric form of the dose-response function is established by theoretical derivation and, if possible, empirical confirmation. Then all available knowledge, other than the theoretical form of the conditional distribution and empirical data already used for that purpose, is brought to bear upon estimation of the parameters of the distribution. To do this, the parameters are recognized as uncertain but subject to professional judgment, and thus, a prior probability distribution is assigned to each parameter. Prior distributions are then refined with dose-response data, to obtain a posterior distribution. Next, the predictive Bayesian dose-response function can be found by multiplying the posterior by the conditional dose-response function, and integrating over the parameter space (Englehardt, 2004). Bootstrap methods (i.e., repetitive Monte Carlo sampling directly from the data or from data summary distributions) whether used in a Bayesian or frequentist framework, may also be used to evaluate parameter uncertainty in dose-response models (Teunis et al., 1996; Haas et al., 1999; Englehardt and Swartout, 2006).

These models can be more complicated than other models described here, are generally less familiar to scientists and managers, and can be difficult to explain.

Bayesian hierarchical models have also been used to develop dose-response relationships for pathogens based on outbreak data rather than feeding study data. For example, Teunis et al. (2005) analyzed *Campylobacter jejuni* dose response using Bayesian methods. Data from both a human volunteer study and an outbreak caused by drinking raw milk were combined in this analysis. The model incorporated both the probability of infection and the conditional probability of illness given infection. First, for the outbreak a certain probability of illness (p0) was assumed for those who were unexposed to the raw milk but might have become ill due to an alternative route of transmission. Second, a beta-Poisson model was used to model the probability of infection given a mean dose (D). Third, a model for the conditional probability of illness, given that the individual is infected and had mean dose D, was developed. Non-informative prior distributions for the parameters were defined. The posterior mode parameter values were calculated by directly maximizing the posterior probability. These values were used to compute the posterior mode dose-response functions for the probability of infection and the probability of illness given infection. Uncertainty intervals for these dose-response functions were computed by using MCMC to simulate vectors of parameter values. Teunis et al. (2008a) also analyzed data from eight outbreaks of *E. coli* O157:H7 using a hierarchical Bayes model and used Bayesian methods to analyze dose-response functions for the Norwalk virus, based on a volunteer study (2008b).

4.2.2 What is the Output of a Dose-Response Assessment?

The output of a dose-response assessment is a value or a set of values for the dose response parameters. For example, for the exponential dose-response model (described in section 4.2.1) a single value ("r") would be required and for dose-response models having more than one parameter, a set of parameter values will be required. For many of the most common dose response functions, the relationship between exposure and risk is linear at low doses (Haas et al., 1999). For exposures to many organisms at once (such as in food), the risk of infection needs to be calculated from the mathematical dose-response function itself.

Computing the risk of infection may be necessary to determine the population at risk for illness, but infection, in itself, is not necessarily adverse. Therefore, information on the rate of illness, given infection, is needed to perform a meaningful risk assessment. Although orally ingested pathogens can cause a number of symptoms, some severe enough to be life-threatening, the only endpoints examined in human studies (for ethical reasons) are related to gastrointestinal illness, primarily diarrhea. The risk of illness is estimated in a similar fashion as for infection, except that the dose-response model is constrained such that an illness response is strictly conditional on infection. The constraint is trivial for the one-parameter exponential model, as the slope is fixed. For models in which the slope can change (more than one parameter), a higher risk of illness than infection can be predicted for some data sets if the constraints are not strict. Illness

without infection is not biologically plausible so constraints are necessary to avoid this implausibility. Applying strict constraints in such models is not a trivial exercise, and is a topic of ongoing research. The output of an illness dose-response assessment generally is a morbidity ratio, which is the fraction of those who are infected that become ill. A common practice is to assume that the risk of illness is constant once an individual becomes infected, no matter what the dose. In this case, the morbidity ratio is simply the number of ill individuals divided by the number of infected individuals. Dose-dependent morbidity ratios, where the conditional (on infection) risk of illness increases with increasing dose, are more difficult to model, requiring the strictly constrained model previously discussed (Refer to Teunis et al., 2005). For drinking water risk assessments, where the exposures are frequently very low, the occasionally large difference between constant and dose-dependent morbidity ratios can be highly significant.

You should be transparent in your discussion of the dose-response output. For example the dose-response is based on a defined health endpoint, a defined human population (e.g., the population used in a clinical trial or an epidemiological study), and is influenced by model selection. Be clear about all the constraining features of the data and what assumptions were made when those data were used to extrapolate to broader human populations or health endpoints. For example, the risk managers who will use the risk assessment results to inform their decision making need to understand the implications of dose-response data based on healthy adults, when their goal is protection of the general population that includes all life stages and sensitive populations.

4.2.3 How do I Fit Models to Existing Dose-Response Data?

A summary of dose-response relationships published in the peer-reviewed literature for waterborne exposures is provided in Table 4.1. In many cases, you can simply select the most appropriate dose response relationship from one that has already been peer reviewed. If an appropriate dose-response relationship for the specific pathogen/matrix combination is not available, you can derive a relationship in several ways. If a valid dose-response data set is available, you should be able to fit the data to a mathematical dose-response model. The dose-response model provides a prediction of the incidence of the effect in the population, given a specific exposure level or dose. The dose-dependent probability distribution is defined by a mathematical equation with one or more parameters. The best values for those parameters are determined by assessing the likelihood of observing the data, given specific parameter values. The parameter values that result in the greatest likelihood of the data are chosen as the most representative ones. The determination of the best parameter values can be done directly on the data (as in a "frequentist" approach), or by updating a prior judgment as to what the parameters might be (a "Bayesian" approach). In either case, the resulting fitted dose-response model is used for predicting human response, either infection or illness, to pathogen exposure for a specific risk-assessment scenario.

The typical dose-response assessment is performed by a direct fit of the functional form to the data using a "frequentist" approach. That is, the fitted parameters represent the frequency with which some event has happened previously. The fitted parameters,

however, are generally used to predict the future occurrence of that event. Proponents of Bayesian methods argue that a Bayesian approach is a virtual requirement when trying to assess *probabilities* of future outcomes, rather than the *frequencies* of past events (Bernardo and Smith, 1994; Berry, 1996; Carlin and Louis, 2001). Bayesian methods provide a rigorous framework for common-sense interpretation of statistical conclusions. Frequentist approaches can only place confidence limits on a result that depends on specific conditions, leading to inferences that might be made in repeated practice. Bayesian proponents point out that most people erroneously interpret frequentist results in the Bayesian sense (Gelman et al., 2004). Bayesian methods generally provide much greater flexibility than do frequentist methods, allowing you to cope with very complex, data-limited problems.

Bayesian methods, however, are not without problems of their own. First, computation of the more complex integrals generally involves complex numerical techniques, with which many practitioners will not be familiar. Most analysts will have to rely on programs, which, in themselves, require some degree of mastery of functional coding techniques. Second, Bayes theorem requires that a prior relationship between dose and response be specified, which can be problematic for pathogens. The prior, which is usually subjective, can vary among investigators. Non-informative priors, generally uniform distributions over a wide range of parameter values, address the subjectivity issue but can make strong statements about prior belief of infectivity. An example of the latter and, perhaps, the only truly non-informative prior, would be the simplest prior on the *r* parameter for the exponential distribution (see Section 4.2.1) of a uniform distribution between 0 and 1 (the full range of *r*). Although this prior says that we really don't know anything *a priori* about infectivity, it establishes a prior expected value of 0.5 for *r*, which is much greater than any known actual pathogen infectivity. As the Bayesian posterior is a compromise between the prior and the data, a lot of data would be required to move the answer towards a less extreme value.

Examples of the use of Bayesian techniques in the literature include a number of analyses of *C. parvum* (Teunis et al., 1999; Messner et al., 2001; Englehardt and Swartout, 2004, 2006; EPA, 2006a). These analyses are largely hierarchical, assessing the aggregate infectivity of several isolates of *C. parvum* treating each one as a distinct strain. The outputs of most of these analyses consist of distributions of uncertainty for "hyperparameters" of *C. parvum* infectivity across strains. Such an analysis was used in support of the Long Term 2 Enhanced Surface Water Treatment Rule (LT2ESWTR) promulgated by EPA in 2006 (EPA, 2006a). One of the analyses did not assume that the *C. parvum* isolates were different, evaluating the aggregate illness-response data across all studies (Englehardt and Swartout, 2006). This analysis and one other (Englehardt and Swartout, 2004) used an unconditional Bayesian form, in which the posterior represented the dose-response function integrated over the entire range of parameter values. This unconditional or "predictive" form is independent of any pre-selected confidence level (e.g., 95 percent) and can be considered to be the expected value under uncertainty. All of the cited *C. parvum* assessments can be considered to be meta-analyses of sorts, as they incorporate data from more than one study into a single aggregate output. Meta-analysis generally allows for a weight-of-evidence approach, where a more objective

review of data may occur. Although none of these analyses did so, in a meta-analysis, individual data sets can be assigned specific weights (other than unity) prior to quantitative analysis. In this case, a Bayesian framework offers a rigorous approach in which to evaluate such weightings. Bayesian model averaging is an technique that formalizes this process (Hoeting et al., 1999).

4.2.4 How Can I Evaluate Uncertainty in Dose-Response?

It is possible to characterize the precision with which the dose response curve has been determined. The statistical confidence limits primarily reflect variability, with some (usually unquantifiable) contribution from uncertainty. Therefore the statistical confidence limits do not represent the full expression of uncertainty. For single hit models some of the uncertainty can be shown by determining the confidence limits to the parameters of the dose response curve and also the upper and lower envelope around the dose response curve (Haas et al., 1999). A likelihood-based approach can be used for this purpose. This yields an m-dimensional region, where m is the number of parameters in the dose response model under evaluation (one for the exponential, two for the beta Poisson, etc.). The computational details of this approach are beyond the scope of this document, but interested assessors are referred to Chapter 7 of Haas et al. (1999).

In hierarchical Bayesian approaches, the output of the uncertainty analysis can be in the form of distributions on the model parameters or "hyperparameters," such as the mean of the fitted beta distribution (the average unit infectivity) for the beta-Poisson dose-response model. A distribution of plausible bounds on the response at any dose can then be generated to obtain, for example, 95% confidence limits on the response at a given dose.

In the predictive Bayesian models, the parameter uncertainty is integrated with the response variability to obtain a single dose-response curve without confidence bounds, but whose shape depends on the amount of information available; the shape becomes narrower (less uncertain) as the amount of information increases (Englehardt, 2004), generally resulting in a prediction of lower mean risk than when information is sparse. Another technique is the parametric bootstrap analysis, in which the result of the initial dose-response model fit to the data is assumed to be the true response relationship. The responses at each dose are then regenerated repeatedly based on the respective fitted response probabilities assuming an underlying response distribution (generally binomial). A distribution of "resampled" responses is obtained for each dose, from which bootstrap confidence bounds on the entire dose-response curve can be calculated.

4.2.5 What is Variability in Dose-Response?

It is likely that all parameters in dose-response have some variability. Dose-response involves the interaction of host and pathogen, so any factors associated with either the host or the pathogen have variability and the interaction itself is also variable. The variability in factors presented in Section 4.1.3 may be quantitative or qualitative in nature. Depending on the needs of the particular risk assessment, you may need to use

statistical techniques to characterize variability in:

a) Dose-response relationships between isolates or strains

b) Host immune responses (both immunity and susceptibility)

c) Duration of host immune responses

d) Host characteristics that influence the dose-response relationship

e) Health effects

4.2.6 How Can I Account for Life Stages and Subpopulations in Dose-Response Models?

Unlike a subpopulation, a life stage is something the whole population passes through, such as childhood. At any given moment only part of the population is in childhood, but referring to children as a subpopulation underestimates the importance of childhood life stages in the larger picture. EPA breaks childhood down into ten age groups based on behavioral and physiological milestones (EPA, 2005; Section 3.7). Some life stages, such as children and the elderly, are more sensitive to pathogens because of host susceptibility and/or behavior patterns when compared to other life stages, such as adults. In chemical risk assessment, life stages and sensitive populations are frequently taken into account by the application of uncertainty factors. The use of uncertainty factors for pathogens, however, is somewhat problematic because of the discrete nature of exposure. Unlike for chemicals, pathogen dose is usually expressed in non-reducible organism-level units as an average, such that fractional doses represent probabilities of exposure to single organisms. Thus, when considering low-dose exposures, a dose of 10^{-4} for a chemical may be in units of milligrams or micrograms, representing perhaps 10^{18} molecules, while the same dose for a pathogen would represent a 1 in 10,000 chance of ingesting a single organism for a given exposure. Depending on relative infectivity of the pathogen, the indiscriminate application of a fixed uncertainty factor could result in an impossible risk (risk of infection greater than probability of exposure) or an arbitrarily high risk inconsistent with the overall exposed population. Accounting for sensitive life stages and populations for a low-dose exposure scenario (e.g., drinking water) would require an estimate of the relative infectivity for that subpopulation compared to the general population, as well as the fraction of that subpopulation with respect to the entire population. With this information, a new subpopulation infectivity parameter could be calculated as a weighted average. For example, simply stating that you believe that a particular life stage is 10 times more susceptible than the healthy adult population represented by the experimental data is not adequate, in itself. In this case, if the r-value for the healthy adult population is greater than 0.1, an impossible risk (> 1) is projected for the life stage. Each case is likely to require a unique solution, with professional judgment playing a large role. For some pathogens, the probability of infection or disease for sensitive life stages and populations may not differ from the normal population, but the severity of the disease outcome may differ.

4.2.7 Can I Use Uncertainty, Modifying, or Adjustment Factors in a Microbial Dose-Response Assessment?

There are no standard guidelines for the application of uncertainty, modifying, or adjustment factors in MRA as are in chemical risk assessment. In chemical risk assessments, uncertainty factors are usually applied as factors of 3 or 10, and are applied to empirically derived effect levels (e.g., no observed adverse effect level) to accommodate for a lack of knowledge associated with interspecies extrapolation, high- to low-dose extrapolation (i.e., effect to no-effect), population variation (i.e., protection of sensitive populations), and extrapolation across exposure durations (e.g., subchronic to chronic). The areas of uncertainty potentially most relevant to MRA are the interspecies, sensitive life stages, and sensitive population extrapolations. As an example, for interspecies extrapolation, there are sound allometric-scaling principles relating the uptake and whole-body distribution of chemicals and strongly conserved mechanisms of toxicity across mammalian species. There are no corresponding agreed upon principles for cross species pathogenicity relationships. Because many pathogens are highly species-specific or produce different effects in different species, and immune response mechanisms can be highly variable across species, use of uncertainty, modifying or adjustment factors to justify extrapolation is highly suspect.

For chemical risk assessment, all uncertainty factors are applied as a divisor of the dose to obtain a quasi-threshold exposure level. For MRA, the assessor would have to pay attention to the absolute value of the computed risks so that implausible or impossible risks would not arise (e.g., product of general population risk and uncertainty factor greater than 1). In essence, a variable-magnitude uncertainty factor scheme would be a likely result, with smaller uncertainty factors for larger average population risks. Furthermore, to obtain an overall population risk (adjusted for sensitive life stages and subpopulations), the assessor would have to know the proportion of the population that is sensitive (for proper weighting of each subpopulation-specific risk). An example of this process can be found in a human population infection-response assessment for *C. parvum*, in which human pathogen-challenge study data were combined probabilistically with assumptions about the size of sensitive and resistant populations to obtain an estimate of overall population response (Englehardt and Swartout, 2004).

Because of the foregoing considerations, it is not usual practice in MRA to use uncertainty or modifying factors in a manner similar to their use in chemical risk assessment. An important distinction between uncertainty factors and adjustment factors is that uncertainty factors account for unknown distributions of sensitivity and adjustment factors account for known differences in response. Case-specific adjustment factors can be employed if strong defensible evidence supports their use. As an example, epidemiological data was used as a justification for the application of adjustment factors in the FDA/USDA/CDC risk assessment for *L. monocytogenes* in ready-to-eat foods. Scaling factors were used to adjust mouse-derived dose-response curves so they were applicable to humans (FDA/USDA/CDC, 2003). The size of the scaling factor was

determined by surveillance data reported to FoodNet[28] for the receptor populations modeled in the risk assessment. Similarly, in the FDA risk assessment on *Vibrio parahaemolyticus*, the dose-response curve was adjusted to reflect CDC's illness estimate. The adjustment factor represents the effect of the apparent differences between the dose-response observed in human volunteers under controlled conditions versus that in the general population when exposure is associated with the oyster food matrix.

4.2.8 What Modeling Methods are on the Horizon?

Physiologically-Based Dose-Response Models

New physiologically based models are being developed to begin to capture the biological complexity associated with dose-response (Blaser and Kirschner, 1999, 2007). Physiologic models begin with the development of conceptual models that break the process from exposure to establishment of infection to the expression of illness into compartments. These compartments serve as "steps" in the process, and the parameters inside each step are captured in the model. Published research and/or new research provide or will provide the values for each parameter. This is one of the strengths of the risk analysis concept, the clear identification of data needs, which can then been translated into research priorities.

[28] http://www.cdc.gov/FoodNet/

5. EXPOSURE ASSESSMENT

The goal of exposure assessment in MRA is to determine the route, frequency, duration, and magnitude (amount) of exposure to a microbial hazard in a population.

Microbial agents may come from more than one source, may be transmitted via multiple routes of exposure, and may be spread via secondary transmission. Moreover, these routes of exposure may be inter-related. Exposure routes relevant to a given microbial hazard are situation-dependent and influenced by the inherent properties of the microorganism and its potential host(s). An exposure source can originate from either natural or anthropogenic events, activities, or locations that generate or release microbial hazards.

A number of factors define microbial exposure including the sources and pathways of exposure, the growth and/or decline in numbers of microorganisms and variable intake amounts among individuals. Often, some of the necessary data for a microbial exposure assessment are either lacking (i.e., need to be extrapolated from data developed for another purpose or limited data that are not representative) or altogether non-existent. Given that complete data and information are rarely available for microbial exposure assessment, you may need to make simplifying assumptions. Such assumptions result in uncertainty about exposure estimates. To support better risk management decisions, you need to objectively characterize uncertainty about exposure assessment results.

This chapter provides general principles and practical guidance for conducting exposure assessments for microbial hazards. Information is organized into five sections: 1) general concepts in exposure assessment (Section 5.1); 2) developing an exposure assessment (Section 5.2); 3) analyzing results from a model (Section 5.3); 4) communication, review and validation of model results (Section 5.4); and, 5) future developments in exposure assessment (Section 5.5). The chapter is also intended to provide information useful to risk assessors, risk managers, decision-makers, risk communicators, stakeholders and the general public, and researchers. Other resources that provide overviews of exposure assessment are those by the WHO/FAO (2008), European Commission Scientific Steering Committee (ECSSC, 2003), Hass (1999), Cox (2006), and Vose (2008).

5.1 What are General Concepts in Exposure Assessment?

This section includes factors to take into account when conducting an exposure assessment. It also discusses variability, uncertainty, deterministic and stochastic risk assessment, and Monte Carlo analysis.

5.1.1 What is an Exposure Assessment?

An exposure assessment is the process of estimating or measuring the magnitude, frequency, and duration of exposure to a microbial hazard(s), along with the number and

characteristics of the person or population exposed. You can provide either a qualitative or quantitative evaluation of exposure, however a quantitative evaluation is always preferable if the data exist and a quantitative risk assessment is needed. Ideally, an exposure assessment describes the sources, pathways, routes, and the uncertainties about exposures.

5.1.2 What is Exposure?

Exposure comprises the sources, mode, route, and extent of contact the host has with the microbial hazard(s) of concern. How often a person is exposed is referred to as frequency of exposure. How long a person is exposed to a microbial hazard is referred to as the duration of exposure.

MRA is typically only concerned with single event exposures. But reoccurring exposures can be included, particularly if secondary transmission is being modeled. The number of microorganisms that correspond to a single exposure is referred to as the dose. The exposure dose may also be the total number of organisms that constitute a set of exposures. Simultaneous exposure to multiple hazards is also a concern in MRA.

5.1.3 What are Sources, Pathways, and Routes of Exposure?

There are various terms used to discuss the origin, movement or spread, and final intake of microorganisms by individuals or populations. Generally, the overall terminology refers to routes of transmission for microorganisms.

The entity (or entities) that supply microorganisms to a particular exposure route is the source. The source of microorganisms could be infected food animals, industrial processes, the environment (water, air, soil), or infected persons. The route of exposure (or route of intake) is where the microorganism comes into contact with the host. The three common routes of exposure are oral, inhalation, and dermal.

The physical movement of microorganisms, over time, from their source to the occurrence of an exposure is referred to as the exposure pathway or route of transmission. Exposure pathways may be complex; exposures may occur via aerosolization, water, food, soil, fecal-oral and/or inanimate sources. The mode of transmission can be wind, flowing water, equipment movement, or vector organism. Not all modes of transmission are relevant for all exposure pathways. For example, neither inhalation via the nose nor dermal exposure are highly relevant for foodborne exposures. The number of microorganisms in a particular medium can increase or decrease across time as a function of changing environmental conditions, throughout the exposure pathway.

Elements of Source Evaluation

While sources of microorganisms may be living or inanimate, the elements of source evaluation are basically the same, again with the caveat that not all modes of

transmission are relevant for all exposure pathways. In a farm to fork model you can consider:

a) How many viable pathogens (or indicators) are present at the source (e.g. infected chicken, contaminated carcass) at time zero?

b) How many are released from the source?

c) Over what time period are they released?

    1) Continuously

    2) Batchwise

d) At what rate are they released?

    1) Counts/unit time (e.g., cfu, pfu, genomes per minutes, seconds, hours, days)

e) What is the form of the release?

    1) Fomites

    2) Spray equipment

    3) Offgases from a fermentor

    4) Waste water

    5) Animal slaughter

f) To what medium are they released

    1) Food

    2) Surface water

    3) Soil

    4) Air

    5) Other surfaces

The evaluation of movement from a source into an exposure pathway is sometimes called a release assessment. The purpose of the environmental release assessment is to identify the sources of potential release, the media of release (air, water, or land) and the magnitude and frequency of release. The release estimates then serve as inputs to the assessment of survival and distribution subsequent to release.

When there are data available to predict releases, four main steps are used in constructing the release assessment.

a) First, you should collect and synthesize information on how many organisms are generated at the site of release. For example for land application of biosolids, this would include the size of each load, the number of loads per unit time, and the concentration of organisms in a load.

b) Second, develop a process description to locate the places where releases may occur. For an industrial process one needs to know where and how the microorganisms are grown and how they are separated from their growth medium. The process description should also consider the manner in which microorganisms may be subjected to circumstances such that they may be inactivated or destroyed.

c) Third, relate each possible point of release to the process involved. In agriculture this could be a drop spreader or spray nozzle. In an industrial plant it could be off-gassing (during separation, as from a centrifuge), from equipment during clean-up, or during product transportation.

d) Fourth, for each release source develop quantitative estimates of release which specify the amount of release, the time frame of release, and the media of release. If inactivation procedures or engineering controls are applied to the release source, then their effectiveness will need to be estimated to quantify the amount released after the control or treatment.

When intentional or incidental releases occur from inanimate sources, quantitative estimates can frequently be obtained. Releases to air from sources such as a fermentor's off-gas can be measured as the viable count per site per unit time. You can treat these as point source releases occurring at approximately rooftop height. A similar approach can be employed for wastewater releases. Modeling of release modes is usually medium specific; the output may be useful in further estimating dispersal of the microorganisms from their source. Incidental releases may be modeled based on empiric evidence compiled for specific activities, but source evaluations of intentional releases to the environment are often complex and case specific.

The dynamic nature of microorganisms is one characteristic that differentiates microbial exposure assessment from chemical exposure assessment. Predicting changes in the number of microorganisms along an exposure pathway is often necessary to accurately estimate exposure doses. Environmental conditions that can influence the growth and decline in the number of microorganisms present in a specific media include, but are not limited to:

a) water activity,

b) pH,

c) carbon source,

d) electron acceptor,

e) sunlight intensity,

f) temperature,

g) population density of the microorganisms and/or other microbiota that compete for nutrients in the media or support pathogen growth e.g. within biofilms on surfaces, and/or

h) presence of disinfectants or antimicrobials.

Depending on the characteristics of the microorganisms, some survive throughout an exposure pathway while others do not.

Many microorganisms have more than one exposure pathway and corresponding route of intake, often referred to as an exposure route or route of entry. Common exposure routes are:

a) inhalation (nose and mouth to lungs),

b) ingestion (oral intake of food, water, soil, and inanimate objects), and

c) direct (via skin, eyes, ears, inanimate objects, and sexual contact).

Some microbial exposure assessments are able to characterize exposure via a primary exposure pathway (e.g., *Escherichia coli* O157:H7 from infected cattle to an individual via undercooked ground beef). Nevertheless, microbial and epidemiological evidence may indicate that some microorganisms spread via cross-contamination pathways; these pathways are sometimes difficult to characterize. Also, many microorganisms (e.g., noroviruses) are spread from an infected person to another who is not. This exposure pathway may be poorly characterized because sufficiently detailed data are often lacking (Zhao, 1998).

In addition to ingestion routes, chemical risk assessment is well developed for inhalation and dermal exposure routes. For example EPA has many guidance document for chemicals such as, *Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry*, the *Exposure Factors Handbook*, and *Risk Assessment Guidance for Superfund Volume I: Human Health Evaluation Manual (Part E, Supplemental Guidance for Dermal Risk Assessment) Final* (RAGS-E) (EPA, 1994, 1997a, 2004d). Inhalation and direct (e.g., dermal) exposure routes and to what degree chemical guidance document might be adapted for MRA may be further discussed in future volumes of this Guideline.

5.1.4 What Environmental Factors Can I Take into Consideration?

Environmental factors are considered when the risk assessment is at the stage of calculating the amount of pathogen that constitutes an exposure. Examples of environmental factors are provided below with a short description.

a) **Ecological Niche** – An ecological niche is an n-dimensional hyperspace[29] where the abiotic and biotic elements in the environment allow a species to survive. As an ecologic niche changes, the amount of pathogen might fluctuate and/or the likelihood of exposure might increase or decrease.

b) **Gradients of Concentrations** – It is rare that microorganisms are equally distributed throughout a medium, therefore gradations of concentrations can be considered in accordance to the media being assessed, and the exposure being characterized.

c) **Persistence** – Persistence in the environment of an organism (in some form i.e., spores) can be considered. Those organisms that are not stable in the environment would pose a different exposure, and subsequent risk, than those that are stable. (See Chapter 3)

d) **Matrix Characteristics** – The characteristics of the matrix where the pathogen is found may determine the amount and state (e.g., dormant, alive, dead) of microorganism available to a receptor. Things like oxygen content, fat content, pH, temperature, water content influence the survivability of microorganisms.

5.1.5 What is an Exposure Scenario?

An exposure scenario (or hazardous event) is the set of conditions or assumptions about sources, exposure pathways, amounts or concentrations of microorganisms, and the characteristics of the exposed individual, population, or subpopulation that constitute one or more exposures. An exposure assessment may comprise many different scenarios. Each scenario is the basis for evaluation and quantification of exposure(s) in a given situation.

An analysis of "what if" options for mitigation measures, interventions, or policy changes is often referred to as a scenario analysis. This type of analysis allows you to evaluate the public or environmental health benefits of various measures that prevent or mitigate exposures.

5.1.6 What are Qualitative and Quantitative Exposure Assessments?

A qualitative exposure assessment is based on data and information which, when considered along with expert knowledge and identification of attendant uncertainties, provides a characterization of exposure in descriptive terms (e.g., high, medium, or low). You will need to conduct qualitative exposure assessment when there is not sufficient numerical data to develop a quantitative exposure assessment.

A quantitative exposure assessment provides numerical expressions of exposure. Such an assessment provides numerical estimates of the likelihood of different microbial

[29] N-dimensional space that has more than 3 dimensions can be called hyperspace.

dose amounts, as well as numerical measures of confidence about its estimates (i.e., uncertainty).

Sometimes multiple quantitative exposure assessments are conducted for a microbial hazard in order to rank sources, vehicles, and/or pathways of exposure based on risk. One example of such an approach is a risk assessment of *Listeria monocytogenes* in ready-to-eat foods (FDA/USDA/CDC, 2003).

5.1.7 What is Variability in Exposure Assessment?

Variability describes a range of possible events. It is purely the effect of chance; it can only be altered by changing the chance of something occurring. Variability in exposure can be caused by differences in location, activity, and/or behavior of exposed individuals at a particular point in time. These sources of variation result in differences in exposure to a microbial hazard(s) in various media. Variability is also caused by differences in the initial occurrence of microorganisms in various media (e.g., air, soil, food, and water). Because microorganisms grow and decline within media along the exposure pathway, there is variability in the amount of microbial hazard per unit of media intake by an individual or sub-population. You may need to characterize variability in:

a) The number of microorganisms initially present in the medium

b) The environmental conditions in which microorganisms exist

c) The processes through which microorganisms move within scenarios

d) The dose of microorganisms per unit of intake (e.g., serving of food, inhalation unit, amount of water ingested; spatial and temporal variability)

e) The amount of intake (inter-individual variability in exposure)

f) Exposures across time (temporal variability)

g) Exposures across geographic location (spatial variability)

The types of variability considered depend on the type of exposure assessment to be developed as part of the overall risk assessment.

5.1.8 What is Uncertainty in Exposure Assessment?

Uncertainty is imperfect knowledge. You can reduce uncertainty by further study or by accumulating more information. Uncertainty may reflect imperfect knowledge of the microbial hazard (e.g., its virulence), environmental pathway/processes, or the human populations under consideration. Sources of uncertainty fit into two broad categories:

a) Uncertainty regarding one or more parameters in an exposure assessment (parameter uncertainty)

b) Uncertainty as a result of incomplete information or scientific theory needed to fully define the causal bases of exposures (structural model uncertainty)

Availability and quality of data and information can reduce the amount of uncertainty in exposure estimates. Objective depictions of uncertainty improve the transparency of information used by decision-makers in managing risk. The process of interpreting the influence of uncertainty on the results of an exposure assessment is referred to as uncertainty analysis.

5.1.9 What is a Deterministic Exposure Assessment?

You can conduct a quantitative exposure assessment using "most likely" or "conservative" values for the variables and uncertain parameters included in the set of scenarios. However, unless data are not available, the use of single point estimates is not a preferred approach to inform decision making (OMB, 2007b; EPA, 2002b). These values are often referred to as point estimates and can result from collapsing the variability and/or uncertainty about random variables or parameters into singular values. Depending on how the point estimates are designed, the results may either represent an average or other extreme exposures (e.g., 95th percentile) among a specified population.

The use of point estimates in an exposure assessment is referred to as deterministic (static) modeling. Point estimates do not account for variability in the occurrence of the microorganisms at the source, variability in growth and/or decline in the number of microorganisms through the exposure pathway, or variability in intake across the population of individuals exposed to the microorganisms. Furthermore, a deterministic exposure assessment does not explicitly characterize the uncertainty about exposures. Without explicit characterization of variability and uncertainty, it is possible that point estimates will substantially over- or under-estimate exposures. If highly conservative point estimates – thought to be protective of public health – are used, the deterministic results may be characterized as worst-case estimates.

In some cases, deterministic modeling may be used to simplify the modeling of a highly complex system. For example, extensive modeling of transmission processes among a population may require simplifying assumptions about contact rates and transmission coefficients.

Another use of deterministic modeling is during your initial analysis of an exposure assessment model. Propagating simple numbers through the model may help with error-checking the mathematics of the model. Also, such calculations can provide early indications of the importance of various model components or pathways. Nevertheless, conclusions from such screening analyses should be cautiously interpreted because omission of variability can generate misleading results.

5.1.10 What is a Stochastic Exposure Assessment?

In contrast to using point estimates, the use of probability distributions for each parameter in an exposure assessment is preferred. A probability distribution includes both a range of values and the likelihood of occurrence for each numerical value. Use of probability distributions throughout the exposure assessment allows you to represent variability in exposures of individuals and/or sub-populations. You can also use probability distributions to characterize the uncertainty in exposure assessments. When developing a stochastic model, you can use point estimates to verify the mathematical formula or confirm that the computer code actually performs correctly for trivial cases (for example, the microbes are not present in the output if the initial concentration is zero).

The use of probability distributions in an exposure assessment is referred to as stochastic modeling. Probability distributions represent the variability and uncertainty inherent in a system. Stochastic modeling can provide more realistic results by accurately characterizing the impacts of known sources of variability and uncertainty on risk estimates. Risk assessments are often concerned with the occurrence of rare events and stochastic modeling may reveal rare but consequential results (e.g., the occurrence of an unlikely, but large, population outbreak.)

Stochastic models often use computer simulations to mathematically combine multiple probability distributions in an exposure assessment calculation. Monte Carlo analysis (more detail in next section) is the most widely used probabilistic method to estimate these combinations. Advanced Monte Carlo modeling techniques can also quantitatively characterize uncertainty in exposure estimates (Gilks et al., 1996).

Stochastic modeling is usually more resource-intensive than deterministic analysis. Defining model inputs as probability distributions can require additional steps in the planning, review, and communication of the exposure estimates. Use of probability distributions, however, provides a framework for incorporating more of the available information into an exposure assessment.

If data for critical variables and parameters are available, then you should consider stochastic modeling for the exposure assessment if time permits. Alternatively, if data and information are insufficient, consideration should be given to the use of other modeling techniques. For example, the use of interval mathematics or fuzzy mathematics may provide a more credible assessment of the probabilistic boundaries of exposure than standard methods using probability distributions (Ferson, 1996).

5.1.11 What is Monte Carlo Analysis?

Monte Carlo analysis is a commonly used quantitative technique for exposure assessments. It involves the random sampling of each of the probability distributions in a model to estimate the likelihood of the model's possible results (Vose, 2008). Each re-

calculation of the model is termed an iteration, and a set of iterations constitutes a simulation.

A cardinal rule of this analytical technique is that every iteration should be possible in nature (Vose, 2008). If followed, this rule can prevent errors in modeling logic. For example, a predicted serving cannot contain 2.7 microorganisms, although the average concentration across some volume or mass may be 2.7 microorganisms. While it is not possible that any individual would be exposed to exactly 2.7 (viable) organisms, the simulation can be acceptable if the exposure distribution is defined and sampled correctly. If a Poisson (discrete) distribution with a mean of 2.7 is used in the risk modeling, the individual Monte Carlo iterations will all take discrete (integer) values, maintaining the realism of each iteration and of the simulation as a whole.

You may face three common problems when using Monte Carlo methods:

a) First, correlations and dependencies between variables may be unknown. If dependent variables are mistakenly assumed to be independent in a Monte Carlo analysis, the likelihood of common occurrences in the real world may not be correctly estimated via simulation. If information on the correlations is not available, then alternative methods (e.g., interval or fuzzy mathematics) may avoid such mistakes because the probability boundaries calculated by these methods can include a full range of correlations in their results (Ferson, 1996).

b) Second, the data necessary to estimate input distributions may be incomplete or entirely lacking. Although inadequate data are a problem for any exposure assessment method, Monte Carlo methods are particularly disadvantaged because these methods require explicit definition of the model inputs. Sometimes uniform or triangular distributions are used when data are sparse.

c) Third, the mathematical structure of the exposure assessment model may be questionable. Risk analysts often acknowledge the limitations induced by these problems and employ sensitivity analysis (or other methods) to assess their influence on estimated exposures (Law and Keaton, 2000; Vose, 2008). Care should be taken to make sure that uncertainties related to model specification are addressed by comparing the quality of fits across different model forms. Where sufficient data are available, methods such as cross-validation may be used. Using this approach, an exposure model is estimated using a portion of the data (usually about 70 percent) and then the model is tested for consistency with the remaining data set.

Examples of Agency guidance on probabilistic risk assessment include, EPA's *Guiding Principles for Monte Carlo Analysis* (EPA, 1997b) and EPA's *Using Probabilistic Methods to Enhance the Role of Risk Analysis in Decision-Making With Case Study Examples* (EPA, 2009b).

5.1.12 How does Exposure Assessment Fit with the Other Components of Risk Assessment?

Fundamentally, risk assessment is a predictive analysis. It intends to "envision how the future will turn out if we undertake a course of action…" (Kaplan and Garrick, 1981).

Predictions are accomplished by answering three questions:

a) What can go wrong?

b) How likely is that to happen?

c) What are the consequences given that it does happen?

The set of answers to the first question outlines the mutually exclusive scenarios to be considered in an exposure assessment. These scenarios are given the symbol s_i where the index i implies there are potentially many, mutually exclusive, scenarios wherein something goes wrong ($i = 1$ *to* N). Each s_i that is identified has an associated probability that it occurs (l_i), as well as some measurement of its consequence (x_i).

To a large extent, planning and scoping and hazard identification will determine the scenarios to consider for the exposure assessment. It is imperative that hazard identification establish the biologic plausibility of causative mechanisms considered in any exposure assessment.

Exposure assessment, therefore, is most invested in determining the likelihood of scenarios and a provisional consequence of the scenarios. In the context of exposure assessment, the provisional consequence is the magnitude of human exposure (usually dose) that occurs at the end of the scenario.

The output of the exposure assessment is commonly combined with the dose-response relationship developed in hazard characterization to predict the probability of an adverse human health outcome. This combination is considered in the risk characterization chapter (Chapter 6).

You can order the scenario triplets from smallest to largest microbial dose (i.e., $x_1 \leq x_2 \leq ... \leq x_N$) and similarly align the scenarios and likelihoods for each of those doses. This organization conveys the likelihood of increasingly larger microbial doses, as well as the scenarios that are responsible for those doses. Likelihood is a number constrained between zero and one. Because the $s_1, s_2, ...s_N$ are defined as mutually exclusive[30] scenarios, it is also true that

[30] Note that each scenario may include exposures through more than one pathway. Each scenario includes defined combinations of exposures that are sufficiently different to justify separate consideration.

$$\sum_{i=1}^{N} l_i \leq 1 \qquad \text{(Eq. 5.1)}$$

(if all s_i represent an exhaustive list of the things that can happen, then this sum will exactly equal one).

In some exposure assessments, l_i will refer to the fraction of all possible exposures – including exposures with a dose of zero – that derive from scenario s_i and generate a microbial dose of x_i. If you know the total exposures for the microbial hazard that can occur in one year (M), then the frequency of microbial dose x_i is $F(x_i) = M \times l_i$ (i.e., the number of exposures with a dose of x_i in one year is the product of total exposures and likelihood).

Given the frequency of microbial doses, the exposure assessment can directly generate a frequency distribution for human exposures (Figure 5.1). This distribution is simply a graphic display of the paired $\{x_i, F(x_i)\}$ values.

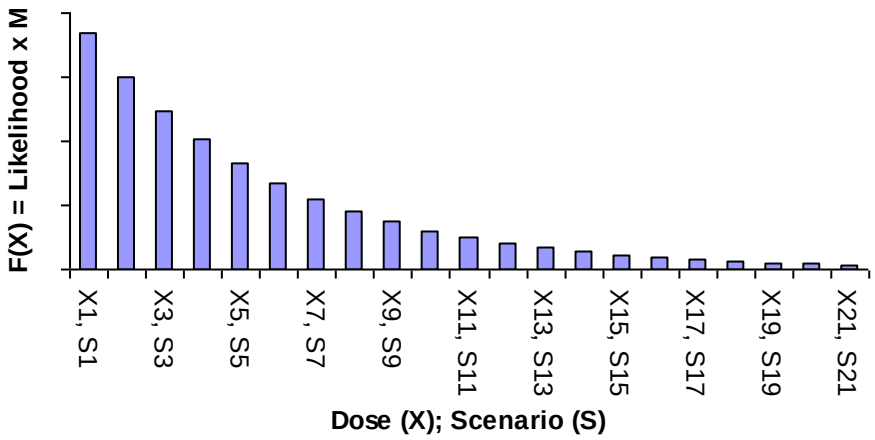

Figure 5.1 An illustrative exposure distribution developed by ordering scenarios (s_i) by the size of dose (x_i) and calculating frequency as the product of total exposures (M) and likelihood of the scenario (l_i).

5.1.13 Do Different Exposure Scenarios Always Generate Different Microbial Doses?

Although exposure scenarios may be mutually exclusive (i.e., represent unique pathways for microbial exposure), it is not necessary for different scenarios to result in different magnitudes of exposures. For example, you might consider consumption of water from a single source to which a treatment, with variable effectiveness, is applied. One scenario might reflect highly effective water treatment and consumption of a single 12 ounce serving that contains no microorganisms. Another scenario might reflect less effective water treatment and consumption of a serving of 6 ounces that contains no

microorganisms. Although the scenarios are different, the resulting dose (0 microorganisms) is the same.

The process of developing an exposure assessment can be complicated and it is not always understood, before the analysis begins, what scenarios will generate what doses. It is possible to design the exposure assessment such that scenarios are grouped by the dose they generate. Alternatively, the scenarios can be described *a priori* and the exposure doses they generate subsequently determined analytically. Depending on how scenarios are defined, it is possible that one exposure scenario may be associated with multiple doses (and their attendant likelihoods).

Consider a scenario wherein a hamburger patty – initially contaminated with 100 *E. coli* O157:H7 organisms and stored at 60˚F for 24 hours, then cooked at 130˚F for 4 minutes – is consumed by a healthy 30-year old male. The likelihood of this scenario might be 0.00089 percent of all servings of ground beef consumed by such a person. The consequence, from an exposure perspective, might be that 1000 *E. coli* O157:H7 organisms are ingested. This scenario, when accompanied by all other scenarios for hamburger patties, is useful because it provides sufficient detail for decision-makers to appreciate the risk of adverse human health outcomes from consuming hamburger patties (especially once the risk characterization is completed). Ultimately, the purpose of risk assessment is to support decisions regarding risk and those decisions typically hinge on the three elements contained here:

a) What are the substantial scenarios?

b) What are the magnitudes of their consequences?

c) How likely are such scenarios to occur?

Short contemplation of this example, however, will undoubtedly raise many questions. How was the scenario identified; surely there are thousands of scenarios at that level of detail (e.g., what about a hamburger that is cooked at 129 degrees Fahrenheit [°F] for 2.5 minutes)? How was the likelihood determined? That likelihood seems very precise; do all servings of hamburger from this scenario have the same likelihood of occurrence? Why does an exposure of 1,000 microorganisms result from this scenario? Do all hamburgers handled this way have the same number of microorganisms at consumption? Are you really this certain about all of this? Are there parts of the scenario that are more influential on the consequence than other parts?

The following sections of this chapter intend to outline general answers to these questions by discussing the process of developing, analyzing, and reporting an exposure assessment. These sections address scenario development, calculation of the likelihoods of scenarios, predictions of exposure doses, uncertainty analysis, and interpretation of exposure assessment results. Nevertheless, the ideas and concepts in these sections are not intended to be dogmatic prescriptions for completing an exposure assessment. In exposure assessment, there are many different approaches that may be valid for solving a

problem. Yet, all valid approaches share some fundamental similarities. It is these similarities that are the focus in the remainder of this chapter.

The concepts of scenario, likelihood, and consequence are fundamental to developing an exposure assessment. When analyzing an exposure assessment, the concepts of variability and uncertainty are crucial. Finally, effective communication and validation are essential considerations when reporting the findings of an exposure assessment.

5.2 How do I Develop an Exposure Assessment?

As discussed in Chapters 1 and 2, the beginning of the exposure assessment starts in planning and scoping. This important groundwork lays the foundation for a successful exposure assessment.

In general, the exposure assessment should be parsimonious, that is, as simple as possible while still including the important sources and steps leading to the exposure of concern. Based on "problem formulation" (see Section 2.1.1), you should make decisions regarding the approach to exposure assessment (e.g., attribution modeling or process modeling, empirical modeling of epidemiological data, probabilistic or deterministic, dynamic or static) and structure of the assessment model (which pathways, single or multiple models) (Hurd and Kaneene, 1993).

Given the complexity of many exposure assessments, the process becomes a multi-disciplinary collaborative effort. Subject matter experts are regularly consulted and their judgment incorporated into the process.

Conceiving an exposure assessment can be daunting. For this reason, some structure is needed. It is useful to begin by describing a conceptual model with all the necessary scenarios, followed by a full mathematical development of the conceptual model and, finally, by collection and analysis of data necessary to inform the model inputs identified in the mathematical model.[31] This structure theoretically ensures adequate attention to, and scrutiny of, the exposure assessment in an ordered and efficient manner.

Efficient exposure assessment is enhanced when precedents exist and are used. For example, beginning with already published conceptual models that require minor modification for a new application avoids unnecessary duplication of effort. Following reasonable precedents is also how standard methods can evolve.

5.2.1 What is the Purpose of the Risk Assessment?

With respect to purpose, most risk assessments can be categorized into two broad categories – retrospective or prospective. A retrospective purpose applies to microbial

[31] Note that this conceptual model may be software driven and is different than the conceptual model that is developed in planning and scoping which illustrates the broad overview of how risk happens.

hazards that are well-established as occurring sporadically or epidemically. A prospective purpose for a risk assessment applies to potential microbial hazards for which the adverse human health effects are not established. This categorization scheme is important because a different series of questions should be considered prior to developing the exposure assessment depending on the purpose category (see Table 5.1). These questions are not exhaustive, but the answers will guide the development of an exposure assessment appropriate for informing specific risk management decisions. In addition, answering these questions may also identify information requirements and methods for collecting this information.

Table 5.1 General Questions Considered Prior to Conducting an Exposure Assessment

Prospective Purpose	Retrospective Purpose
Is disease onset only a potential at this point, with the luxury of time to provide an answer?	Is disease onset imminent or already occurring, thus requiring an immediate answer?
Should the exposure assessment be structured as an in-depth analysis using less conservative assumptions?	Should the exposure assessment be structured more as a screening analysis, using default and/or more conservative assumptions?
Should the evaluation focus on both long-term and short-term exposures?	Should the evaluation focus only on short-term exposures?
Should the analysis focus on both low-level and high-level exposures?	Should the analysis focus only on high-level exposures?
Should attempts be made to measure (sample) or model exposures within the body?	Should concentrations be measured (sampled) or modeled in the media of concern?
Should the evaluation consider all potential exposure pathways for that particular microbial agent?	Should the evaluation focus only on those exposure pathways of imminent concern?
Should the analysis attempt to consider aggregate and/or cumulative exposures to multiple microbial agents?	Should the analysis focus only on the microbial agent causing the adverse health impact and only this exposure?

5.2.2 Which Scenarios Can I Consider?

Scenario development is the conceptual and creative part of exposure modeling. It melds considerations of purpose and scope with established or putative causal pathways. Although standardized frameworks for some microbial exposure assessments have been suggested (e.g., for food safety applications, see Nauta, 2002), there are few hard and fast rules for how or what scenarios should be considered in an exposure assessment.

Despite the lack of standard scenarios, those included in a specific exposure assessment should be clearly communicated and understood. Explicit diagrams (e.g., conceptual model) can be developed early in a project and these diagrams can include detailed descriptions of the inputs, parameters, flow and relationships of these components in the exposure assessment. It is useful to establish meaningful symbols within the diagrams to represent these model components early in the project. Consistent

use of symbols will encourage clear communication among risk assessors and risk managers.

Detailed diagrams with consistent symbols can be discussed with risk managers early and often in an exposure assessment project. This approach encourages open and clear communication of the modeling approach. Exposure assessment is an iterative and collaborative process; clear descriptions of the model with constructive feedback from persons outside the project will facilitate improvements at the conceptual model stage.

Exposure assessments should be parsimonious (simple as possible, but not simpler). Although hazard identification may suggest that the chain of causation for exposure is complex, the exposure assessment should only incorporate the complexity needed for the purposes of the risk assessment. In other words, risk assessments should leave avenues unexplored that, although interesting, do not specifically address risk management options. For example, the exploration of unproven causal relationships is rarely useful in exposure assessments.

Conceptually, an exposure assessment begins by considering the occurrence of the target microorganism at some place and/or time (i.e., its source). It is plausible that an exposure assessment could begin by characterizing the distribution of doses (or concentration of microorganisms) at the time of exposure. Such a beginning would not include consideration of sources and processes that produced the exposures. Nevertheless, if the purpose of the risk assessment was investigative and risk managers strictly sought estimates of the potential adverse human health events that could occur, then this approach would be satisfactory. Of course, it would only work if data are available to estimate the distribution of doses just prior to exposure.

Often exposure assessments begin by considering the occurrence of microorganisms at a place and time that is somewhat distant and prior to the actual human exposures. Availability of microbial occurrence data is one justification for where and when to begin the conceptual model. For example, if the only microbial data available refers to its occurrence prior to the application of some treatment process, then the exposure assessment may begin at that place and time. The purpose of the risk assessment is another justification for where and when to begin the conceptual model. For example, if the purpose is to set a regulatory performance measure for some treatment process, then the exposure model will need to begin its considerations of microbial occurrence at some point prior to the treatment process.

Once the beginning of the exposure assessment is determined, you can use predictive microbiology to determine how microbial occurrence changes before a dose reaches an exposed human. Processes refer to events or phenomena that influence microbial occurrence between the beginning and end of the exposure assessment. As discussed later, generic processes include growth, attenuation, mixing and partitioning. Which and how many of these processes are included in an exposure assessment depends on the planning and scoping and hazard identification stages of the project.

Many microbial exposure assessments involving food or water will include three general sequential stages: bulk processing, bulk transport, and consumption (Haas et al., 1999). Including these stages is often necessary to examine the influence of factors (or covariates) on microbial levels at the point of exposure. This inclusion is especially important when the purpose of the risk assessment is to predict changes in risk from one or more changes in these stages. For example, a risk assessment that examines a policy to require more controlled refrigeration of a food after it is produced – to limit growth of the target microbe – will include the bulk transport stage in the exposure assessment so that the effect of the policy relative to current conditions is measurable.

To complete scenario development, you should determine how each exposed population will come in contact with the microorganism of concern. The three well-recognized routes of initial exposure are inhalation, ingestion, and direct contact. These routes will influence the mathematical scale of the model. For example, inhalation exposures will likely depend on concentration measures of microorganisms (e.g., microorganisms per cubic meter) while ingestion exposures may depend on tracking actual counts of microorganisms to determine the average number of microorganisms in a serving.

Three examples illustrate different approaches to developing exposure scenarios:

a) To estimate the risk of viruses in water, an exposure assessment considered the volume of water consumed and the average concentration of viruses per liter of water supplying a large city (Haas et al., 1993). In this exposure assessment, no attention was given to mechanisms potentially responsible for the average virus concentration or to factors that might cause variability in virus concentration across time or water supplies.

   For this example, a scenario could represent one possible combination of concentration (e.g., viruses per milliliter) and water consumed (e.g., milliliters per person per day). Therefore, the set of scenarios would include all possible combinations. Alternatively, this analysis is an example as a single scenario (e.g., water consumed) with variable average dose of virus per day.

b) To estimate the risk of tuberculosis (TB) transmission on a commercial airliner, an exposure assessment examined the spatial variability in concentration of this mycobacterium (Ko et al., 2004). This assessment included air transfer rates between different cabins in the plane, respiration rates of potentially exposed individuals, distance from an infectious source, and the rate of expired TB organisms from an infectious source. Hazard identification had indicated that all these factors might influence exposures of passengers to TB on a plane. The exposure assessment focused on the cumulative exposure to infectious TB during a long (> 8 hours) intercontinental flight. For this example, scenarios reflected the combinations of cabin location of exposed individuals, location of source individual and airflow direction.

c) To estimate the risk of human illness from *E. coli* O157:H7 in ground beef, an exposure assessment included causative factors during on-farm production, slaughter, and storage/preparation stages (Ebel et al., 2003). This complex farm-to-table exposure assessment examined the influence of season, live animal prevalence, transport, dehiding, carcass decontamination, carcass chilling, carcass fabrication, grinding, storage/handling, cooking and consumption on the predicted exposure per ground beef serving. Hazard identification indicated these factors might influence exposures. Control of many of these factors was considered by risk managers.

For this example, scenarios reflected the combinations of different sources (e.g., prevalence of different classes of infected cattle presented for slaughter by season) with effectiveness of decontamination procedures with times/temperatures of storage and cooking. The unique combinations that represent individual scenarios were too numerous to count. Furthermore, ground beef servings were created from combinations of scenarios that produced the beef that went into a load of ground product. Therefore, the exposure assessment did not list distinct scenarios but, instead, produced frequency distributions for levels of *E. coli* O157:H7 per serving of ground beef for low- and high-prevalence seasons of the year.

An exposure assessment depends on the microbial agent's properties and the environmental transmission factors relevant to exposures. Microbial agents may stem from more than one source, may be transmitted via multiple routes of exposure, and may be spread via secondary or vector transmission. An exposure source can originate from either natural or anthropogenic events, activities, or locations that generate or release hazards. Exposure sources can be classified as point sources or non-point sources. Exposure routes include inhalation (nose and mouth to lungs), ingestion (oral), and direct contact (skin, eyes, ears, and sexual). Exposure routes are situation-dependent and medium specific. An exposure pathway encompasses both exposure source and route, and generally is described by a source and release from a source, an exposure point, and an exposure route. Table 5.2 illustrates the exposure assessment components and their relationship to various exposure points.

Table 5.2 Transmission Pathways for Microbial Hazards

Source	Release	Exposure Point		Exposure Route
Natural or Anthropogenic Point or Non-point	Natural Accidental Intentional	Food	• Meat • Poultry • Eggs and Egg Products • Fish and shellfish • Produce • Dairy Products • Other Food Products	• Ingestion • Direct Contact
		Water	• Surface Water (Drinking) • Ground Water (Drinking)	• Ingestion • Direct Contact • Inhalation
			• Recreational Water • Compost Tea[32]	• Incidental Ingestion • Direct Contact • Inhalation
		Soil	• Surface Soil • Subsurface Soil • Sediment • Manure • Biosolids	
		Air	• Ambient Air • Indoor Air	
		Surfaces	• Porous • Non-porous	
		Biota	• Plants • Animals, including humans	

The dynamic nature of host-pathogen interactions, unique to infectious disease risk assessment, can lead to secondary transmission. The strictest definition of secondary transmission pertains to direct human-to-human contact between a primary case (infected or ill) and a secondary case that becomes infected or ill from that contact. Broader definitions include secondary cases that arise from contact with fomites or food or water contaminated by primary cases. Where secondary transmission includes infection from pathogens in the environment (e.g., fomites), it would not be considered secondary unless

[32] Compost tea is made by soaking or steeping compost in water. The resulting compost tea is used for fertilizing plants.

it occurs in the context of an outbreak where primary cases have already been identified. In such a case, the term "secondary transmission" is not used in the strictest sense, but is commonly used by public health professionals in the context of an outbreak.

5.2.3 What are the Exposed Populations I Could Consider?

Identifying the exposed individuals or subpopulations of interest is crucial to determining what data are needed for an exposure assessment. In some cases there may be statutory requirements or Agency policy that require certain populations to be considered. The following factors are inherently tied to the exposure scenario:

a) **Exposure Duration and Frequency** – Certain individuals/populations may have a comparatively greater exposure duration or frequency in a given environment. For some microorganisms, it is possible that exposure to a low concentration for a long duration may be a concern even if the concentration would not pose any health risks under short-term exposures.

b) **Exposure Routes** – Knowing the characteristics of the exposed population helps develop the appropriate exposure pathways to consider in the exposure assessment.

c) **Sensitive Individuals/Populations and Life Stages** – Some individuals/subpopulations may be more susceptible to infection or more likely to develop severe manifestations of infection. For example, while healthy individuals may recover from an *E. coli* infection, it can be deadly to young children, the elderly, and people with compromised immune systems. Similarly, chronic smokers may have impaired mucociliary clearance mechanisms, and therefore, may be more susceptible to respiratory infections.

Once the exposed individual/subpopulation or life stage has been determined, it is important to include any scenario-specific conditions in the conceptual model. Sensitive populations and/or life stages which may have different exposure considerations include:

a) Young children (up to 10 different age groups (EPA, 2005)

b) The elderly

c) Persons with compromised immune systems

d) Pregnant women

e) Chronic smokers

f) Military personnel (deployed and non-deployed)

g) Occupationally exposed individuals

h) Other groups based on behavioral patterns (e.g. subsistence fishing)

This list is not comprehensive. You should select individuals/subpopulations of interest based on the purpose of the risk assessment. However, your groupings may be limited by data availability. Any differences in grouping between the subpopulations considered in the dose-response and the exposure components of risk assessment should be carefully explained and the implications of those differences should be discussed.

5.2.4 What are Common Approaches to Exposure Modeling I Can Use?

Although there are more elaborate classification schemes (Hurd and Kaneene, 1993), most exposure assessments use an attribution and/or process modeling approach (Cox, 2006). Process modeling is the more common approach and it generates more traditional results (i.e., likelihood and dose) from the exposure assessment. Attribution modeling, in contrast, is used less frequently and does not explicitly estimate the likelihood of microbial doses. Instead, this approach implicitly synthesizes the exposure output with hazard characterization to generate links between model inputs and numbers of human illnesses. Some refer to attribution and/or empirically-based models as 'black-box' approaches because causative mechanisms are not explicitly included (ECSSC, 2003). Nevertheless, attribution modeling is empirically based and is often available to assessors when the problem is rich with surveillance evidence.

Attribution Modeling

Empirical estimates of the number of human illnesses per year caused by the microorganism are sometimes available. These data might be used, in conjunction with other information, to back-calculate the fraction of illnesses attributed to various scenarios. This approach is infrequently used in microbial exposure assessment because epidemiological surveillance data is prone to data gaps that may not be well characterized (under reporting of illnesses is significant and varies). Nevertheless, national surveillance systems combined with thorough investigations of outbreaks may generate sufficiently valid conclusions about sources and causes of those illnesses. If such data are available, it is feasible to determine the number of exposures from a target source that cause human illnesses. Furthermore, the estimated number of illnesses that result from the target source can be divided by the estimated number of total exposures to determine the probability of illness per exposure.

Ultimately, the number of illnesses from a microorganism that are attributed to a source represents the total number of illnesses that could be avoided by eliminating that source. If complete elimination of the source is not feasible, then further analysis might suggest what fraction of a baseline number of cases could be avoided by a proposed risk management option that improves the effectiveness of some particular process.

For illustrative purposes, assume 1,000 cases of illness caused by microorganism B are detected by public health surveillance each year (Figure 5.2). Analysis of the

surveillance system suggests that only one of every 50 cases that occur is detected via this system. Therefore, the true number of cases is estimated to be 50,000 per year. Epidemiologic evidence suggests that 20 percent of cases are caused by Source 1 (e.g., ground beef). Research evidence suggests that 50 percent of source 1 exposures are directly attributable to process 1 (e.g., under-cooking). Based on these values, you estimate the number of illnesses attributed to Source 1 is 50,000×0.20=10,000. You estimate the number of those illnesses attributed to process 1 is 10,000×0.50=5,000.

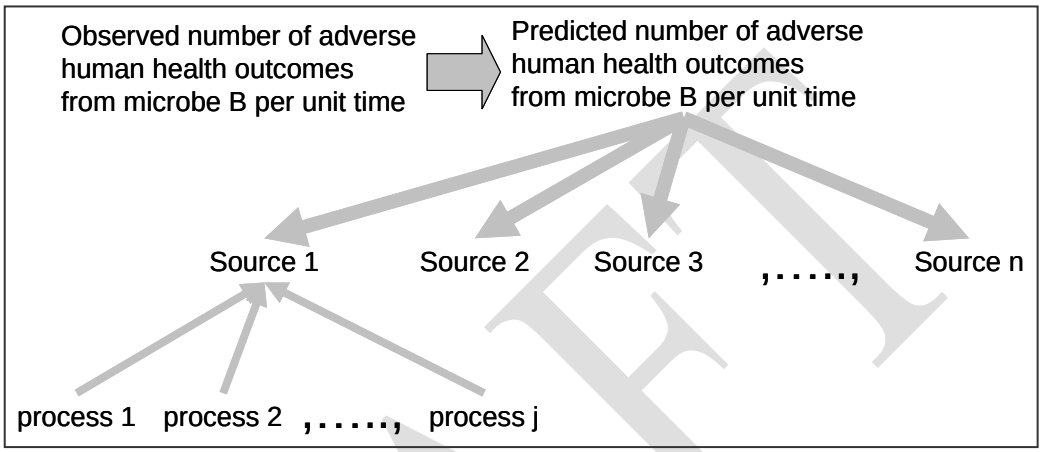

Figure 5.2 Schematic illustrating direction of inference when using an attribution approach to exposure assessment.

Process Modeling

For many exposure assessments, planning and scoping will suggest that attribution modeling is inadequate. A dearth of reasonable epidemiologic surveillance evidence sometimes precludes attribution modeling, as well. In these cases, a process modeling approach can be used.

Generally, process modeling involves the characterization of processes that occur from the initial source of the microbe to the point of exposure. The influence of processes associated with growth or attenuation of microbial populations is one of the purposes of the predictive microbiology field. Research of microbial kinetics and lethality provides important insight to process modeling.

Simulation of the effects of processes on microbial occurrence is commonly accomplished using Monte Carlo sampling techniques (see Section 5.1.11). Nevertheless, simple probabilistic models may not require Monte Carlo sampling and other dynamic modeling methods, such as those based on differential equations, may be solvable using other mathematical techniques (e.g., Cox, 2006).

Regardless of the technique, process modeling is concerned with predicting exposures given the initial microbial occurrence. Its direction of inference is opposite

that of attribution modeling (Figure 5.2) and calculations tend to follow an input-output flow from initial microbial levels through each process in the model.

Process modeling begins with some prediction of microbe occurrence prior to exposure (Figure 5.3). Processes that occur subsequent to this beginning point influence the microbial population to grow, maintain, or decline. These processes have been referred to as transformations and the mathematic effect of these transformations is sometimes modeled using transformation ratios (Haas et al., 1999). Such ratios simply describe the level of microorganisms after the process to the level before the process. You can define

$$\varepsilon_j = \frac{\text{amount of microbe after process j, } (x_j)}{\text{amount of microbe before process j, } (x_{j-1})} \qquad \text{(Eq 5.2)}$$

and stipulate that this value is reasonably estimated from predictive microbiology. It might also be invariant to the initial microbe level so that the ratio is constant across all amounts of the microbe. If you are given an amount of a microbe before the process (x_{j-1}), you can calculate the amount of microbe after the process as $x_j = \varepsilon_j \times x_{j-1}$, where the *j* subscript could refer to a time step somewhere between the beginning and end of the model when process *j* is applied. For example, time step *j* may be a decontamination step applied to water, food or something else in which the microorganism occurs. If the exposure assessment predicts the amount before the decontamination process, then this equation predicts the amount after that step. Nevertheless, ε_j is an abstraction of the complicated interactions that likely exist between microorganisms and their environment. Sometimes those complex interactions need to be explicitly modeled using more complicated math and/or simulation. Note that this approach does not incorporate cases where toxin generation during processing, before exposure is an issue.

A modular approach to exposure assessment process models has been suggested for food safety applications (Nauta, 2002). One advantage of subdividing a model is that complicated components in one module can be summarized into a simpler format before propagating the output of that module to the next module. For example, a module may predict microbial growth as a function of storage times and temperatures and other factors. The resulting prediction may be a continuous distribution describing the exponential growth of the microbial populations that are input to this module. Nevertheless, it may only be necessary to consider a small number of discrete levels of microorganisms in the next module (e.g., by $\log_{10}$ units), so this module's continuous results can be summarized before calculations in the next module are completed.

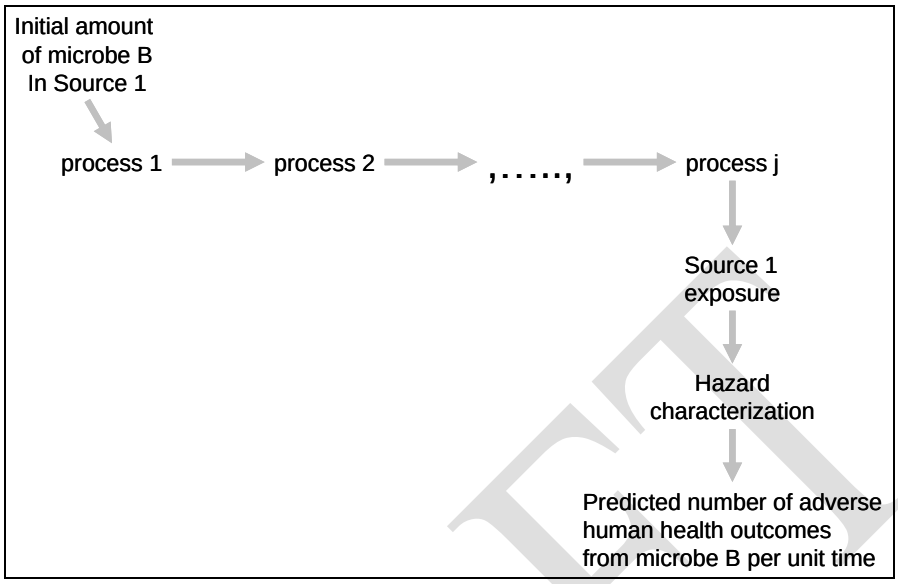

Figure 5.3 Schematic illustrating direction of inference when using a process modeling approach

5.2.5 How is Scenario Analysis Used in Exposure Assessment?

Scenario analysis begins the process of identifying those scenarios that constitute a risk as well as those that are not a risk. It simplifies the next stage of exposure model development; quantifying the likelihood of those scenarios that end in an exposure dose.

In developing scenarios during the conceptual development phase, you also start the process of defining the mathematical relationships between steps. Are these additive or multiplicative? Are there correlations between different inputs? What other factors influence the relationships among inputs?

Compared to scenario development, determining likelihood and dose is usually a more mathematical exercise. The relationships between model inputs need to be mathematically described; often statistical methods are employed to quantify these relationships. Also, the process of converting conceptual relationships into explicit mathematical relationships commonly involves additional assumptions beyond those represented in the conceptual model.

Before data availability or analysis is considered, the mathematical development needed to determine likelihood and dose should be determined. Once a tentative mathematical model is completed, its structure can be communicated with outside reviewers and risk managers. Typically, the mathematics will provoke discussion about the data needs of the model. A clearly defined mathematical model provides an opportunity for specialists to review and comment on the course of the project. If

scrutiny of the mathematics determines the need for change, then you can make changes before a large investment in data acquisition and analysis is completed. Frequent outreach and feedback is one manifestation of effective exposure assessment.

An Illustrative Example

In a very simple illustrative example, you want to predict the exposure risk from servings containing 10 units of microbe B (Figure 5.4). Two scenarios are considered: one where the serving is cooked such that all the microorganisms in the serving are destroyed and another where the serving is not cooked and all the microorganisms survive. From available data or expert opinion, you determine that the likelihood of cooking such servings is f, where $0 \leq f \leq 1$. Therefore, the likelihood (l_1) of the cooking scenario (s_1) is f which is inseparable from an exposure dose (x_1) of zero units of microorganism B.

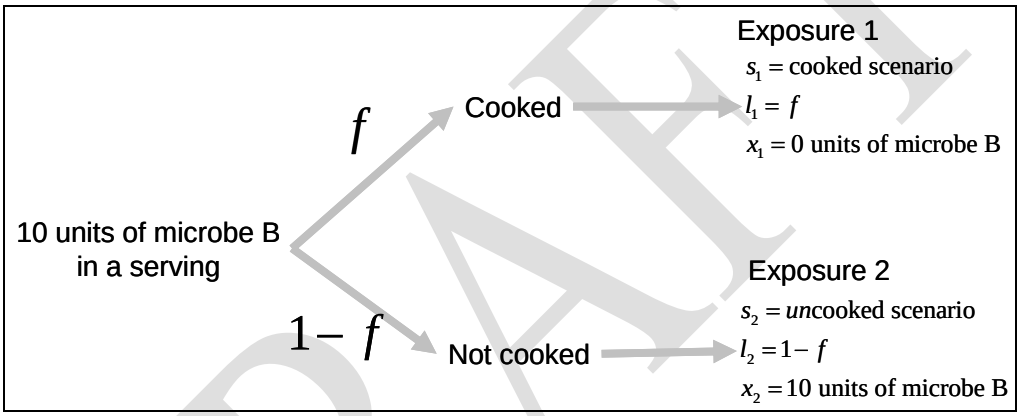

Figure 5.4 A simple illustrative example of two exposure scenarios resulting from an initial amount of microorganism B in a serving of food. In this case *f* is the probability that a serving will be cooked such that all of microbe B is destroyed. Conversely, 1-*f* is the probability that a serving will not be cooked and the total amount of microorganism B survives to expose a consumer.

In a slightly more complicated illustrative example, you consider three average concentrations of microorganism B per serving, three levels of cooking effectiveness, and three amounts of food or water consumed per serving (Figure 5.5). This example generates 27 different scenarios for the exposure assessment.

Besides the complication of more scenarios, this example also uses a concentration measure for microorganism levels instead of a fixed number of microorganisms in a serving. Concentrations are used in many food and water microbial exposure assessments. It is sometimes recommended that modeling microbial concentration be avoided and physical counts of microorganisms should instead be explicitly tracked (ECSSC, 2003; Nauta, 2005). One justification for such an approach is that it avoids mass balance mistakes that can occur when processes necessarily change the microbial numbers. For example, a partitioning of some bulk quantity into subunits

necessarily must account for all the organisms that existed in the bulk quantity. Yet, an average concentration per subunit might result in an incorrect assessment of the exposure dose per subunit. Nevertheless, for problems without partitioning or mixing processes, using concentration may be sufficient.

You may assume this example pertains to some bulk product in which three average concentrations (e.g., microorganisms per milliliter or per gram) are possible. You define each concentration as λ_1, λ_2, and λ_3 and refer to the likelihood of each of these as $f(\lambda_1)$, $f(\lambda_2)$, and $f(\lambda_3)$. A similar approach is used for cooking effectiveness levels (ε_i and $g(\varepsilon_i)$) and consumption amounts (υ_i and $h(\upsilon_i)$) to define their values and likelihoods[33].

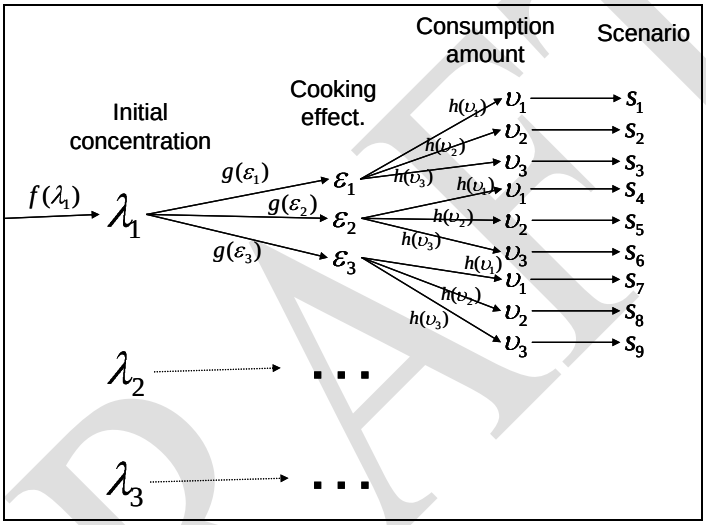

Figure 5.5 An illustrative example of a slightly more complicated exposure assessment

The dose and likelihood for each of the 27 scenarios generated for this exposure assessment can be calculated directly from the information given. For example, the average dose for scenario 1 (x_1) is the product of concentration (λ_1), cooking effectiveness (ε_1) and serving size consumed (υ_1). Similarly, the likelihood of scenario 1 (l_1) is the joint probability of each of these events occurring; if you know these events are independent, then $l_1(x_1,\lambda_1,\varepsilon_1,\upsilon_1) = f(\lambda_1) \times g(\varepsilon_1) \times h(\upsilon_1)$.

The multiplications for this simple model are common for many process models. Exposure dose is often the result of multiplicative, input-output, relationships; in this case the average dose for a scenario is $x_i = \lambda_i \times \varepsilon_i \times \upsilon_i$. The unit of x_i is microorganisms per

[33] Note that branch likelihoods must sum to one (i.e., $\sum_1^3 g(\varepsilon_i) = 1$) so if we know two of the likelihoods then the third is also known (e.g., $g(\varepsilon_3) = 1 - g(\varepsilon_1) - g(\varepsilon_2)$.

serving; λ_i is microorganisms per volume or mass; ε_I is unitless ratios; and υ_i is volume or mass per serving. Predictive microbiology sometimes prefers to represent microbe quantities in $\log_{10}$ units. A logarithmic treatment will convert the multiplicative calculations of the model to addition or subtraction (e.g., $\log(x_i) = \log(\lambda_i) + \log(\varepsilon_i) + \log(\upsilon_i)$) while the joint likelihood remains the product of the likelihoods of each variable in the model.

Although the results from this example are trivial, it is illustrative to interpret them. These results explain which scenarios generate the highest doses and the likelihoods that those doses occur. By summing the likelihoods of scenarios in which zero organisms occur, the prevalence of non-zero exposures can be determined (i.e., $prevalence = 1 - \sum_i f(x_i = 0)$).

The relative importance of different concentrations, cooking effectiveness levels and amounts consumed might also be assessed by calculating conditional expected dose values. For example, the conditional expected dose value for one concentration ($E\ x \mid \lambda = y$) is the average dose calculated when only that concentration is considered but all the values for cooking effectiveness and amounts consumed are still possible.

$$E\ x \mid \lambda = y\ = \frac{y \times \sum_i \sum_j \ \varepsilon_i \times \upsilon_j\ \times\ g(\varepsilon_i) \times h(\upsilon_i)}{f(\lambda = y)} \qquad \text{(Eq 5.3)}$$

The magnitudes of the conditional expected values suggest the influence of these inputs on the average exposure dose (Table 5.3). For example, the largest change in average dose occurs across the possible values for initial concentration, but the smallest average dose occurs if cooking is completely effective.

Table 5.3 Results of a simple exposure assessment

λ_i	$f(\lambda_i)$	ε_i	$g(\varepsilon_i)$	υ_i	$h(\upsilon_i)$
1	0.7	0	0.75	10	0.25
10	0.2	0.5	0.2	100	0.5
50	0.1	1	0.05	150	0.25

Variable	Value	$E\ x \mid \text{variable} = y$
λ_i	1	14
	10	135
	50	675
ε_i	0	0
	0.5	249
	1	462
υ_i	10	12
	100	116
	150	173

	Calculated values		Sorted values	
scenario	dose,	likelihood,	dose,	likelihood,
s_i	x_i	l_i	x_i	l_i
1	0	0.131	0	0.750
2	0	0.263	5	0.035
3	0	0.131	10	0.009
4	5	0.035	50	0.080
5	50	0.070	75	0.035
6	75	0.035	100	0.020
7	10	0.009	150	0.009
8	100	0.018	250	0.005
9	150	0.009	500	0.021
10	0	0.038	750	0.010
11	0	0.075	1000	0.005
12	0	0.038	1500	0.003
13	50	0.010	2500	0.010
14	500	0.020	3750	0.005
15	750	0.010	5000	0.003
16	100	0.003	7500	0.001
17	1000	0.005		
18	1500	0.003		
19	0	0.019		
20	0	0.038		
21	0	0.019		
22	250	0.005		
23	2500	0.010		
24	3750	0.005		
25	500	0.001		
26	5000	0.003		
27	7500	0.001		

As the previous example illustrates, explicit tree-diagram schematics of exposure scenarios are daunting when the inputs can assume multiple values and the number of processes is increased. A full graphic depiction of all 27 scenarios for the previous example would not fit on a single page.

For more complicated exposure models, the inputs are simply treated as random variables that are mathematically combined. In the simple example, you can represent all three possible values for initial concentration as $\tilde{\lambda}$ where the tilde symbol signifies that concentration is a random variable. You can similarly define $\tilde{\varepsilon}$ and $\tilde{\upsilon}$.

Using symbols for the random variables in the model, exposure can be mathematically written as $\tilde{x} = \tilde{\lambda} \times \tilde{\varepsilon} \times \tilde{\upsilon}$ where the dose delivered to humans is also a random variable by virtue of the fact that it is a function of random variables. Statistical moments of $\tilde{x}$ (e.g., its mean and variance) might be predictable, but when the likelihood distributions for $\tilde{\lambda}$, $\tilde{\varepsilon}$ and $\tilde{\upsilon}$ involve more than a trivial number of values, other techniques (e.g., Monte Carlo simulation) are often used to determine $\tilde{x}$. Nevertheless, the techniques essentially mimic the procedure followed for the simple example; the

possible values for $\tilde{\lambda}$, $\tilde{\varepsilon}$, and $\tilde{\upsilon}$ are multiplied together and their joint likelihood is determined.

Once an exposure model becomes more complex, the identities of individual scenarios are difficult to determine. It is common for exposure assessments to focus on predicting the likelihood of doses without explicitly identifying scenarios. In these situations, sensitivity analysis is used to sort out the relative influence of model inputs on the exposure distribution. Nevertheless, it is sometimes crucial to identify the higher risk scenarios; thinking about the model as a scenario tree is one useful technique for elucidating those scenarios.

Random variables can be discrete or continuous. Exposure assessments often use a mixture of both. Although it is intuitively appealing to consider microbial counts as discretely distributed random variables, it is not always essential that they be treated as such. Naturally continuous distributions, like weight measures or measures of effectiveness, may also be treated as discrete random variables in some models to simplify their calculations without any loss of information. Like most decisions in exposure assessment, the choice of distribution is informed by planning and scoping. Nevertheless, such decisions should be made with an understanding of the biologic plausibility of the choice.

Determining the likelihood of doses of microorganisms is the fundamental objective of most exposure assessments. The key to effective exposure assessment, therefore, is to explain the mathematical relationships among the random variables that contribute to exposure. Once you explain and justify the mathematical model, the process of determining the exposure distribution is relatively straight-forward. Nevertheless, much of the work of conducting an exposure assessment involves collection and analysis of available data for the different random variables in the model, as well as explicit representation of the uncertainty about inputs to the model or the model itself.

5.2.6 What is the Role of Predictive Microbiology in Exposure Assessment?

The field of predictive microbiology is important to many microbial exposure assessments. This field is concerned with quantifying the dynamics of microbial populations and these dynamics often depend on environmental and other biologic factors. Useful discussions on the mathematics and statistics of predictive microbiology are available (Ross and McMeekin, 1994, 2003; Haas et al., 1999; ECSCC, 2004; Vose, 2008). These references also cite seminal research in this discipline. Within the field of food safety, the Center of Excellence in Microbiological Modeling provides research and products related to pathogen modeling.[34]

Functional relationships that describe microbial dynamics are typically of an input-output form. For example, an exponential growth model is;

[34] http://ars.usda.gov/Services/docs.htm?docid=8392

$$N_t = N_0 e^{k \times t}$$

where N_t and N_0 are the number of microorganisms at times t and zero, respectively, and *k* is an exponential growth rate constant. Rearrangement of this relationship illustrates derivation of a transformation ratio for growth;

$$\varepsilon = \frac{N_t}{N_0} = e^{k \times t} \qquad \text{(Eq 5.4)}$$

If the constant, *k*, in the above equations is a negative value, then the same relationship can serve to predict attenuation of microorganisms in time unit t.

The exponential growth rate is only constant for particular environmental conditions. At a minimum, most exposure assessments will consider environmental conditions to be variable between scenarios. In such cases, *k* is some function of temperature, pH and/or other conditions (i.e., *k = f*(*environmental conditions*)). The reader should refer to the Center of Excellence in Microbiological Modeling or the FoodRisk.org[35] websites for research regarding how various environmental factors – and microbial strain differences – influence microbial growth and/or attenuation behavior. Specific guidance for statistical fitting of experimental microbial growth or attenuation data can be found in these references.

Growth or attenuation functions can be deterministic (i.e., one set of parameters predicts one amount of change) or stochastic (i.e., one set of parameters predicts a probability distribution for amount of change). In the context of an exposure assessment, however, the predictions from either a deterministic or stochastic function will be stochastic because the environmental parameters upon which growth or attenuation depend are variables. This source of variability relates to human behaviors such as storage times and temperatures that vary across individuals.

Clearly, human behavior can be highly variable; and behaviors such as exposing raw foods to high temperatures for extended periods of time can dramatically affect the dose of pathogens ultimately consumed. Although data on microbial growth or attenuation behavior can be generated readily in experimental laboratories, data on human food handling behaviors must be collected via well-designed human population surveys. Such data are available for some commodities, such as ready-to-eat foods (Kosa et al., 2007; Pouillot et al., 2010), but not necessarily for all food commodities. Although data regarding refrigeration temperatures may be applicable to most perishable foods, storage time within the refrigerator may depend on the particular food; this phenomenon can only be captured via food-specific surveys. In addition, times and temperatures that foods experience during transport from retail to homes, during food preparation and prior to (or following partial) consumption are sometimes needed. Actual human behavior

[35] http://www.foodrisk.org/resource_types/tools/predictive_micro.cfm

data that captures variability in cooking processes is also sometimes important for estimating the microbial attenuation achieved prior to consumption.

You can apply more complex growth and attenuation models in exposure modeling. For example, the Gompertz equation – or modifications thereof – includes specific parameters for lag time and asymptotic maximum density (Haas et al., 1999). Lag time is a characteristic of many growth curves; measured from time zero, it is the elapsed period before exponential growth begins. The maximum density that a microbe can attain before competition for nutrients among the microorganisms halts growth is another modeling characteristic studied by predictive microbiologists.

Predictive microbiology provides insight and data concerning the behavior of microorganisms across different environmental conditions. Nevertheless, such insight and data needs to be translated and extrapolated from experimental studies to natural conditions when applied to exposure assessment. Adjustment of results from controlled experimental settings to highly variable (and uncertain) natural conditions can be difficult. Therefore, care should be taken when applying predictive microbiology to exposure assessment.

Common difficulties for direct application of predictive microbiology to exposure assessment are: accounting for variable temperatures across time and accounting for the competitive effects of other microorganisms on the growth characteristics of a target microbe. Varying temperatures suggest variable transitions between growth, maintenance and attenuation of microbial populations. Depending on whether growth or attenuation is a process with memory or is memory-less, the modeling techniques needed for microbial dynamics will be different (Vose, 2008). The existence of other microorganisms in media can influence the growth rate or maximum density a target microbe can achieve, which is termed the Jameson effect (Ross and McMeekin, 2003).

ILSI (2010) looked at mechanisms that have an impact on physical distributions, characteristics of frequency distributions employed to model microbial distributions, and the impact of both physical and frequency distributions on illness risk and food safety management criteria. ILSI outlined six mechanisms that can impact the microbial distribution in a foodstuff: contamination, growth, death, joining, mixing, and fractionation (ILSI, 2010).

Although not necessarily a part of predictive microbiology, the processes of microbial transfer and cross-contamination are also not well researched. There is a need for further development of modeling approaches for these potentially important processes. Some default techniques have been suggested for use in food safety applications (ECSCC, 2003).

5.2.7 How Can I Address Secondary Transmission of Disease in the Population?

The approach described heretofore assumes that exposures result directly from the media of interest (e.g., food or drinking water) and the potential for person-to-person transmission of disease is not taken into account. Such an approach generally assumes that multiple or recurring exposures constitute independent events with identical distributions of contamination (Regli et al., 1991). Furthermore, secondary transmission and immunity are most often assumed to be negligible in this approach. Nevertheless, such assumptions may not be valid.

To more completely assess all possible exposures, it may be necessary to consider possible secondary transmissions that result from a primary infection. Such an approach commonly requires consideration of a disease transmission model. A variety of models have been formulated, mathematically analyzed, and applied to infectious diseases (Hethcote, 2000). Mathematical models of disease transmission have become important tools that have led to understanding the transmission characteristics of infectious diseases in communities and better approaches to decreasing the transmission of these diseases (Hethcote, 2000; King et al., 2008; Riley et al., 2003). Modeling infectious disease processes such as person-to-person transmission of infection and immunity requires dynamic methods where the number of individuals that are assumed to be susceptible to infection is time-varying and risk is manifest at the population level (Anderson and May, 1991; Hethcote, 1976, 2000).

Epidemiological disease transmission models stratify a population of potentially exposed humans into different states according to disease status:

a) Susceptible

b) Diseased (infectious and symptomatic)

c) Immune (partial or complete)

d) Carrier (infectious but asymptomatic)

Only a portion of the population is in the susceptible state at any point in time, and only those individuals in a susceptible state can become infected through exposure to pathogens. Members of a population may move between model states and model parameters predict the numbers of people that are in each of the epidemiological states at any given point in time. Factors affecting the population dynamics include the level and frequency of exposure, the ability of individuals in infectious states to infect susceptible individuals, and the temporal processes of the disease (e.g., incubation period, duration of disease, duration of protective immunity). The rate parameters may be determined through literature review or through site-specific data.

Disease transmission models may also be used to determine the primary exposure. Such models may be necessary to predict the level and frequency of contaminated media when little or no empiric evidence is available. For example, assessing exposures that result from the inadvertent slaughter of a Highly Pathogenic Avian Influenza-infected U.S. poultry flock requires modeling the epidemic spread of the virus within that flock (USDA, 2008). This approach is needed because there is no evidence available concerning the occurrence of this pathogen among U.S. poultry flocks.

Secondary transmission modeling for airborne microorganisms such as anthrax and severe acute respiratory syndrome (SARS) virus may be discussed in more detail in a future volume of this Guideline (Riley et al., 2003; Bartrand et al., 2008).

5.2.8 What Data Can I Use in an Exposure Assessment?

Ideally, the data needed for an exposure assessment are determined by its specific conceptual and mathematic models. If the needs are clearly determined before effort is expended in collecting and analyzing data, a fuller and more efficient treatment of relevant data can be accomplished.

Exposure assessments data usually stem from either population- or experimental-based surveys or studies. These data are preferably from published or reviewed research and are fully relevant and representative of what is needed in the exposure assessment. However, this is not always the case. An extensive discussion of data types and sources is available (WHO/FAO, 2008).

The broad categories of data needs for exposure assessment are: microbial, processes, and exposed humans. Within each of these categories, however, is an array of data types that may be needed for specific analyses.

Data on Microorganisms

Data about the occurrence and amount of microorganisms within the medium of interest (e.g., water, food, air) is important for process modeling. It is desirable to have occurrence data for multiple points between the beginning of the model and the point of exposure.

Prevalence data provide presence/absence data for the occurrence of a microorganism in a medium. Such data support estimation of the proportion of some population in which the microbe occurs (i.e., $prevalence = \frac{\#\ of\ \ units\ with\ microbe}{\#\ of\ \ units\ in\ population}$) during some cross-section of time. Observational studies that solely report prevalence are rarely directly applicable to exposure assessments. Instead, the apparent prevalence must be adjusted for the probability that units with the microbe would be detected if actually present (i.e., sensitivity) in order to estimate the true prevalence. Apparent prevalence is also influenced by the probability that units without the microbe might be

incorrectly detected (i.e., 1 – specificity), for example, if there were cross-reactivity in an immunological assay.

Counts of microorganisms in sampling studies are desirable for exposure assessments. Such data may arise from microbiologic techniques such as direct plating, observation or most probable number assays (Haas et al., 1999). These data provide an empirical distribution of the number of samples that contained each count of microbe observed. Nevertheless, it is important to also know the sensitivity and specificity of the methods used in count assays to accurately interpret the data.

A well-designed exposure assessment fully characterizes the microbe (or microorganisms) on which it is focused. Oftentimes, occurrence data will not be specific for the target microbe and additional data will be needed to interpret the relevance of the occurrence data. For example, count data for all *Salmonella* serotypes on broiler carcasses would need to be adjusted if the focus of the exposure assessment was *Salmonella enteric* Typhimurium. If you knew that the count data were adequately described by a Poisson distribution and you also estimated that 20% of all *Salmonella* were *Salmonella enteric* Typhimurium, then you might simply model the counts of *Salmonella enteric* Typhimurium as distributed according to a Poisson ($0.2\times\lambda$) distribution. Yet, other data may suggest this simple approach does not adequately account for the clustered occurrence of specific *Salmonella* serotypes on broiler carcasses.

Process Data

General processes common to many exposure assessments include growth and attenuation of the target microbe(s), as well as mixing and partitioning of the medium in which the microbe occurs (Nauta, 2002). The processes modeled in an exposure assessment should be informed by the evidence used to construct the conceptual model.

Predicted changes in microbial amounts resulting from growth or attenuation processes may be available from predictive microbiology research. Nevertheless, these predictions are often functionally dependent on environmental factors. Therefore, you need data to characterize the variability in parameters such as temperature and time in order to employ predictive microbiology in an exposure assessment.

Partitioning of water, food or air into smaller subunits prior to exposure is a common problem in exposure assessment that requires industry or ecologic data to solve. Mass balance considerations may be required to account for recycling or cross-contamination of microorganisms in some scenarios. Some of these data may come from industry- or government-sponsored surveillance systems; but sometimes expert experience will be the only information available.

Human Characteristics and Behavioral Data

Demographic and behavior data concerning exposed humans will be specific to the subpopulations and media considered in the exposure assessment. Much of the data used to characterize subpopulations will come from routine government surveys. These surveys provide demographic data by geographic region, age, sex and other factors. The estimated proportion of the population that represents a susceptible population may be available from epidemiologic research. Extrapolations from non-representative data may require substantial modeling and expert judgment to accomplish.

Data on human behaviors that influence the exposure assessment will be needed. Some behaviors, like time and temperature of storage or cooking, are highly variable among the human population. Some of these behaviors have been the subjects of on-going research projects.[36] These data inform the growth and inactivation processes via their predictive functional relationship with microbial counts.

Specific data on some human behaviors that increase the likelihood of exposure to a particular microbe (e.g., preference for raw meats or seafood, occupational exposure to microbe rich environments, cohabitation with infected individuals) are sometimes difficult to locate. Nevertheless, some frequency and contact rates have been summarized for water and air media (Haas et al., 1999). Similarly, government surveys can provide data on variability in consumption, inhalation or contact amounts across individuals and groups of individuals.[37]

Some well known and frequently used sources of human consumption data include CDC's National Health and Nutrition Examination Survey (NHANES)[38] and USDA's Continuing Survey of Food Intakes by Individuals (CSFII), which as of 2002 have been integrated and maintain the name NHANES (Dwyer, 2003). The FoodNet Population Survey Atlas of Exposures is also a useful resource.[39]

5.2.9 How do I Use Data in an Exposure Assessment?

Data provide evidence about the inputs to the exposure assessment, but data also influence the magnitude of uncertainty surrounding its results. Weak or absent data are usually associated with large uncertainties while data that are substantial, relevant and representative contribute little uncertainty to an exposure assessment.

Population-based, observational data are commonly used to estimate the parameters for random variables in exposure assessments. Statistically robust approaches for selecting appropriate probability distributions, estimating the parameters of those distributions and comparing alternative distributions are explained elsewhere (Vose, 2008 and Haas et al., 1999). Often the process of fitting data to distributions is complicated

[36] www.cfsan.fda.gov/~lrd/ab-foodb.html
[37] www.ars.usda.gov/main
[38] http://www.cdc.gov/nchs/nhanes.htm
[39] http://www.cdc.gov/foodnet/surveys/FoodNetExposureAtlas0607_508.pdf

because the data were generated by imperfect detection systems. Adjustments for imperfect detection sensitivity and specificity are discussed in those same references.

Although there are pros and cons to strict application of either classic statistical ("frequentist") or Bayesian estimation methods, it is often the case that the results of the two approaches are very similar. Results will tend to differ when the available dose-response data are very limited and/or when there is substantial information other than the numerical dose-response data that leads to a very informative (i.e., precise) prior.

Procedures for statistical fitting of data to distributions include appropriate methods for determining the uncertainty in the estimated distribution parameter(s). This uncertainty is propagated through the exposure assessment and combined with other sources of uncertainty to quantify the total uncertainty about the resulting exposure distribution.

Data of questionable relevance to the specific exposure assessment require special consideration. Similarly, data that are not entirely representative of the populations modeled should be carefully used. It can be argued that data pertaining to one microbe are also relevant to another. Such 'surrogate' relationships should only be exploited when there is an absence of data that is directly relevant. Establishing the credibility of a surrogate for the target microbe may require a high standard of proof. It is usually preferred to use directly relevant data and honestly represent its uncertainty than to mix highly relevant and surrogate data in an attempt to reduce uncertainty.

Highly representative data are generated from random sampling of all relevant subpopulations. For example, an exposure assessment pertaining to the United States would preferably use human behavior data generated from a representative sample of U.S. persons. Nevertheless, representative data may not be available for some model inputs. Instead, data from specific regions or other countries may be available. Based on comparison of other factors it may be concluded that one or more of these other data sources could be a reasonable substitute. In such cases, you can make an effort to determine the best substitute and only use its data in the exposure assessment. It is usually inappropriate to mix multiple sources of less representative data because the resulting estimates often imply more confidence than is legitimate to claim.

5.3 How do I Analyze a Model's Results?

The general purpose of an exposure assessment is to translate the technical inputs of a model into a description of the likelihood of exposure doses in some defined part of the human population. Quantitative assessments estimate numeric values while qualitative assessments may use ordinal metrics (e.g., high, medium, and low) to signify the magnitudes of exposures.

Quantitative exposure assessment models usually comprise random variables that consequently generate variability in exposures. Therefore, a common output of the exposure assessment is the frequency distribution of possible doses that come in contact

with the human population of interest. This distribution is an estimate of the actual variability in exposures that occurs in nature. If the model is calculated based on input values thought to accurately reflect current conditions, this variability reflects what is occurring at present. Exposure assessments often refer to such predictions as the baseline exposure. If the model is calculated based on proposed risk management changes, then the variability reflects a prediction about the future.

It should be noted that the exposure distribution calculated from a model is also constrained by some unit of time. This time component must be explicit to correctly interpret and extrapolate the variability predicted by the model.

Analysis of the exposure distribution includes determining the sensitivity of this distribution to changes in the random variables (or other model inputs) used to predict it. Sensitivity analysis determines which model inputs are the primary drivers or predictors of substantial doses with relatively high likelihoods of occurring. Conclusions about the important inputs directly inform risk managers by suggesting what changes in the system will cause the greatest reduction in exposures.

If all model inputs were perfectly known, then the output of an exposure assessment might arguably consist of a single exposure distribution. Yet uncertainty potentially pervades all aspects of an exposure model and the resulting uncertainty about its predictions can be incorporated into any analysis. Uncertainty analysis is concerned with determining the influence of the various sources of uncertainty on the predictions of the exposure assessment.

5.3.1 How do I Report Exposure in an Exposure Assessment?

The natural output of an exposure assessment is an exposure distribution; this distribution provides likelihood or frequency values for the range of possible doses that constitute exposures. You should clearly identify the applicable time period and the exposed human population to which the exposure distribution applies. The most common format for reporting exposures is tables or graphs.

An exposure distribution may reflect the possible doses an individual could experience in, for example, one year. The objective of the risk assessment should determine the type of exposure distribution reported. It could reflect those doses relevant to a highly sensitive population, life stage, or to the entire population.

An exposure assessment will convey the variability of doses for the relevant population per unit time, but it may also include consideration of the variability of the entire distribution across time. A dynamic exposure assessment that predicts trends in exposure across time may report the trends based on the statistical expected values of the individual exposure distributions or it may report the actual distributions for each time period considered.

It is essential that you communicate the magnitude of uncertainty about the true exposure distribution. A number of techniques may be used to convey the uncertainty, but no method is universally applicable and all methods can be computationally intensive (Lammerding and Fazil, 2000; Cullen, 1999). Second-order modeling – in which variability is derived for one combination of uncertain inputs and the process is repeated until a full range of plausible combinations is achieved – is a common technique to accomplish a clear separation of variability from uncertainty (Vose, 2008). Nevertheless, the complexity of this technique may preclude its widespread application.

Multiple exposures to various doses for the same individual can complicate exposure assessments. It is not uncommon for exposure assessments to assume that exposure to non-zero doses is an infrequent phenomenon within a population. Therefore, it is typically assumed that there is a one-to-one correspondence between exposures and individuals, or at least the same individual is unlikely to be exposed twice within a short period of time. Nevertheless, in some cases this assumption is not robust, especially when considering exposures that may be clustered in space and time. Microbial dose-response models typically describe the likelihood of an adverse outcome as a function of a single exposure to some dose. If persons are expected to face multiple exposures, then the process of combining the exposure information with a typical dose-response function is be different from a standard one-dose one-person approach (Haas et al., 1999).

5.3.2 How do I Determine a Change in Exposure and Subsequent Risk?

The purpose of many MRAs is to determine how risk management decisions might change the risk of an adverse human health outcome. From the perspective of an exposure assessment, this usually requires calculating the model with and without the proposed change. For example, risk managers may want to evaluate the effect of some new mitigation process on the frequency or level of exposure doses. A baseline exposure prediction is compared to an exposure prediction based on the inclusion of the new mitigation process.

In large exposure models, measuring change in exposure is complicated by the role of uncertainty in the model's predictions. The baseline model predicts an exposure distribution with its attendant uncertainty. The mitigation model will predict a different exposure distribution with its own attendant uncertainty. But, the uncertainties between the two predictions are not independent of each other. It should be clear that the same things that contribute uncertainty to the baseline predictions usually apply to the mitigation predictions. Therefore, measuring the change in exposure between the two predictions is not trivial. Typically, the change you are interested in quantifying is in the number of adverse human health outcomes. That change cannot be calculated until risk characterization takes place and the problem of dependent uncertainties is compounded by the inclusion of dose-response uncertainty.

A direct method for accurately measuring changes in exposures or adverse human health outcomes is to model the baseline and mitigation predictions simultaneously using

some parallel modeling structure. This method is computationally daunting, however, and is not commonly followed.

Another approach can provide boundaries for the magnitude of change in numbers of adverse human health outcomes. In this approach, two uncertainty distributions about numbers of adverse human health outcomes, generated by separately calculating the baseline and mitigation predictions, are subtracted from each other assuming perfect positive correlation and assuming perfect independence. Generic equations for each approach are:

Independence or perfect correlation

$$E\ B-M\ = E\ B\ - E\ M \quad \text{(Eq 5.5)}$$

Independence

$$Variance\ B-M\ = Variance\ B\ + Variance\ M \quad \text{(Eq 5.6)}$$

Perfect correlation

$$Variance\ B-M\ = Variance\ B\ + Variance\ M\ - 2\times\sqrt{Variance\ B}\times\sqrt{Variance\ M} \quad \text{(Eq 5.7)}$$

These equations provide the expected value and variance of the difference using the two approaches. It may be appropriate to assume some parametric distribution for change in adverse human health outcomes; in that case these moments can be used to determine that distribution's parameters. The results assuming independence and perfect correlation provide boundaries for the more accurate predictions that could be generated using parallel calculation. In some cases, the bounds may not be sufficiently different to cause concern but in other cases this analysis may suggest the need to invest in the development of a parallel model structure.

5.3.3 What is Sensitivity Analysis?

Sensitivity analysis examines the relative influence and importance of a model's inputs on its output (see Section 6.7). A well-designed exposure assessment model should comprise inputs that influence exposures, so the important idea of sensitivity analysis is measuring the 'relative' influence. Sensitivity analysis may be completed as part of an exposure assessment or it may be done as part of risk characterization.

There is no universal standard for conducting sensitivity analysis (Frey et al., 2003). In fact, multiple approaches may be legitimately used because each approach may examine a different type of influence. The typical sensitivity analysis examines the magnitude of change in exposures for some change in inputs (i.e., $\frac{\Delta output}{\Delta input}$). One challenge is determining how a change in exposures should be measured. In some cases,

change in the average dose value may be satisfactory (e.g., measuring conditional expected dose). In other cases, the change in the variability of doses may be of interest. If sensitivity is measured using analysis of variance techniques, then changes in the average and variance can be assessed together (Frey et al., 2004). Correlation or other quantitative measures of association (e.g., spider plots, tornado charts) are commonly available in commercial software packages used for Monte Carlo simulation (Vose, 2008).

To proceed with sensitivity analysis, it is important that the objective of the analysis is clear to the analyst. This objective can be informed by the overall purpose of the risk assessment. Sensitivity analysis for its own sake is rarely rewarding when applied to complex models and often a frustratingly inefficient use of the analyst's resources. For a focused exposure assessment, with specific purposes and scope, it is likely that focused analysis on specific components' importance and influence (examined in multiple ways) may be more useful for risk managers.

The major challenge for sensitivity analysis is that it is difficult to separate sensitivity from uncertainty in most exposure assessment models. Uncertainty about model components can result in a very mixed description of the importance of model components. Theoretically, a model input may be highly influential across part of its uncertainty range but much less influential across another part. At the least, acknowledgement of such discrepancies should be communicated to risk managers. Again, focused sensitivity analysis facilitates deeper analysis of a few components instead of superficial analysis of many components.

5.3.4 What is an Uncertainty Analysis?

The goal of uncertainty analysis is to identify those model inputs whose uncertainty substantially contributes to the total uncertainty about exposures implied by the exposure model (see Section 6.7). Uncertainty is a lack of knowledge; therefore, uncertainty can only be reduced via accumulation of new knowledge. Uncertainty analysis suggests where to focus future data gathering efforts and/or scientific research. Like sensitivity analysis, there is no standard method for conducting uncertainty analysis.

Although the objective of uncertainty analysis differs from sensitivity analysis, the results of uncertainty analysis are usually not independent of the results of sensitivity analysis. If uncertainty about an input contributes substantial uncertainty about the model's results, then that input usually is also likely to be identified as highly influential through sensitivity analysis.

Random variables and parameters in an exposure model are subject to parameter uncertainty (Morgan and Henrion, 1990). This source of uncertainty refers to sampling and measurement errors that are inherent to empirical data. Statistical techniques may be available to quantify parameter uncertainty.

Uncertainty about model structure is another source of uncertainty that may be propagated through an exposure assessment. This source may refer to use of surrogate variables, model simplifications or alternative specifications of the model processes. An example of the latter reference might be uncertainty about whether to include a process (e.g., cross-contamination) in an exposure model. It is often difficult to quantify the magnitude of uncertainty about model structure. Nevertheless, such uncertainty can substantially alter exposure predictions from a model. Uncertainty associated with model specification can be investigated by testing the differences in fit and predictions of multiple model forms. If the models are of the same general form (e.g., exponential family), then the effects of including or excluding covariates can be evaluated using likelihood-based criteria such as the Akaike information criterion.

If economic information is combined with uncertainty analysis, a value of information analysis approach may yield insights relevant to the goal of uncertainty analysis. Yet, these methods have rarely been employed in microbial exposure assessment. For a simple dichotomous decision, value of information methods can evaluate the economic returns of hypothetical new information relative to the choice made prior to acquiring the new data. These methods highlight an important point about new information; if additional data will not change a decision, then those data are not valuable. Therefore, risk managers can acknowledge that their decisions hinge on the degree of uncertainty about results from a model. Newly acquired data will presumably reduce that uncertainty, but its value may be naught if risk managers were not influenced by the magnitude of uncertainty about the original results.

The techniques for uncertainty analysis are similar to those for sensitivity analysis. A factorial design for uncertainty analysis enables evaluation of how alternative values for uncertain model inputs influence the model results. For example, if there are k uncertain inputs and two realistically extreme values (i.e., high and low) for each input are proposed, then the model can be calculated 2^k times to examine the influence of each input's uncertainty on the results (ECSSC, 2003). This approach supports examining interactions between the inputs. For example, one extreme value of one input may slightly influence the model's results, but when the model is calculated with that extreme value and certain other extreme values of other inputs, the magnitude of its influence substantially increases.

Analysis of uncertainty is daunting if the number of uncertain parameters or alternative model structures is large. A simple, univariate alternative to the 2^k factorial design is to calculate results for each of the two extreme values of each input independently; this only requires 2k re-calculations of the model (ECSCC, 2003). The predicted change in results for each input can be graphically displayed to demonstrate their relative influence on the model's results.

5.4 What Can I Put Into an Exposure Assessment Report?

Communication of exposure assessment results is challenging because the intended audience is usually diverse. A presentation of these results has to satisfy technical specialists as well as those who are less specialized in quantitative methods (ECSCC 2003, WHO/FAO, 2008). Consequently, a balance between technical details and general conclusions of the analysis is a goal of any communication (see Chapter 8).

The credibility of an exposure assessment is enhanced through peer-review and feedback from public outreach. If communication of the exposure assessment is reasonably transparent and balanced, reviewers can focus their attention on the merits of the analysis and contribute to improving its accuracy and validity.

The output of an exposure assessment is usually an exposure distribution. Thoughtful contemplation of the best formats for communicating this distribution and its uncertainty should precede any final decision. A number of graphical formats for presenting risk results are available (Vose, 2008). Tabulated results can be more useful in some cases and both table and figures may be needed in other cases. A limited number of moments (e.g., mean, variance, skewness) along with meaningful quantiles of this output are routinely provided in a report. You can consider different preferences for data display when selecting the best format. All tables, graphs, and other figures should have clear, concise narrative text to help the reader understand what is being presented.

If specific processes included in the exposure assessment are the subjects of risk management choices, then you should summarize outputs from these processes in the report. Plots of the central tendencies of microbe counts that illustrate trends across the breadth of a model can be useful for some readers.

To clearly communicate the scenarios considered in the exposure assessment, you can illustrate diagrammatically the conceptual model in the report. A well-annotated conceptual model will enhance reader's understanding of the analytic approach taken.

You can list all inputs used in the model and clearly define their reference symbol and name, describe their purpose in the model, provide the probability distribution name and parameter values for random variables, and explain how you handled uncertainty for variables and parameters. You can communicate important inputs by using graphical depictions of their distributions.

Include the mathematical structure of the model transparently, but concisely, in the report. Because it will only be useful to a specialized audience, it is usually appropriate to place the mathematics in an appendix. Nevertheless, this mathematical description of the model will provide the greatest opportunity for clearly explaining the exposure assessment to the specialized audience. This is the audience that can verify the validity of the model or identify errors in its logic or assumptions.

Inclusion of a discussion of important assumptions is necessary. A transparent treatment of assumptions improves the reader's understanding of the analysis; although some might disagree with assumptions made, it is much preferred that they understand the reasons for the assumptions. Models are always imperfect depictions of reality and their results are always conditioned on assumptions. The strength of a model, then, is based on the strength of the justifications of its assumptions.

Presentation of the results of sensitivity and uncertainty analyses in tabular or graphical formats is also helpful and may be required depending on the use of the risk assessment. It is crucial that such presentations are meaningful to both a specialized and non-specialized audience. Complex analyses that do not illuminate important conclusions will create confusion for the reader.

Exposure assessments can generate large volumes of analytic output. Nevertheless, presentation of the exposure assessment requires deliberation by the analyst; the reader of a report expects that care is taken in what is presented and how it is presented. Avoid arbitrary decisions about the content of the report.

5.5 What are Possible Future Developments in Exposure Assessment?

The discipline of microbial exposure assessment continues to grow and evolve. Compared to more established analytic fields like economics, epidemiology or statistics, microbial exposure assessment is still relatively new. Therefore, it is expected that methods and approaches will continue to improve.

One technique of increasing interest for exposure assessment is the use of MCMC simulation (Gelman et al., 2004). The MCMC method is based on Bayes Theorem, but it is designed to solve problems that would normally be intractable using standard Monte Carlo approaches. Using MCMC methods, "prior distributions" are specified for model inputs and the empiric evidence is integrated with the model to determine which combinations of model inputs best describe—in a probabilistic sense—the empiric evidence.

Many exposure assessments are generated to guide government policy development. Traditionally, risk assessments are conducted independent of economic analyses, but risk assessment products are commonly incorporated into economic analyses conducted to support government policy (Williams and Thompson, 2004). In the future, it is expected that economic analysis will be integrated into the risk assessment to provide more accurate and useful policy information.

Within exposure assessment models, costs and tradeoffs between alternative processes could be incorporated so that their effects on the model's results were explicitly calculated. For example, it might be the case that a process with variable effectiveness is associated with extraordinarily high marginal costs at very high levels of effectiveness, but low or absent costs at moderate or low levels of effectiveness. To fully appreciate

such phenomena, the exposure model needs to incorporate the economic factors. Otherwise the effects of these economic factors may not be understood by risk managers.

As mentioned for uncertainty analysis, VOI methods are very useful for determining economically-efficient future data gathering efforts (Yakota and Thompson, 2003; Disney and Peters, 2003). In the future, it is expected that these methods will become standard for exposure and risk assessment analyses.

Exposure assessment models can serve as templates for the incorporation of better data when it becomes available. In the future, exposure assessments may be updated with data collected explicitly for the purpose of exposure assessment. Traditionally, exposure assessments use data generated for other purposes. Risk assessors then must struggle with incorporating such data into an exposure assessment. If data are collected for an exposure assessment, however, it is likely that the data will be better structured for that purpose. The result of using data that is "fit for purpose" is improved exposure estimates for risk management decisions.

MRA deals with many different types of problems; there are a number of approaches and methods that may provide satisfactory solutions. Nevertheless, exposure assessment will continue to evolve towards standard approaches and methods that represent accepted defaults for certain types of problems. Increasing academic attention and scrutiny of exposure assessment will almost certainly bring greater consistency in methods. Because the field of MRA is still emerging, it is also critical that risk assessors search for reasonable precedents in methodology when embarking on a new exposure assessment project. If precedents are adopted and improved, standardization of methods and models can eventually occur.

International groups such as the WHO/FAO and Codex Alimentarius will continue to provoke thoughtful discussions about the most appropriate exposure assessment methods and approaches. Such discussions will contribute to improved standardization of exposure assessments in the future.

6. RISK CHARACTERIZATION

As introduced in Section 1.1, risk characterization is one of the four fundamental components of risk assessment. Risk characterization is the integrating component of the risk assessment process that "characterizes" or describes and summarizes microbial health risks. It is the final integrative step of risk assessment. This chapter addresses what a risk assessor can do to take the pertinent elements from the previous components of the risk assessment (hazard identification, hazard characterization, dose-response, and exposure assessment) and integrate them into a coherent, understandable, and informative conclusion that is useful for decision makers and stakeholders. Further, the risk characterization discusses scenario, model, parameter, data, and analysis options that risk managers should understand and consider when interpreting the results of the risk assessment. The risk characterization, in a sense, brings to full circle the initial planning and scoping for the risk assessment described in Chapter 2. The content of the risk characterization is intended to reflect the issues and questions laid out during planning and scoping.

For further detail and discussion on risk characterization, good references are the NRC reports (NRC, 1983, 1994, 1996, 2009), EPA's *Risk Characterization Handbook* (EPA, 2000a), and *An Examination of EPA Risk Assessment Principles and Practices* (EPA, 2004b). This chapter is not intended to provide the level of detail about risk characterization provided in the previous references, but rather provides the microbial risk assessor with guidance on what information to include and how to integrate the information from the previous three chapters.

6.1 What is Risk Characterization?

In its most general sense, risk is the possibility (and if estimated, probability) of suffering harm. For the purposes of MRA, hazard may be causal or associated with adverse outcome as a representation of intrinsic effects expressed by a microbe. Risk contains elements of both hazard and exposure. Thus, risk is generally understood to be the integration of intrinsic effects, represented by Hazard, and the values for Exposure. It is usually represented by some form of the equation:

$$Risk = f_{Hazard} \bullet f_{Exposure} \qquad \text{(Eq. 6.1)}$$

Hazard identification allows you to select and focus on specific features of subject organisms associated with the potential to cause harm. Exposure analysis provides a description of the routes and an estimate of the degree to which a host may be exposed. None of these components can stand alone to characterize risk. When combined with host factors in an evaluation of dose-response, one can get a quantitative Hazard Characterization of that potential once a host is exposed. Risk characterization takes the specific identified hazards, examines the probabilities of their existence under specific exposure scenarios, and combines those probabilities with those of the likelihood that the agent will encounter the host in sufficient quantity to cause an effect.

Risk characterization is the final step of the MRA process in which all preceding data collection and analyses are combined to convey the overall conclusions about potential risk to humans. During risk characterization, the results of the iterative risk assessment process are integrated and documented in a descriptive risk characterization summary. Risk characterization communicates the key findings and the strengths and weaknesses of the assessment through a conscious and deliberate transparent effort to bring all the important considerations about risk into an integrated analysis by being objective, transparent, clear, consistent, and reasonable (OMB, 2007b; EPA 2002b; EPA 2000a). For these reasons, the risk characterization needs to be complete, informative, and useful for decision-makers. Therefore, this section of the risk assessment needs to be both sufficiently technical to be scientifically accurate, taking into consideration the uncertainties and reporting the assumptions but also comprehensible by an educated lay audience. As noted below this is the component that most directly leads to a regulatory/management decision and serves as communication tool for stakeholders.

Risk characterization describes the ways in which exposure and dose-response (quantitative) or exposure and hazard assessments (qualitative) are used together to formulate a statement of risk. Risk characterization can be quantitative, when values are available for all terms in the risk equation, or it may be semiquantitative, when only some values are available. In many cases, default values/assumptions based on known conditions are used in place of measured ones. Further, when the data are not useful to present a quantitative estimation of risk, then a qualitative description of the risk may be all that can be presented in a risk characterization, which may be sufficient in certain cases. Regardless of quantitative versus qualitative, the risk characterization should address the risk management questions posed in the planning and scoping phase (and any questions that may have been added or revised during the assessment itself).

Moreover, risk characterization forms the starting point for formulating risk management considerations and provides a foundation for (regulatory) decision-making. It characterizes both quantitative data and qualitative information in technical and non-technical terms, explaining the extent and weight of evidence, results, and major points of interpretation and rationale. It also summarizes the strengths and weaknesses of the evidence, conclusions, uncertainties, variability, potential impact of alternative assumptions, and discusses scenario, model, parameter, and analysis options that may deserve further consideration as the results from the assessment are subsequently used for decision making purposes.

As an example of how one agency views risk characterization, EPA's Policy Statement on risk characterization is as follows:

> Each risk assessment prepared in support of decision-making at EPA should include a risk characterization that follows the principles and reflects the values outlined in this policy. A risk characterization should be prepared in a manner that is clear, transparent, reasonable and consistent with other risk characterizations of similar scope prepared across programs in the Agency. Further, discussion of risk in all EPA

> reports, presentations, decision packages, and other documents should be substantively consistent with the risk characterization. The nature of the risk characterization will depend upon the information available, the regulatory application of the risk information, and the resources (including time) available. In all cases, however, the assessment should identify and discuss all the major issues associated with determining the nature and extent of the risk and provide commentary on any constraints limiting fuller exposition. (EPA, 2000a)

6.2 What are the Elements in a Risk Characterization?

During the risk assessment process, you should have identified areas where policy options were considered, where management decisions and assumptions were made, and where uncertainties are important. The point of risk characterization is not to reiterate the details of each chapter of the risk assessment, but rather to integrate those chapters to arrive at the risk assessment output (e.g. risk estimate, risk ranking, or other output), describe the relevant findings, cross-reference the exposure and dose-response assumptions (e.g., do the age grouping in exposure assessment and dose-response assessment match), and discuss other salient elements (as described below).

Risk characterization consists of two principal steps—risk estimation and risk description. Risk estimation is the compilation of the types and magnitude of effects anticipated from exposure to the microbe or medium and can be qualitative or quantitative depending on the data and methods used. The risk estimation is derived from the output components of the risk assessment (hazard identification, hazard characterization, exposure assessment, and dose-response analysis). Specifically, the results from the characterization of exposure can be expressed as the number of organisms to which an individual is exposed in a defined amount of time and/or for a certain consumption rate. Resultant estimates of the potential for adverse human health effects can be expressed as an individual risk estimate (e.g., 1 per 1000 probability of illness) or as a population level risk estimate (100 illnesses per year in a region with a population of 100,000 individuals). As described in further detail below, the risk estimation can also be modeled to consider time-dependent elements such as secondary (person-to-person) transmission, host immunity, and multiple routes of exposure (ILSI, 2000).

The second component of risk characterization, risk description puts the risk estimation into context by summarizing the event of interest according to its nature, severity, and consequences and discussing and quantifying (to the extent possible) (1) the uncertainties associated with the key components within the risk characterization; (2) the variability associated with key inputs to the model(s); (3) the confidence in the resulting risk estimates through a weight of evidence discussion; (4) the limitations of the analysis; (5) the critical assumptions; and (6) the plausibility of the results. Clearly, your professional judgment will also be necessary to determine what should be included in the risk characterization. You can consider the following elements in risk characterization (adapted from the EPA *Risk Characterization Handbook*, EPA 2000a):

a) **Key information** – Consider 1) the studies available and how robust they are; 2) the major risk estimates you calculated, the assumptions and the extrapolations made during the estimated risk calculation, and the residual uncertainties; 3) the use of default parameter values, policy choices and risk management decisions made, if any; 4) whether the key data used for the assessment are considered experimental, state-of-the art, or generally accepted scientific knowledge; and 5) variability.

b) **Context** – Consider 1) how you are addressing the risk management questions; 2) how the estimated risk from this microbial hazard compares to other estimates for this hazard, if available. Include discussion of regulatory requirements or if there are regulatory values to consider.

c) **Sensitive Populations** – Consider 1) the range of people that may be affected, including innately susceptible populations (e.g., ethnic groups, gender, socioeconomic and/or nutritional status, other genetic predisposition) and those that are highly exposed; 2) a quantitative characterization for each sensitive population may not be necessary or possible. For example, where there are many sensitive population groups for a given microbial hazard, it may be sufficient to estimate risks for the most sensitive group, and then assume that as long as that group is protected, other groups may be protected adequately. If the quantitative portion of the risk assessment is strongest for the general population due to data availability, then some data-based adjustment for sensitive populations may be considered. Both results can be presented and discussed.

d) **Life Stages** – Consider the age groupings evaluated and note any life stages that may have particular vulnerability due to behaviors or situations that influence exposure patterns and/or innate susceptibilities. For microbial hazards with only short term effects the different life stage may be treated as sensitive populations. However, if any longer term effects (e.g., health endpoints that span a 70 year life) are of interest then life stages may need to be considered differently than sensitive populations. For example, everyone in the general population passes through childhood life stages and exposures to pathogens could vary at different childhood life stages. Thus, depending on the scope of the risk assessment, consideration of childhood life stages may be necessary as risk estimates may vary for the range of important life stages.

e) **Scientific Assumptions** – Describe, 1) where key data gaps exist; 2) what the key assumptions you used during your assessment are and the impact they may have on the assessment outcome. Also note if there is precedent in other risk assessments for the approach or assumptions employed, and note the justifications for selection of any default parameter values that are used.

f) **Policy Choices** – Describe, 1) if your office has different policies about how to assess risk (e.g., different uncertainty factors or different levels of regulatory

concern); 2) any policy choices that may bound the scope of the assessment. If appropriate, include discussion of consistency with other Agency approaches or decisions.

g) **Variability** – Describe 1) how variability arises from true heterogeneity in characteristics such as dose-response differences within a population, or differences in microbial levels in the environment; 2) if the values of some variables used in an assessment can change with time and space, or across the population whose exposure is being estimated. The discussion of variability should link to the discussion of assumptions, because variability considerations may be lost or assumed when values for parameters are selected. This element is critical and discussed at length in the previous chapters. An inadequate discussion of variability can result in a loss of transparency or in the worse case a misleading risk assessment.

h) **Uncertainty** – Describe 1) what uncertainties exist in the assessment (e.g., measurement uncertainty, model uncertainty, uncertainty due to data gaps); 2) how you addressed uncertainty (e.g., uncertainty analysis, sensitivity analysis); 3) what impact reducing scientific uncertainties could have on your assessment; 4) where possible, quantitative uncertainty analyses of the data should be presented; at a minimum, a qualitative discussion of the important uncertainties should be provided. This element is critical and discussed at length in the previous chapters (additional references: Morgan and Henrion, 1990; Frey et al., 2004). As with variability, an inadequate discussion of uncertainty can result in a loss of transparency or in the worse case a misleading risk assessment.

In practice it is often difficult to separate variability and uncertainty, because many uncertainties are in the area of characterization of variability (e.g. lack of data for certain conditions). In cases where there is uncertainty about variability, be sure to be clear about your use of the two terms.

i) **Bias and Perspective** – Consider 1) that in the light of uncertainty and default choices, your agency may proceed in the direction of more public health protection compared to less protection. If this is the case, this perspective needs explicit explanation; 2) it is not always clear where agency/public health bias enters into your agency risk assessment - to the extent it may make a difference in the outcome of your assessment, highlight the potential bias so the impact will not be overlooked or misinterpreted by the risk manager. For example explain the implications of selecting a 50^{th} versus 95^{th} percentile in a data set.

j) **Strengths and Weaknesses** – Throughout the risk characterization you should highlight and/or describe 1) major imbalances among the components of the assessment, e.g., the case for the microbe posing a hazard may be strong, while the overall assessment of risk is weak because there are no data about whether there is exposure to the microbe; 2) the data/information that present the strongest evidence for your conclusions as well as the data that may not be as strong

(weakness) and how you incorporated that into your conclusions. You should also discuss the quality of the data used and how the data quality pertains to variability and uncertainty.

k) **Key Conclusions** – Describe 1) the key points that need to be communicated for knowledgeable interpretation of the risk assessment; 2) that small subset of key findings supporting information (strengths and weaknesses, results from sensitivity/uncertainty analyses) that really makes a difference in the assessment outcome.

l) **Alternatives Considered** – Consider 1) if there are plausible alternatives to the risk estimated in your assessment and how you dealt with those alternatives (e.g., alternative models that could be used, different hazard pathways); 2) the limitations of making comparisons among the alternatives; 3) where appropriate, how your conclusion about risk compares to other possible risks. If other risks are compared, the discussion should highlight the limitations of such comparisons as well as the relevance of the comparisons

m) **Research Needs** – Describe the key data needs and/or methodology gaps that were identified during the course of the risk assessment.

Each element described above is important to address in a risk characterization; however, no single element is necessarily more "critical" than others. As the risk assessor, you need to be aware of all these elements and address them appropriately in the risk characterization. For each element you considered describe the data you found, or if you found no data or information for a particular element, or you used a default assumption that also needs to be stated.

6.3 How Do I Prepare a Risk Characterization?

Because the purposes of MRA vary, the mechanisms of risk characterization, including risk integration, can also be quite varied. Some organizations prefer to attempt separate hazard and exposure characterizations, and combine the results once all the pieces are described and quantified. Others find that it is valuable to have feedback from the major assessment components throughout the assessment timeframe, and thus have some elements of risk integration running simultaneously with the component assessments. Hybrids of these approaches are possible, as well. Your selection of an approach to characterization and integration may be driven by institutional practices, completeness of data, and/or timing of availability of data.

It is common to have an iterative process in which a simple scoping characterization (e.g., risk profile) is generated early in the process. This allows the assessors to identify data gaps, to recommend additional data generation or gathering, or to use default assumptions as a starting point to evaluate the potential implications of scenario being evaluated. Later iterations may adjust the scope of the characterization by focusing in on the most important hazards or exposures identified in the initial iterations,

or expand as needed when additional hazard or exposure elements are identified or additional data are provided.

As discussed in Section 2.7 and Chapter 7, there are many different levels of decisions that need to be made during risk assessment. As the assessor, you take responsibility for decisions involving scientific judgment. Other decisions that are ultimately policy calls, are informed by science, but will most likely be made by risk managers. Risk managers may want to evaluate policy decisions and make revisions during different iterations of the risk assessment. Any changes in policy decisions or scope can be tracked for transparency. It is unlikely that scientific judgments will change unless more data become available or compelling scientific arguments are made during internal or external peer review. It is not unusual for risk managers to adjust policy decisions or refine the scope or questions for the risk assessment during the risk assessment process. In many ways, this process highlights the importance of the iterative nature of risk assessment. You should be prepared for these types of changes and provide clear documentation to help delineate scientific judgment from policy decisions. Very often something that could be scientific judgment if data were available becomes a policy decision when data are lacking. Determining the threshold between those two situations can be unclear.

Some of the driving forces for this level of variation in performing risk characterization are available resources, time constraints, legal requirements, and Agency culture. In some cases, a first attempt at a complete risk characterization, in the sense that all evaluation elements are accounted for to at least a limited extent, is desirable to provide managers with an initial scoping of likely outcomes before all information has been fully evaluated. Sometimes, certain factors are given precedence at the outset of an assessment, such as a particular exposure scenario, and interchange between the hazard and exposure components can take place throughout the course of an assessment. This is generally the case when examination of a specific health effect or exposure model is the driving force behind the risk assessment. In other cases, it may be necessary to isolate the hazard and exposure components to avoid results from one having an undue influence on the assessment of the other. This may happen if the type of assessment is intentionally broad and the desire is to not eliminate scenarios, or emphasize specific effects until a first pass is made. This can result in an iterative assessment, with feedback among the components occurring during later iterations.

In selecting an approach for risk characterization, be careful to ensure that any simplifying assumptions that are employed are in fact appropriate and clearly identified. Within that context, and to the extent possible, you should demand higher quality input data and fewer simplifying assumptions when seeking increased accuracy and precision from your risk assessment. From a modeling perspective, biological “realism” is often counter-balanced by analytical or computational complexity. The increase in the complexity of a model structure can increase variability and/or uncertainty due to increased needs associated with model specification (EPA, 2004c). On the other hand, a simpler model involves implicit or explicit assumptions that may or may not be realistic or appropriate for a particular situation. More complex models should be considered or

used under conditions in which the added complexity may provide sufficient additional insight that the additional complexity is warranted (King et al., 2008; Soller and Eisenberg, 2008). As discussed in Chapter 5, statistical methods, such as use of the Akaike Information Criterion or similar likelihood-based measures, to judge the desirable level of complexity in statistical models. Representative MRA model forms are discussed in Section 6.5.1.

6.4 Are All Risk Characterizations Quantitative, and What Do I Do When Quantitative Data are Unavailable for Some Elements of the Risk Characterization?

While many assessments are quantitative, risk characterization can also be qualitative and/or descriptive. It is also possible that some parts of the analysis can be quantitative, while only approximations and/or defaults are possible for other elements (this is sometime referred to as a semi-quantitative analysis).

Although "risk" is often thought to imply a probability of an adverse health effect, and therefore thought of as quantitative, some assessments can only be expected to approximate risk probabilities. In certain cases, the risk characterization can at best be thought of as a screening exercise, providing only a sense of whether the risk might be judged high, medium, or low. Sometimes one can obtain values for some of the components of hazard and exposure characterization, but not all. In these cases, it may be possible to utilize default values for the missing data elements to provide enough information to establish limits of risk for the organism and conditions in question. A bounding analysis may be appropriate. For example, you can do a deterministic analysis with plausibly conservative values for the unknown parameters and see if the resultant risk is above the level of concern. If it is not above the level of concern no further data on these parameters is probably necessary. If the risk is above the level of concern, then you have found out that gathering more data on the unknown parameter value(s) would be justified.

Finally, another type of assessment that can be qualitative is a relative risk assessment. These types of assessment have been valuable for numerous agencies, especially for evaluating the potential benefits of management actions (treatments) or alternatives in conditions where rigorous and quantitative data were not available.

6.5 Are There Different Forms of Risk Characterization? When Do I Apply Them?

Risk characterization should be complete, informative, and useful for decision-makers. The appropriate level of detail for any particular assessment will be a function of the goals of the assessment, the questions that the assessment are intended to answer, and the data that are available to conduct the assessment. Whether or not a particular level of detail is appropriate for a particular situation will depend on the purpose of the assessment.

When hazards are well defined and specific exposure scenarios allow accurate exposure and dose-response calculations, a quantitative risk characterization may be in order. This is generally the case with event-driven retrospective assessments. Prospective assessments often lack sufficient information and data to make the appropriate calculations for detailed quantitative assessments. In some cases, default assumptions and/or parameter values can substitute for measured values so that calculations can be made, but with considerable uncertainty. Qualitative risk characterization may be useful, when risk management choices need to be made and a general sense of risk is all that is required by managers. For example, if default values, coupled with an understanding of the uncertainty that accompanies their use, can enable completion of a semi-quantitative assessment that gives enough information for decision making, then that assessment is more useful than the failure to produce any assessment, due to the inability to cope with the lack of data for which the default assumptions substitute. Finally, the appropriate form of risk characterization may change during the risk assessment, if questions to be answered change or if additional data become available.

The availability of data and the appropriate type of risk characterization are generally related. Assessments of specific events can sometimes result in precise calculations of exposure and dose-response, assuming the agent is well identified. In some prospective analyses, estimates of organism concentrations in a particular source may be easier to estimate than the actual exposure of particular populations. In these types of cases, default values may need to be substituted for accurate population estimates. Similarly, dose-response relationships are only available for a limited range of organisms (refer to dose-response chapter for a list of available dose response relationships), so quantitative assessments can only be conducted for those microbes that have known (or derivable) dose-response relationships.

With respect to quantitative risk characterization, you can employ a variety of model forms for the assessment of infectious disease transmission and the potential impact or benefit of intervention efforts/management actions. Particular characteristics of each model form allow for the capture of different aspects of the disease transmission system (EPA, 2004b). The two most commonly employed classes of MRA models are static and dynamic models. An overview of these types of models along with pros and cons of each is provided by Soller and Eisenberg (2008). Exclusion from the following discussion does not preclude use of a particular model form; however, justification for use of a particular model form should be included in the risk description.

For examples of risk characterization within MRA you can refer to Foodrisk.org for a searchable database of MRAs.[40]

[40] http://foodrisk.org/risk_analysis/RA/RAs.cfm

6.5.1 When is a Static Model Appropriate?

A static model would be appropriate in those cases where the central question is concerned with the probability of infection or illness relative to the dose of pathogens acquired from a single exposure. Such models can handle complex details about the course of events that lead to exposure and infection and can be analyzed by well-established statistical techniques that require fewer assumptions than do dynamic models (discussed below). Static models are useful for analyzing situations where the effect of an intervention directed to individuals (e.g., point-of-use remediation) is more important than the effect on transmission throughout the population; they are not appropriate for measuring indirect effects at the population level (e.g., the effect of water treatment interventions on risk due to secondary transmission).

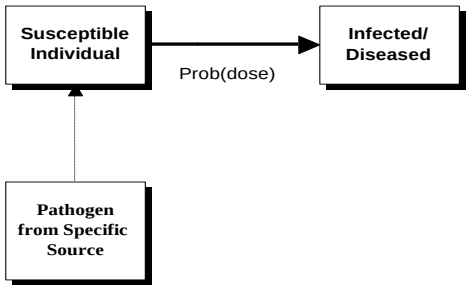

Figure 6.1 Static Risk Assessment Conceptual Model

Some infectious diseases are not readily transmitted from person to person but are acquired, to the best of current knowledge, only by consumption of or contact with contaminated environmental materials (e.g., *L. monocytogenes* from food, *Naeglaria fowleri* infection from water). In other cases, although an agent may have the potential to be transmissible, the particular situation is such that the person-to-person component is unknown or thought to be negligible. Understanding the pattern of human infections from such pathogens or exposure scenarios may be best achieved through the use of static models (parallel to those used for toxicological risk assessments) (Figure 6.1).

These chemical risk assessment-based models are used to estimate risk at an individual level and typically focus on estimating the probability of infection or disease to an individual as a result of a single exposure event. With respect to microbial contaminants in the environment or a particular media (food, water), a fundamental simplifying assumption of static model-based analysis is that exposure events and infection/disease are independent; that is, the outcome from one exposure event does not affect a subsequent exposure, and one individual's outcome has no impact on any other individual's outcome. Thus, secondary transmission and immunity are most often assumed to be negligible or are of similar magnitude and effectively cancel each other out. (It is generally assumed that secondary transmission would increase the level of infection/disease in a community relative to a specific exposure to pathogens, and

immunity would decrease the level of infection/disease in a community relative to a specific exposure to pathogens.)

6.5.2 When is a Dynamic Model Appropriate?

Risk managers and regulators are often concerned with risk on a societal or population scale. Thus, individual risks need to be translated to the level of the exposed population or some other relevant part of that population. When an infectious agent that occurs in the environment or a particular media is also contagious, its impact on a population can be significantly influenced by the interactions between contagious and susceptible individuals. To assess the full impact of human exposure to pathogens, you should consider addressing risk at the population level in addition to individual risk at the dose-response level. For a thorough evaluation of risks that are manifest at the population level, MRA methods should explore the relative importance of secondary transmission and immunity, and thus capture and integrate the dynamic interplay of hosts, agents, and environments.

Dynamic MRA models take two main forms: deterministic or stochastic. "Deterministic" means that the model output is strictly determined by the starting conditions and the values of the parameters in the equations that define the system. In stochastic models, events are treated as stochastic (random) events rather than deterministic ones. Deterministic dynamic MRA models are suitable for large populations of individuals randomly interacting with one another. In this form, the population is divided into epidemiological states such as: (1) susceptible, (2) diseased (infectious and symptomatic), (3) carrier (infected but asymptomatic), and (4) immune (partial or complete). Only a portion of the population is in a susceptible state at any point in time, and only those individuals in a susceptible state can become infected through exposure to pathogens. The dynamic aspect of the model means that members of the study population move between epidemiological states at different rates, and thus, the number of individuals in each state changes over time. A representative conceptual model for this type of MRA model is presented in Figure 6.2.

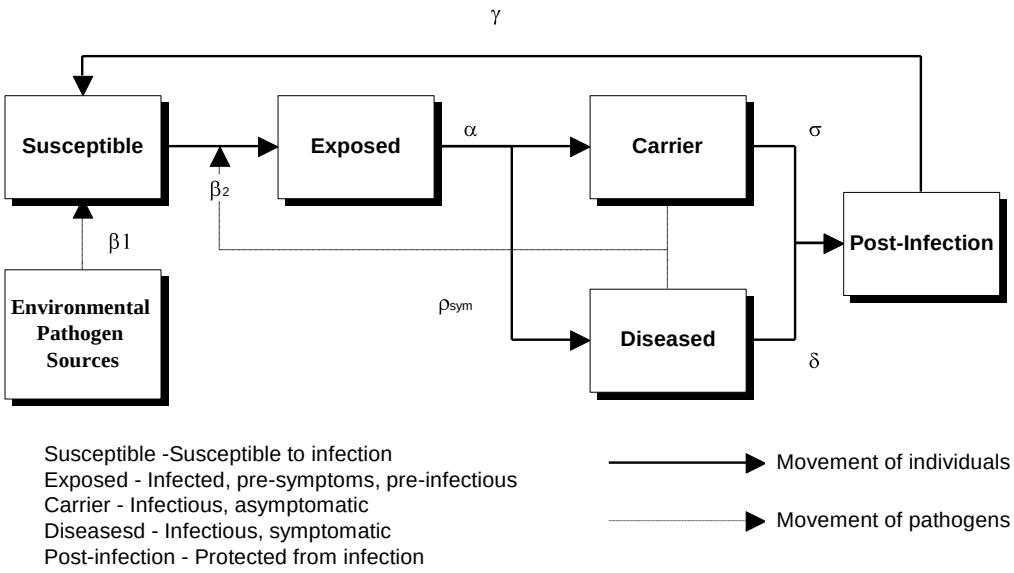

Figure 6.2 Dynamic Risk Assessment Conceptual Model
(Source: Soller, 2009; Soller and Eisenberg, 2008)

Deterministic dynamic MRA models are expressed mathematically as a set of differential equations. These equations describe the rate of change in the number (or density) of individuals in a particular state (or compartment) over time and have defined parameters and starting conditions. Deterministic dynamic MRA models have a number of limitations. If they are used to model relatively small populations, the assumption of homogeneous mixing of the individuals in the population can lead to miss-estimation of disease. These models also require appropriate parameter values for transmission rates and some of this information can be quite difficult to determine accurately. Lack of knowledge and data, as well as inherent biological variability, suggest a need for uncertainty and sensitivity analyses of parameter values. Furthermore, random events such as local introduction or local die-out of a disease in a neighborhood of a heterogeneously mixing population are difficult to incorporate into these models (EPA, 2004b).

In a stochastic form, dynamic models incorporate probabilities at an individual level and are evaluated by an iterative process (e.g., susceptible person A has a probability of contacting person B, who has a probability of being infectious). This type of model also uses states for classifying the epidemiological status of the population and subpopulations under study, but differs from the deterministic dynamic MRA models in that the compartments contain discrete individuals rather than the numbers or densities of persons that are represented by the compartments in deterministic dynamic MRA models.

In stochastic dynamic MRA models, events are treated as random (stochastic) events rather than deterministic ones. These models employ distributions of outcomes rather than the average outcomes as do the deterministic models; a stochastic model will

produce different results each time it is run. Stochastic forms are suitable for small populations and heterogeneous mixing patterns where stochastic events can have a major impact. In a small population, chance events, such as an infectious person contacting only immune persons during the infectious period of illness, may have a substantial impact on the transmission dynamics of the disease (EPA, 2004b).

Based on this information, risk characterization can be thought of as modular, with different modules requiring specific data for calculations. Some modules require detailed data, while others may only require default estimates or descriptive information. Risk management options may obviate the need for precise data on exposure components, if exposure can be limited by specific actions. Thus, the appropriate module (and corresponding level of detail) for any particular assessment will be driven by the goals of the assessment, the questions that the assessment are intended to answer, and the data that are available to conduct the assessment.

6.6 How are Sensitivity and Uncertainty Analyses Related to Risk Characterization?

The discussion in this section is limited to data sensitivity and uncertainty and does not include sensitivity and uncertainty in overall decision-making, which may consider decision-maker judgments and values beyond the risk assessment.

Although uncertainty and variability are different, in practice it can be difficult to separate the two, particularly when uncertainty about variability is important (Section 6.2). It may be practical to characterize uncertainty and variability together if you clearly describe what you are doing. Uncertainty analysis "is the computation of the total uncertainty induced in the output by quantified uncertainties in the inputs and models" (Morgan and Henrion, 1990). Uncertainty analysis is a key concern for risk managers because it provides information about the overall reliability of the risk estimates. Measures of model "uncertainty" communicate to risk managers the risk assessor's best judgment as to the overall quality of the numerical risk estimates generated by the MRA. Confidence intervals, "credible ranges" developed through Bayesian analyses, and other measures of dispersion in risk should be presented clearly, and their meaning communicated clearly. Similarly, clear graphical or tabular presentations are very useful. To the extent that intermediate calculations add value and understanding to the results, they can also be included. Key assumptions related to model selection, input data, and parameters should be provided and discussed, as well as their implications for the model results and uncertainty. Any conservative assumptions that are built into the model should be explained and the impact of using less conservative assumptions should be discussed.

It is also important to carefully evaluate the impact of known sources of variability in model outputs. This is generally done through use of one or more forms of sensitivity analysis. Sensitivity analysis can also help determine whether more resources should be put into parameter estimation. Sensitivity analysis "is the computation of the effect of changes in input values or assumptions (including boundaries and model

functional form) on the outputs" (Morgan and Henrion, 1990). Sensitivity analyses techniques range from simply conducting a small number of additional model runs with different parameter values to performing a fully probabilistic evaluation of the effects of variations in parameter values on model outputs. The specific approach that is taken will depend on the nature of the data and models supporting a given assessment. USDA identified several sensitivity analytical techniques useful for MRA (Frey et al., 2004). The methods evaluated ranged from simple and intuitive (varying input values across their observed ranges, scatter plots) to more complex statistical procedures (e.g., classification and regression tree [CART]). For any given risk assessment, it is likely that one or more of these methods will be useful for sensitivity analysis.

Although sensitivity analyses are useful for evaluating the effects of the variability in single parameters on risk estimates, when multiple parameter values vary, the results of sensitivity analyses should be interpreted cautiously (EPA, 1997a). If the variations in parameter values are independent of one another, it is easy to overestimate the impact of varying more than one value because using upper or lower percentile values for more than one variable can yield point estimates of risk that are overly conservative or insufficiently protective. If the variability in risk parameters is correlated, the impact of their variations may not be easy to estimate using sensitivity analysis. In such cases, a more detailed and comprehensive analysis may be required, usually employing probabilistic approaches such as Monte Carlo or related simulation techniques. Where the variability in model parameters can be partitioned into components mainly reflecting variability and uncertainty, "two-dimensional" Monte Carlo analysis can be employed to estimate the relative importance of these two components.

The EPA Exposures Factors Handbook (EPA, 1997a) provides several approaches to quantitative uncertainty and sensitivity analysis (Table 6.1):

Table 6.1 Approaches to Sensitivity and Uncertainty Analysis Recommended in EPA's Exposure Factors Handbook (Source: EPA, 1997a)

Approach	Description	Example
Sensitivity analysis	Changing one input variable at a time while leaving others constant to examine affect on output	Fix each input at lower (then upper) bound while holding others at nominal values (e.g., medians)
Analytical uncertainty propagation	Examining how uncertainty in individual parameters affects the overall uncertainty of the exposure assessment	Analytically or numerically obtain a partial derivative of the exposure equation with respect to each input parameter
Probabilistic uncertainty analysis	Varying each of the input variables over various values of their respective probability distributions	Assign probability density function to each parameter; randomly sample values from each distribution and insert them in the exposure equation (Monte Carlo simulation)
Classical statistical methods	Estimating the population exposure distribution directly, based on measured values from a representative sample	Compute confidence interval estimates for various percentiles of the exposure distribution

In addition, Morgan and Henrion (1990) discuss in detail the following four techniques for sensitivity and uncertainty analysis, including:

a) **Deterministic** – One-at-a-time analysis of each factor holding all others constant at nominal values

b) **Deterministic joint analysis** – Changing the value of more than one factor at a time

c) **Parametric analysis** – Moving one or a few inputs across reasonably selected ranges such as from low to high values in order to examine the shape of the response

d) **Probabilistic analysis** – Using correlation, rank correlation, regression, or other means to examine how much of the uncertainty in conclusions is attributable to which inputs

6.7 How are Quality of Life Measures Important in MRA?

Quality of life measures are usually included in cost-effectiveness analyses (CEA) rather than within risk assessment. You should be aware of how your risk assessment results might be used, such as in a CEA. For example, EPA has used quality-adjusted life

years (QALY) and Morbidity Inclusive Life Years (MILYs)[41] in the regulatory impact analysis for the Final Clean Air Interstate Rule (EPA, 2005b, Appendix G) and the LT2ESWTR. (EPA, 2006a, Appendix U).

Quality of life captures the impacts that illness has other than medical costs and lost work hours. It is particularly relevant for chronic illnesses that cause pain, suffering, and a sacrifice in lifestyle. One concept, known as QALY, is a method for assigning a numerical value for quality of life and translating that numerical value to a monetary measure (WHO, 2001). Duration and severity of illness can also be used to characterize quality of life, but these are not expressed in monetary units, so would not be utilized in the same manner as QALYs. Disability adjusted life-years (DALYs) are recommended in WHO *Water Quality: Guidelines, Standards and Health* to integrate the effects of a single agent, compare the health effects of different agents or conditions, and to inform the debate on acceptable risk (WHO, 2001). WHO expects that "DALYs will play an important role in prioritizing risk factors, determining levels of acceptable risk, setting health targets and appraising effectiveness [of policy or mitigation] through examining public health outcome." DALYs and QALYs are not calculated in the same manner and have reversed scales of measure. DALYs measure a health gap, with full health represented as 0 and full disability (death) as 1.0; QALYs measure health expectancy, with full health represented as 1.0 and lowest possible health state (death) as 0 (Airoldi and Morton, 2009; Gold et al., 2002; Rice et al., 2006).

It is important to note that QALYs and DALYs are not objective measures and require a descriptive conceptualization of health states. In addition, there can be significant differences in ranking due to ethnicity, gender, and area of residence (different cities; urban versus rural). Thus, there is much controversy regarding the validity of these measures partially because there is no accepted "gold standard" for determining criterion validity (Gold et al., 2002).

Agencies may have different histories of involving economists in risk assessment. For example, EPA has recognized for many years the need for early involvement of agency economists in risk assessments. EPA's primary concern has been that without this early involvement outcome measures from the risk assessment may be of a form that cannot is not useable in cost benefit analysis. In addition, there is increasing recognition that the risk generation process often involves predictable human response to environmental factors or firms' or individuals' responses to economic and social conditions. Economists are trained to model and estimate these kinds of responses. The inclusion of economists on risk assessment teams as scientific analysts, not as managers, could improve the accuracy of risk assessments by allowing risk assessment to incorporate models of production systems and human behavior that influence risk levels (Williams and Thompson 2004).

[41] MILY combines QALYs saved from avoided cases of non-fatal morbidity with life years resulting from mortality risk reductions (assigned a weight of 1.0).

6.8 How Can a Risk Assessment be Validated?

Validation of an assessment can occur at multiple levels (ECSCC 2003, WHO/FAO 2008). Validation of the conceptual and mathematical models, the computer algorithm and the assessment's predictions all occur before a risk assessment can be considered validated. Except for the final level, the validation process is often accomplished through review by persons outside the risk assessment project.

Model validation and verification in risk assessment are general terms that are sometimes used to refer to rigorous data driven evaluation of models, but more often they are used interchangeably to refer to a less rigorous "reality check" that may have poorly defined validation criteria. Risk assessors should be aware of the differences between model validation and verification and whether a model has been validated for interpolation or extrapolation. Researchers gathering data for the USDA use the more formal rigorous definitions of verification and validation as follows (Oscar, 2005): "Verification… is the successful outcome of the performance evaluation process where the model predictions were compared with the data used in model development (that is, dependent data). In contrast, validation… is the successful outcome of the performance evaluation process where model predictions were compared with data that was not used in model development (that is, independent data)."

The iterative nature of most assessments suggests that the models have been reviewed several times by risk managers and/or experts at the conceptual and mathematical development phases of the project. Nevertheless, public and peer review is usually solicited to examine the results of the assessment. Close scrutiny of the conceptual and mathematical models – and the computer algorithm – by specialists knowledgeable in statistics, epidemiology and mathematics will serve to sanction the validity of the mechanics of an assessment model.

The output of an exposure or dose-response assessment (a dose-response relationship, an exposure distribution) is often not readily measured in nature. Surveillance data may be available for some outputs of the models and statistical measures of agreement between the model's predictions and empirical observations are helpful in describing the accuracy of the model. Creative uses of empiric evidence may serve to support a contention of validity. Nevertheless, most risk assessments cannot meet this burden of proof concerning their validity. Such is the nature of many risk assessment problems; their verification primarily stems from the logic and reasoning built into the models used to solve them.

Because validation implies different criteria in different situations, any discussion of validation should refer to how the validation was performed so that readers may understand the degree of rigor the validation effort entailed. One method of validating the risk assessment findings is to compare the outputs to epidemiological data to determine whether the risk estimates are consistent with reality. The following examples illustrate MRA model validations:

a) **Rotavirus in Drinking Water:** To confirm the validity of the output results of the epidemiologically-based model used in a case study of rotavirus in drinking water (Soller et al., 1999), a dynamic model was modified using actual data and best judgment to analyze and simulate a 1981 rotavirus outbreak in the Eagle-Vail and Avon communities in Colorado (Hopkins et al., 1984). Although a rigorous direct comparison of the results from the actual outbreak and the rotavirus simulation could not be conducted due to a lack of specific surveillance data (e.g., concentration data, secondary spread), a qualitative comparison was made to assess the plausibility of the output from the model. The overall attack rate for diarrhea and/or vomiting during the rotavirus epidemic was reported to be approximately 32% (Hopkins et al., 1984). Using virus detection or serological methods, it was estimated that a total of approximately 23% of the population became ill from rotavirus exposure during this event. The results of a 5000 trial Monte Carlo simulation of the outbreak using the model showed that about half of the trials resulted in average daily disease prevalence rates ranging from 7.5% and 25%, which compares favorably to the historical estimate of 23%. Thus, it may be inferred that the output from the model seems plausible and intuitively consistent with the actual outbreak data.

b) ***Cryptosporidium* in Drinking Water:** Teunis and Havelaar (1999) conducted a case study of *Cryptosporidium* in drinking water and discussed the importance of and opportunities to attempt validation of their calculated estimates of yearly individual infection risk through comparison with actual epidemiological data on endemic/epidemic cryptosporidiosis. Their approach also provides a logical and transparent methodology to integrate quality of life-based approaches into the risk assessment by expressing all health effects in one single metric—the DALY. Such an approach has the added advantage of not being disease-specific and lends itself for risk comparisons (e.g., with chemical risks, for economic evaluations).

Whether or not formal validation is possible, peer review is an important aspect of evaluating models (OMB, 2004).

7. RISK MANAGEMENT

7.1 What is Risk Management?

Risk management is a ubiquitous term used in settings as diverse as financial investing, military planning and public health. Within federal public health agencies, risk management refers to activities ranging from high-level policy making to routine, sometimes *pro forma,* risk control applications in operational risk management. This chapter provides an overview of the risk management processes likely to be encountered by microbial risk assessors.

The NRC "Red Book" initially defined risk management in very broad terms as "the process of evaluating alternative regulatory options and selecting among them." (NRC, 1983). By 1996, a follow-on NRC committee described the activities of risk managers:

> Risk managers are supposed to deal with broad social, economic, ethical, and political issues in choosing from among a set of decision options by using the results of the risk assessment and their understanding of the other issues. Making tradeoffs, which may be called risk-benefit, cost-benefit, or risk-risk evaluations, is part of risk management. (NRC, 1996)

The NRC reports focused on management processes occurring with a single risk assessment. During the mid-1990s, the Presidential/Congressional Commission on Risk Assessment and Risk Management argued that risk management should no longer be thought of a process that focuses on decisions about managing one risk at a time. Rather, governmental agencies need to confront the task of managing risks from multiple hazards and exposures. To this end, the Commission provided a framework for risk management recommending that the risk management process include steps to (P/CC, 1997):

a) Formulate the problem in broad context

b) Analyze the risks

c) Define the options

d) Make sound decisions

e) Take actions to implement the decisions

f) Perform an evaluation of the effectiveness of the actions taken

The engineering and systems analysis view of risk management has been described by Haimes (2004): risk management is the process focused on controlling risks by addressing:

a) What *can be done* and what are the *options* for controlling risks?

b) What are the *trade-offs* in terms of risks, benefits and costs?

c) What are the *impacts of risk management decisions* on future options for risk management?

Federal state and local public health agencies are charged with the responsibility of preventing, mitigating, or controlling risks to the public's health. As a general concept, it is clear that the "mission" of risk management is accomplished using risk management processes at several levels. For example, Table 7.1 describes risk management as a strategic, applied, or operational function in the agency. The strategic level is concerned with managing the agency's portfolio of risks; the applied level—the primary focus of this Guideline—concerns the risk management processes surrounding specific risk assessments; and operational risk management deals with risk management that is guided by standard operating procedures.

Table 7.1 Classes of Risk Management in Federal Agencies

Class of Risk Management	Description
Strategic Risk Management (and Policy Making)	Long-term, broadly based view of the agency's entire risk portfolio. Interface with the public, industry and governmental stakeholders about policy issues such as the level of acceptable risk, the risk-based decision- making process
Applied Risk Management	Charters and collaborates with risk assessors on newly identified or emerging risks, new scenarios for known risks and risk mitigation scenarios. The information gained from the risk assessment is used in risk management decisions about controlling the risk
Operational Risk Management	Implements prescribed administrative, engineering or other controls to maintain risk at appropriate levels or below.

The Codex *Draft Principles of and Guidelines for the Conduct of Microbial Risk Management* includes the following 8 principles (Codex, 2007a).

a) Principle 1: Protection of human health is the primary objective in MRM.

b) Principle 2: MRM should take into account the whole food chain.

c) Principle 3: MRM should follow a structured approach

d) Principle 4: MRM process should be transparent, consistent and fully documented.

e) Principle 5: Risk managers should ensure effective consultations with relevant interested parties.

f) Principle 6: Risk managers should ensure effective interaction with risk assessors.

g) Principle 7: Risk managers should take account of risks resulting from regional differences in hazards in the food chain and regional differences in available risk management options.

h) Principle 8: MRM decisions should be subject to monitoring and review and, if necessary, revision.

7.2 When and How Can Risk Managers be Involved in Risk Assessments?

Risk management begins before risk assessment. Sometimes, the recognition of potential problem and the general hazard identification occurs externally to the agency and is brought to the attention of risk managers by the public, stakeholders, or other governmental organizations. Risk managers typically determine the need for a risk assessment and provide the risk assessment team with the specific risk analysis to be performed. This often includes setting the analytical boundaries and constraints for the risk analysis. For example, the risk assessment might be focused on risks from exposures to the hazard only within the U.S. borders or, perhaps to a particular subpopulation at risk of illness from exposures to the hazard. The initial problem formulation and discussion about boundary conditioning are often accomplished interactively with risk assessors who have the particular knowledge about what can be performed quantitatively and whether or not a quantitative risk assessment can be accomplished within the project time constraints.

Risk managers should work interactively with you (risk assessors) during the planning and scoping activities to collaborate on defining clear, scientifically defensible "risk questions" before the analytical components of risk assessment are executed (FDA, 2002). Forming a risk question is analogous to stating a testable hypothesis at the outset of a basic research project: it is a necessary antecedent to designing an objective and informative project. Here, risk managers are generally aware of the type of information needed to answer policy questions, the resources available to mount complex and large-scale risk assessments, and relevant stakeholder concerns. You will probably rely on risk managers for a "big picture" perspective of the agency's entire portfolio of risk management activities and how the current risk assessment fits into the agency's work plan.

Risk managers have valuable insights into the value and potential problems of risk assessments. Thus, high quality risk management requires risk managers to interface with you at various stages throughout the entire risk assessment process so that they can help you anticipate problems in the analyses and redirect resources, if necessary, to improve or ensure the quality of information resulting from the risk assessment. Additionally, risk managers might become aware of new information about the risk in

question that might be useful for focusing the risk assessment on a modified risk question. The frequency of risk assessor and risk manager discussions will likely depend on the complexity and nature of the risk being investigated.

The risk managers should explain clearly why the assessment is being performed and what questions need to be addressed. The risk managers should also advise the assessors, economists, engineers, and other contributing experts involved in the planning and scoping of any interested party, affected party, or policy interests to be considered in the context of the risk issue. These factors may influence the risk management options, management goals, key participants, data sources, selection of assessment endpoints, or the schedule for the development of the assessment. The risk manager and appropriate others should discuss any regulatory basis for the risk assessment and what kind of information is required to satisfy such requirements.

Risk assessment teams usually have a lead risk assessor.[42] The lead risk assessor is responsible for ensuring that risk assessments are properly performed and documented and that the key information from risk characterization is elevated up the management chain and communicated to senior management. The lead risk assessor should ensure that the risk characterization integrates other considerations specified in applicable statutes, Agency and office policies, executive orders, and other factors to make and justify regulatory decisions. The lead risk assessor's specific responsibilities might include:

a) Ensure that all risk assessment work products produced by or submitted to your organization are well written and characterized.

b) Provide advice, guidance, and support for the preparation, conduct, and completion of an appropriate risk assessment for your decision.

c) Play a major role in managing and documenting the planning and scoping process.

d) Ensure that sufficient funds are designated in the office's budget request to conduct a risk assessment.

e) Establish a realistic risk assessment schedule.

f) Ensure that the products prepared by individual risk assessors for their portion of each risk assessment document are integrated into a complete risk assessment.

g) Establish systems to maintain records of the risk assessments prepared by risk assessors under your supervision.

[42] Synonyms for this position include, technical integrator, risk assessment team leader, risk assessment team liaison, and risk assessment manager.

h) Ensure that the key points from the risk assessment are carried forward in all deliberations or considerations for decision making.

i) Review implemented decisions for the degree of implementation, efficacy, and ongoing relevance.

7.3 How are Risk Management Options a Useful Component to Include in a Risk Assessment?

"Risk managers use information from risk assessment and economic analysis, together with information about public values and statutory requirements, to make decisions about the need for and methods of risk reduction" (P/CC, 1997). To accomplish risk management decision-making, a decision among options for risk management controls requires that the decision alternatives for risk management be specified. The characterization of options often means that risk assessors are asked to calculate risks given one or more proposed risk management scenario. Risk managers use these scenarios, the inputs of benefit-cost assessments and other information to make decisions about the best option for controlling risks.

One of the principles of risk management is that the risk management analysis and the proposed risk control strategy should be commensurate with the level of risk. The reality of this principle in practice is that it also often relates to uncertainty about the magnitude of risk. Highly uncertain risk estimates sometimes lead risk managers to expend additional risk assessment resources in an effort to reduce the uncertainties before decisions about controlling the risks are taken.

From the risk manager's perspective, risk assessment is only one among several tools that can be used to inform the risk decision made by decision makers. Decision making by risk managers can call for benefits-risk assessments, risk-risk analysis, VOI analysis, or trade-off analysis as additional information useful to making decisions about managing risks. Decision analysis might be used to create a systematic and transparent decision making process that evaluates the importance of factors ranging from the objectively scientific to social values. For most scientific endeavors, "risk-informed" or "risk-based" decision making benefits from a formal decision analysis that provides a systematic analysis of complex scientific information, concerns of stakeholders, the constraints on risk management options caused by gaps in data, models or policies, and need for transparency in governmental decision making.

7.4 What are Some Other Inputs into Risk Management Decisions About Controlling or Accepting Risks?

Decision making about risks often requires balancing results of a risk assessment with the results of benefit-risk and risk-risk tradeoff analyses; the need for risk management resources to address other risks in the agency's portfolio; and political pressures from stakeholders in industry, the public, or legislatures. The most important input from risk assessment into the decision-making is a high-quality risk

characterization from which risk managers can evaluate the scientific underpinnings of risk estimates, including a characterization of uncertainties in the estimates.

Risk managers make decisions under uncertainty. The results of benefit-risk and other analyses prior to risk management decision making often include equivalently uncertain estimates for the impact of proposed risk controls on the reduction or elimination of risks. Here, risk managers can employ formal decision analysis to trade-off decision alternatives, based on both objective and subjective assessments, for a transparent decision. In other situations, the decision to control or accept risks might be well-defined by existing regulation or guidance. Ultimately, the goal of risk management is to achieve an appropriate level of risk. The affirmation of this goal might not occur until after risk management controls are applied and the risks have been evaluated iteratively.

One of the most difficult aspects of risk management for regulatory functions is setting an acceptable or tolerable level of risk. Different approaches to setting standards have different ways of incorporating tolerable risk levels. You should be aware of what approach has historical precedence in the field that applies to your risk assessment. A summary of some approaches to acceptability that have been suggested to regulators is provided below (adapted from Humber and Almeder 1986, Fischhoff et al. 1981, Lave and Romer 1981, Lowrance 1976):

a) **Reasonableness** – This is a commonly cited principle in safety judgments. For example the Consumer Product Safety Commission is mandated to "reduce unreasonable risk of injury."

b) **Custom of usage** – The U.S. Food and Drug Administration (FDA) "generally recognized as safe" (GRAS) determination is for food substances which do not have to be regulated as additives because among other things they have a history of usage. Table salt and sugar are examples.

c) **Prevailing professional practice or professional judgment** – Originally established for physician's clinical practice, the principle is also used for local building standards and toy design. The underlying assumption is that sanction by custom is safer than untested. In professional judgment professionals rely on personal experience, accepted professional practice and their clients' desires to judge risks.

d) **Best available practice, highest practicable protection, and lowest practicable exposure, best available technology** – air and water quality regulations are associated with these standards, but they still require judgment. Lowest practical is also known as "as low as reasonably practical" (Vatn 2004).

e) **Risk benefit (degree of necessity or benefit)** – A rough balancing of risks and benefits is attempted.

f) **The Delaney Clause or No-risk** – named after former New York Congressman James Delaney who added language to the Federal Food, Drug, and Cosmetic Act that states "no [food] additive shall be deemed to be safe if it is found…to induce

cancer in man or animal." It has been criticized for omitting other health endpoints and because it ignores dose-response relationships. It attempts to lower risks to zero.

g) **No observable adverse effect level (NOAEL)** – because no adverse effects are observed at a given level of chemical, that level is deemed acceptable. It is customary in chemical risk that when the NOAEL is based on animal data, a safety factor of 100 is applied to arrive at the level for humans (decimal point is moved to the left to make the appropriate level 100 times lower).

h) **Cost-effectiveness** – Equates the cost of saving lives or preventing adverse effects across programs.

i) **Formal benefit-cost analysis (also referred to as cost-benefit analysis)** – quantitative framework that includes explicit dollar value of a human life or human wellbeing. Performance of cost-benefit analysis still only serves to inform the risk management decision. It cannot determine the decision (Williams and Thompson 2004).

j) **Risk-risk** – balances various risks against each other.

k) **Quantitative risk assessment** – is able to provide a range within the actual risk that the acceptable risk can fall. Alternative management assumptions can be tested. [43]

For chemical risk assessment usually the risk level is stated quantitatively. For microbial risk level, regulators very often refer to quantified risk reduction, without actually stating the level of risk associated with the risk reductions or commenting on the acceptability of the level of risk. Some levels of risk that are customary for microbial hazards in different media include:

a) **Ambient recreational water** – The U.S. and the most stringent European Union and WHO standards are associated with about one to two percent (1-2%) increased risk of gastrointestinal illness due to exposure to ambient water during recreational activities (EPA, 2004c).

b) **Foods** – Hazard Analysis Critical Control Point (HACCP) plans are common and are not linked to acceptable health risk levels, but are designed to ensure contamination of food is prevented or otherwise mitigated. Food Safety Objectives and Performance Objectives are stated goals (often numeric in nature) for public health, processing, transportation, or retail safety (Crouch et al., 2009; Havelaar, et al., 2004; Rieu et al., 2007). Management options are often linked to quantified public health outcomes.

c) **Biosolids** – Microbial standards are currently based on operational standards which should provide pathogen levels that are below detection limits. The standards are not linked to level of health risk.

[43] Proposed control measures must really reduce risks, not transfer them somewhere else (de Koning 1987).

d) **Air** – There are no standards or Threshold Limit Values for microbial pollutants in the United States, but numeric criteria for mold and bacteria levels have been set in other countries. Indoor levels are compared to outdoor levels.

e) **Vaccinations** – Historically accepted or rejected risk levels of different adverse health outcomes are compared to benefit:risk analysis for new vaccinations (FDA, 1999).

f) **Occupational** – The General Duty Clause provides employees with workplaces that are "free from recognized hazards that are causing or are likely to cause death or serious physical harm." Target risk levels for quantified health outcomes are not discussed (29 CFR 1910.1030).[44]

7.5 What are Some Operational Risk Management Tools and Approaches?

A major reason for the development of the public health field was for the prevention of microbial diseases in human population caused by unsanitary conditions (e.g., John Snow and the 1854 *Cholera* outbreak) or the presence of vectors for disease transmission. The regulatory tools at the disposal of the public health risk manager span an entire range of options to prevent the occurrence of pathologic organisms (e.g., low-acid canning regulations) to limiting the means of primary or secondary transmission to vaccination of the host (e.g., for *Bacillus anthracis)* to improve host resistance to disease. Sanitary engineering designs that provide clean drinking water and separate waste water channels are credited with preventing countless outbreaks of disease (e.g., cholera, typhoid fever) and saving countless lives in more modern times. Even the education and outreach programs to promote safe handling of raw foods during preparation are part of risk management strategies to reduce the incidence of foodborne illness.

Generally speaking, operational risk management controls for health hazards are classified as physical, administrative and management controls (Table 7.2). For the particular case of microbial hazards, a "biological" classification for the host could be added for the possibility of immunization against some of the microbial hazards.

[44] http://www.osha.gov/pls/oshaweb/owadisp.show_document?p_table=STANDARDS&p_id=10051

Table 7.2 Class of Risk Management Controls for Operational-Level Microbial Risk

Class of Operational Risk Controls	Examples
Physical	• Barriers and physical factors that eliminate or reduce the chance of contamination and growth of pathogens • Pasteurization with heat or ionizing radiation • Packaging • Disposable gloves
Administrative	• Engineering designs: process controls • Work practices and standards • Sanitary practices by personnel • Training program design
Managerial	• Training execution • Supervision • Appropriate skills for the tasks that might lead to contamination or favorable growth
Biological (specific microbes)	• Vaccination • Food animal treatments with benign organisms

7.6 What is Risk Management for the Intentional Use of Regulated Microorganisms?

Microorganisms, whose intentional uses fall under federal laws, including, but not limited to, the Federal Insecticide, Fungicide, and Rodenticide Act, the Federal Food, Drug, and Cosmetic Act, the Plant Protection Act, the Endangered Species Act, and the Invasive Species Act, often have specific risk management options available to the regulators. These options can involve placing restrictions on their use or, if justified, denial of permission for their use. Restrictions on their use can vary according to the specific law and regulations involved. For example, for pesticides, the restrictions can be used to limit exposure by specifying on the approved labels exactly when, where, and how much can be applied. Furthermore, protective clothing, including respiratory masks, may be required to be used. Restrictions can also be set on what levels are acceptable to appear on food crops, although due to the potential for growth subsequent to application, microorganisms with any potential for human toxicity and/or pathogenicity are generally not allowed to be used on food crops. Containment requirements can be placed on intentional uses of regulated microorganisms for field testing and for industrial uses. Genetic engineering can be used to reduce risk concerns for intentionally used and regulated microorganisms, e.g. by deleting toxin genes. In addition, marker genes can be used to better identify the specific approved uses. Restrictions on the storage and movement of the regulated microorganisms can also be used to mitigate risk.

8. RISK COMMUNICATION

A risk assessment is only as good as your communication of its output. All of the time and effort you spend conducting a risk assessment is for naught unless there is clear communication of risk assessment results to risk managers and other stakeholders. Effective risk communication requires diligence and has the same level of importance as any other component of the assessment.

This Guideline does not provide a detailed treatment or guidance for risk communication; you can refer to other places for such detail (Sellnow, 2008; Morgan, 2002; Lundgren and McMakin, 1998). Rather, this chapter provides an initial understanding of how risk communication plays an important role in the risk assessment process.

Risk assessors are not expected to carry the load of risk communication. You need to work with the appropriate communication offices in your Agency (e.g., public affairs office, Congressional outreach office). These communicators need to be part of the team during planning and scoping and throughout the remainder of the risk assessment process.

8.1 What is Risk Communication?

Risk communication describes and exchanges information about risk, including its form and severity, and what can be done to lessen or avoid it. It includes two equally important objectives:

a) Inform risk managers about risk so that they may make informed decisions.

b) Inform the public about risk so that they understand the nature of the risk and what is being done or will be done about it.

Risk communication is the interactive exchange of information and opinions concerning risks and risk management among risk assessors, risk managers, consumers, and other interested parties (WHO, 2000).

8.2 What are the Aspects of Risk Communication?

Risk communication principles include (OMB 2007b):

a) Risk communication should involve the open, two-way exchange of information between professionals, including both policy makers and “experts” in relevant disciplines, and the public.

b) Risk management goals should be stated clearly, and risk assessments and risk management decisions should be communicated accurately and objectively in a meaningful manner.

To maximize public understanding and participation in risk-related decisions, agencies should:

1) explain the basis for significant assumptions, data, models, and inferences used or relied upon in the assessment or decision;
2) describe the sources, extent and magnitude of significant uncertainties associated with the assessment or decision;
3) make appropriate risk comparisons, taking into account, for example, public attitudes with respect to voluntary versus involuntary risk; and,
4) provide timely, public access to relevant supporting documents and a reasonable opportunity for public comment.

The aspects of a good risk communication plan (see Section 2.6.6) include:

a) Involvement and input of risk managers and stakeholders throughout the risk assessment process
b) Clear risk management questions that are understood by managers and other assessors
c) Awareness by managers and other stakeholders of the strengths and limitations of the assessment

Social and personal behaviors are strongly influenced by risk managers' pronouncements, but only if the underlying risk assessment process is transparent to the public and considered credible. Trust is based on open communication and the credibility of provided information; thus, the public must be aware of the science behind the risk assessment. People who write risk communication statements should consider acknowledging both the power and the limits of the risk assessment process and the data used in the risk assessment.

8.3 Who are the Stakeholders of MRAs?

Stakeholders are people or organizations that may be affected by the relevant decision and thus, have an interest in the outcome of the risk assessment. They typically include people such as those in industry who will be responsible for implementing and financially affected by new rules and regulations borne of the risk assessment, as well as the general public. For example, a risk assessment that results in new regulations for *Escherichia coli* in ground beef would affect all persons who produce and consume ground beef. Anyone interested in a risk assessment may be reasonably termed a stakeholder. Stakeholder groups will self-identify but should also be sought out.

Risk managers, assessors, and communicators are not typically considered stakeholders because they should remain unbiased. If the results of the risk assessment could affect them and the outcome was altered because of this knowledge, this would be considered a conflict of interest.

8.4 With Whom Can I Communicate?

Even if risk communication specialists are on the team, you as the risk assessor may serve in a risk communicator role and be responsible for communicating with stakeholders and developing outreach materials. If this is the case, you can work closely with appropriate Agency offices to follow protocol. For example, many agencies will request that communication with members of Congress be done through a specific office. Communication strategies and materials for different stakeholders should be tailored to that particular audience. For example, communication with technical experts could have a high level of technical detail, while communication to the lay public would be less technical.

The language used in communication materials should conform to your Agency's standards, but needs to be absolutely comprehensible by almost all being addressed. Agency communications, public affairs, and/or outreach offices should be consulted to assure that the message can be understood. Your agency may also have specific guidance for coordinating with other federal, state, and local health and environmental agencies.

8.5 When Can the Process of Risk Communication Begin?

There is a tendency to view risk communication as a final stage to the risk analysis process—something that occurs upon *completion* of the risk assessment and risk management. It is critically important that risk communication strategies be developed at the *beginning* of a risk assessment (during planning and scoping), to inform risk managers and other stakeholders *throughout* the risk assessment. The best place to start the risk communication process is during planning and scoping discussions in which a risk communication specialist can draft a risk communication plan (i.e., the communication strategy, including the risk assessor's role).

During development of the risk assessment, the team leader can be proactive in implementing the risk communication plan, particularly communicating with risk managers and stakeholders. For example, one way to do this is by announcing that the Agency plans to conduct a risk assessment. An announcement can be placed in the Federal Register. Other good venues for announcing risk assessments (and subsequent activities) include the agency's web site, advertising/announcement sections of trade and professional journals, and at professional meetings.

8.6 Can I Communicate in Writing, Orally, or Both?

Both. Oral communication is needed at many points throughout development of the risk assessment. It is appropriate to keep risk managers and other stakeholders informed about progress of the risk assessment. For example, you may wish to schedule a weekly, (or other appropriate frequency) phone call with managers to keep them updated. Similarly, periodic conference calls or public meetings with stakeholders lessen the chance that they are caught off guard when the risk assessment is completed.

Communication in writing is almost always appropriate, especially since a written record is usually needed. For example, it is good to have the risk assessment questions in writing. Doing so entails back-and-forth work with risk managers to identify and clearly articulate the purposes of the risk assessment. By solidifying the questions in writing, you help ensure that risk managers, risk assessors, and stakeholders clearly understand what the risk assessment is intended to do.

The most current risk communication message should be available in some written form (e.g., fact sheets), as well as electronically, throughout the process. Information should be available on your Agency's website (although it should also be accessible to all who do not have computer access). Coordination with local health authorities in most cases is also important, and they may be able to suggest useful approaches to communicating with their public.

Virtually all completed risk assessments include a written report. The TCCR principles discussed and integrated throughout this Guideline will help with the effective written communication in the form of the report (Section 1.10). In addition, many risk assessments are presented publicly as presentations or seminars.

8.7 Who Decides What to Communicate?

The risk manager—in consultation with the risk assessor—is responsible for deciding what information to communicate. Any formal communication or information release should be firmly based on the documented findings of the risk assessor. Therefore, you are responsible for communicating risk to the risk manager for a full understanding of the potential risks that would then be considered in the decision making process. Again, risk communicators can assist in the delivery and presentation of the information to the public.

MRA is an inexact science, requiring judgment calls and policy decisions on the part of highly trained, experienced professionals. This message should be included in any communication of risk, following a statement of known facts and preceding prescriptive risk reduction measures. Mathematical constructs and underlying assumptions should be made clear. Although there may be differences of opinion among risk assessors, the risk manager is ultimately responsible for deciding on a transparent and clear message on which the audience can evaluate agency actions.

8.8 What Information Can be Communicated?

The content of the formal risk assessment (as communicated to the public or to stakeholders) is determined by the risk manager, but you are responsible for presenting all available data, including those that challenge or do not support points in the assessment. The information that the risk manager may decide to communicate includes:

a) Data on human disease identified as either historical or experimentally-derived, or projections generated by modeling

b) Underlying uncertainties or data gaps

c) The degree of potential hazard to sensitive populations such as children, the elderly, and immune compromised people (focusing on susceptibility and severity)

d) Explanation that the different pathways that were deemed relevant were explored thoroughly

e) The possibility of person-to-person transmission (if applicable)

f) Any results from animal testing and how these data may be relevant to humans

g) The potential for zoonotic transmission between humans and animals (if applicable)

h) Potential actions to reduce exposures (for example, following posted signs regarding swimming, fishing, or harvesting clams)

You will have to provide the risk manager with your best professional judgment at that time as to the degree of risk associated with a specific hazard. This judgment can be accompanied by all relevant data about both the hazard and the target population. Probably some of the most important pieces of information you need to communicate are descriptions of the probabilities, uncertainties, and possible sources of biases or error (assumptions, generalizations) in any interim or final assessment.

8.9 How is the Communication Process a Continuous Dialog?

Effective risk communication is a continuous process that requires constant feedback throughout the risk assessment. This begins at the planning and scoping stage, when all parties involved (including stakeholders as appropriate) should have a chance to comment on the need for a risk assessment, the scope of the risk assessment, and the other factors discussed in planning and scoping. Doing this helps to promote buy-in and helps to ensure respect for the process. Communication of risk from assessors to managers is constant throughout the process. Communication of risk from risk managers to the public is usually episodic, but nonetheless, scheduled regularly.

As an example, interactive communication occurs between risk managers and risk assessors in developing risk management questions. A risk manager may say s/he wants a risk assessment to address "Salmonella." It is then up to you to press for more specifics. At this point, perhaps the manager refines the question to "What is the effect of increased cooking temperatures on illnesses from Salmonella?" Then you may indicate that data are only available for a specific serotype, Enteritidis for example, but not all serotypes. Thus, the manager can work to refine the question to include Enteritidis only. In the end, iterative communication helps ensure clear and concise questions, which in turn increases the likelihood of a useful risk assessment. This iterative dialog can be conducted in person to speed up the process, but written documentation of the

understandings between risk managers and risk assessors is important for clarity and the orientation of new team members.

Communication can also be iterative when describing results of the assessment to managers and other stakeholders. In virtually all cases, there is considerable room for improvement in risk assessments. Therefore, it is unwise to present results from risk assessments as if they were written in stone. Instead, the results should be presented as a best effort, with the idea that feedback from managers and stakeholders will likely improve the assessment.

8.10 How In-Depth Can I Communicate?

Depth of communication depends on the audience. For example, if you are describing the risk assessment to another risk assessor, perhaps one who will peer review the assessment, then your communication should be very detailed. If, on the other hand, you are presenting the results to a high-level risk manager in the space of ten minutes, in-depth details should be avoided unless specifically sought. When communicating with stakeholders, it is important to present results in a clear manner without talking down to the audience. For example, do not say, "This next part is complicated, so I'll put it in terms you can understand." Instead, start with a broad description of the work and then proceed to the details as both time and the audience's needs dictate. Lastly, it is very important to write a detailed report of the assessment. That way, even though you may be unable to give specifics during a talk, briefing, or other venue, you will always be able to refer your audience to the written report. The detailed report should be sufficiently detailed that another team of risk assessors with appropriate expertise can replicate the risk assessment. In addition to a formal detailed report, summary graphics, executive summary, fact sheets, and other types of overview documents can aid in communication efforts.

8.11 What Can I Do if the Message Is Not "Getting Through?"

It is incumbent upon you as a risk assessor to convey the results of the assessment. If you cannot do this, then the utility of the assessment is lessened. Accordingly, when you communicate the results of the assessment (or any aspect of the assessment for that matter), work to engage your audience. Take time to explore if your audience understands the points you are trying to communicate. If not, take a step back and work to clarify the parts of your message that are confusing. Look at this as an opportunity to exchange information and thoughts with your audience. Keep in mind that the difficulty may lie in your communication, not in your audience's comprehension. It may help if you communicate through various means. For example, in addition to speaking, it may help to take out pencil and pad and sketch your message. Be sure to allow time throughout your communication (be it a formal slide show or an informal conversation) for the audience to seek clarification. When experiencing problems getting through to the audience, consult communication specialists within your Agency that can assist with communication.

8.12 How Can I Communicate Risk Successfully?

Successful risk communication requires strategic planning. This planning requires in turn a very thorough review of the costs and benefits of specific actions (or inaction), and considers possible outcomes. Successful risk communications consider the public to be stakeholders, inasmuch as Agency decisions and actions affect them directly. The stakeholders base their own behaviors on information (however anecdotal) provided by trusted sources. Strategic risk communication practices may help to develop audience understanding and ultimately gain stakeholder and public cooperation.

Strategic risk communication involves planning how to address stakeholder questions identified during planning and scoping and later during the assessment process. The responses to these questions should be straightforward and couched in simple language (rather than technical jargon). Concepts need to be packaged correctly, i.e., clearly, truthfully, and respectfully. This last requirement—that relevant concepts be presented in understandable ways that enable discussion amongst all stakeholders, rather than as abstruse knowledge suitable for expert analysis only—is critical in establishing successful communication.

Risk managers turn to strategic risk communication when they need "buy-in." The best way to achieve cooperation of public and stakeholders in, rather than agitation against, risk-management decisions is by helping the audiences choose among options that involve cost (risks) and safety (benefits). Usually no one on the team can foresee the full range of audience responses. The starting relationship between Agency and audience may be skeptical and/or confrontational, pitting audience experience against expert analysis. Strategic risk communication helps the audience to gain insight into the problem (and/or proposed actions) and to establish exactly what aspects of the proposed action (timing? approach?) are within their control. Ideally, successful risk communication will form the basis for mutual trust; but at the very least, strategic risk communication transfers information.

People respond not only to what is said, but how it is said. In addition, people response to the way in which actions are carried out. Stakeholders value the qualities of listening, understanding, and responsiveness on the part of the Agency. Other important factures include their own perception of risk acceptability, due process (in which stakeholders are able to participate in judging risks and predicted benefits), their personal sense of the risk-manager's credibility, and, above all, open communication (transparency). Successful risk communication is developed with full appreciation of the technical complexity of the situation in question, the controversy about or unavailability of the requisite science, the sensitivity of the communication environment, the potential relevance of political realities, and the perceived credibility of parties involved.

Other preparations can also contribute to successful communication. For example, practicing presentations before audiences such as co-workers can test the understandability of the presentation and increase the presenter's comfort level. The length of presentations is also an important consideration, since typically audiences do

not want long presentations without opportunities for to ask questions. Furthermore, practical information such as what actions can be taken by individuals or communities to interrupt exposures (e.g., beach closures to limit exposure to fecal contamination) can be included whenever the risk assessment is presented to the public, even if the actions suggested are not directly addressed or considered in the risk assessment.

8.13 How Can I Handle Media and Congressional Office Requests?

Risk assessors are sometimes approached directly with questions by external news sources and congressional offices. It is the responsibility of the risk manager and/or your communications offices (e.g., public affairs office, congressional liaison office), not the risk assessor, to communicate risk. If you are contacted directly from someone in the media or a congressional office, inform that person that your communications office is authorized to answer their questions (e.g., requests for interviews, background information, policy questions). Provide them with your communications office contact information (phone number, e-mail address). If you are contacted directly via e-mail, simply forward the message to your supervisor and communications office. No matter how you are contacted, inform your supervisor and include as many details about the request as you can.

However, there will be occasions that, with appropriate permissions, you can provide technical information relevant to the risk assessment being queried. Remember that there is no such thing as "off the record." Assume everything said is on the record; even "background" information is still a response. Once you provide information, nothing really prevents the recipient from including that information in their article or report. A good reporter for example may not use "off the record" information as directly attributable to you, but will have a lead to contact others to verify it and then may use or release that information. You might also consider having a press officer on the phone or during interviews to make sure that the appropriate information is communicated. You may need to be aware of press deadlines and might be asked to contribute graphics to include in articles where appropriate.

8.14 When Can Risk Communication End?

Once a decision has been made, fully implemented, and openly communicated as per the risk communication plan, subsequent communication efforts may not be as intensive. However, there will likely be a need to monitor how the implemented decision is accomplishing its goal(s) or not, and communication will be critical at that time. You need to be aware that resultant actions based on the risk assessment may need to be re-evaluated and addressed further in the future (and this can be part of your communication plan). However, be careful not to mislead the public into expectations of involvement and re-evaluation that might not be appropriate.

Risk communication shouldn't have an absolute end; it can be an ongoing process. In another vein, risk managers can take the initiative to incorporate risk communication into routine functions. For example, the various microbial societies

(most notably, in the United States, the ASM) produce informational outreach material targeting specific age groups and educational levels. These professional societies seek to expand public understanding of the role of microorganisms in human affairs beyond disease causation through staff dedicated to the effort. Agency public affairs offices might fulfill their responsibility to the public by teaming with these societies and by working with internal environmental-education staff to develop guidelines and procedures for ongoing risk communication. At least one senior public affairs manager could be tasked to work with Agency experts to identify and communicate risk specific to children, seniors, and other sensitive populations. Interaction with public health and safety agencies (the Centers for Disease Control and Prevention [CDC] or the Occupational Safety and Health Administration [OSHA]) would be advisable, as would consultation with academic and industrial clinicians. A database of these and other external consultants can be developed in anticipation of potential outbreaks.

The risk communication network just described would be expected to develop outreach materials continuously, and to establish name recognition for the responsible Agency via routine (e.g., bimonthly) public education broadcasts or activities. Such learning opportunities could be easily incorporated into local public school curricula, and ideally would be organized and distributed by agency offices nationwide. The network, once in place, can also provide informational materials to concerned individuals (and Internet blogs) or to news outlets.

9. GLOSSARY

The term definitions in this glossary are from the EPA Thesaurus of Terms Used in MRA unless otherwise noted (EPA, 2007b). For the original sources of the definition, see the Thesaurus.

acceptable risk
This is a risk management term. The acceptability of the risk depends on scientific data, social, economic, and political factors, and on the perceived benefits arising from exposure to an agent. Tolerable risk is a synonym.

analysis of variance
This is a statistical technique that isolates and assesses the contribution of categorical factors to variation in the mean of a continuous outcome variable. The data are divided into categories based on their values for each of the independent variables, and the differences between the mean outcome values of these categories are tested for statistical significance.

analysis plan
This is a plan that provides all the details of exactly how each part of the risk assessment will be performed. It usually describes in detail what analyses will be performed, how they will be performed, who will perform the work, schedules, resources, quality assurance/quality control requirements, and documentation requirements.

appropriate level of protection (ALOP)
Codex defines ALOP as the level of protection deemed appropriate by the member (country) establishing a sanitary or phytosanitary measure to protect human, animal, or plant life or health within its territory. The term is also used more broadly to refer to risk levels selected for regulations, rules, and risk assessments.

CODEX
The Codex Alimentarius Commission was created in 1963 by FAO and WHO to develop food standards, guidelines and related texts such as codes of practice under the Joint FAO/WHO Food Standards Programme. The main purposes of this Programme are protecting health of the consumers and ensuring fair trade practices in the food trade, and promoting coordination of all food standards work undertaken by international governmental and non-governmental organizations.[45]

conceptual model
Ecological risk assessment defines a conceptual model as a written description and/or a visual representation of actual or predicted relationships between humans or ecological entities and the chemicals or other stressors to which they may be exposed. ILSI (2000) states that a conceptual model depicts the purpose, defines the scope and scale,

[45] http://www.codexalimentarius.net/web/index_en.jsp

determines appropriate variables and identifies data needed for risk assessment. It can also serve as a preliminary or exploratory risk assessment.

cost-benefit analysis and cost-effectiveness analysis (CEA)
See OMB (2003) for full descriptions of cost-benefit analysis and CEA.

data objectivity
Data objectivity "focuses on whether the disseminated information is being presented in an accurate, clear, complete, and unbiased manner, and as a matter of substance, is accurate, reliable, and unbiased."(OMB, 2002)[46]

dose-response
This is a relationship in which a change in amount, intensity, or duration of exposure to a pathogen is associated with a change in the manifestation and magnitude of human health effects.

dose-response assessment
This is the determination of the relationship between the magnitude of exposure (dose) to a chemical, biological or physical agent, and the severity and/or frequency of associated adverse health effects (response).

dose-response curve
This is a graphical representation of the quantitative relationship between administered, applied, or internal dose of a chemical or agent, and a specific biological response to that chemical or agent.

dynamic model
This considers the individual within a community rather than the isolated individual. Time-dependent elements such as secondary transmission, host immunity, and animal reservoirs are included.

endpoint
For chemical risk assessment an endpoint is an observable or measurable biological event or chemical concentration (e.g., metabolite concentration in a target tissue) used as an index of an effect of a chemical exposure. For ecological assessment an endpoint is an explicit expression of the environmental value that is to be protected, operationally defined by an ecological entity and its attributes. For example, salmon are valued ecological entities; reproduction and age class structure are some of their important attributes. Together "salmon reproduction and age class structure" form an assessment endpoint. For MRA an endpoint is usually a health effect or infected state. However indicators or conditions associated with human health could also be endpoints.

[46] http://www.whitehouse.gov/omb/fedreg_reproducible/

epidemiology triad
This is the traditional model of infectious disease causation. It includes three components: an external agent, a susceptible host, and an environment that brings the host and agent together, so that disease occurs.

exposure
Exposure is contact made between a chemical, physical, or biological agent and the outer boundary of an organism. Exposure comprises the sources, mode, route, and extent of contact with the microbial hazard(s) of concern. How *often* a person is exposed is referred to as frequency of exposure. How *long* a person is exposed to a microbial hazard is referred to as the duration of exposure.

exposure assessment
This is the process of estimating or measuring the magnitude, frequency, and duration of exposure to a microbial hazard(s), along with the number and characteristics of the person or population exposed. The route of exposure is also considered.

exposure pathway
The exposure pathway is the physical and temporal movement of microorganisms from their *source* to the occurrence of an exposure. For chemicals the exposure pathway is the route a substance takes from its source (where it began) to its end point (where it ends), and how people can come into contact with (or get exposed to) it. An exposure pathway has five parts: a source of contamination (such as an abandoned business); an environmental media and transport mechanism (such as movement through groundwater); a point of exposure (such as a private well); a route of exposure (eating, drinking, breathing, or touching), and a receptor population (people potentially or actually exposed). When all five parts are present, the exposure pathway is characterized as "complete", that is, capable of contributing to human health risks.

frank pathogen
A microorganism capable of producing disease in both healthy and compromised persons.

hazard
The term hazard can be interpreted in a number of ways. It may be defined as the stressor agent capable of causing an adverse effect on the recipient or the adverse effect itself. The selection of the definition is a policy decision driven by the existing statutes, regulations, or consistency with in-house processes. Codex considers a microbiological hazard is a hazard arising from bacteria, viruses, yeasts, molds and algae, parasitic protozoa and helminthes, and their toxins or metabolites.

hazard identification
This is the process of determining if data support the case for a chemical or a microbe causing adverse health effects in humans and what those effects might be.

health effect
This is the clinical manifestation of disease associated with a specific pathogen, including symptomatic and asymptomatic infections, clinical illness, mortality, and sequelae.

health endpoint
This is an observable or measurable biological event used as an index to determine when a deviation in the normal function of the human body occurs.

host
This is a person or other living animal, including birds and arthropods, that affords subsistence or lodgment to an infectious agent under natural conditions. In an epidemiologic context, the host may be a population or a group.

host specificity
This is the characteristic of a pathogen that renders it capable of infecting one or more specific hosts.

immunocompromised
Immunocompromised individuals have a weakened immune system, making them more susceptible to infections than the general population.

incubation period
This is the time from the moment of inoculation (exposure) to the development of the clinical manifestations of a particular infectious disease.

indicator
An indicator is any biological entity or processes, or community whose characteristics show the presence of specific environmental conditions.

infectious dose
This is the number of organisms that make individuals ill or carriers. It should be noted that methods to count microbes may not be counting individual microorganisms. For example a colony forming unit (cfu) may be a clump of cells that formed one colony on a plate. An infectious dose is the minimum number of organisms that will result in entry through the host barriers, survival of the pathogen, and multiplication in the host. Infection may or may not result in symptomatic illness. On a population basis, there is no discernible minimum infectious dose for pathogens (FAO/WHO 2003). Instead there is a probability distribution for infection associated with different dose levels reflecting intra- and inter-individual variability in the pathogen-host relationship.

infectivity
Infectivity describes the ability of a pathogen to enter, survive and multiply (infect) a host.

microorganism
These are viruses, bacteria, yeasts and simple fungi, single-celled algae, protozoa, all are defined as being organisms that can only be seen with the aid of a microscope. Most are beneficial but some produce disease (pathogens). Non-pathogenic microorganisms are critical for recycling energy and nutrients globally, such as in soil, the oceans, composting and sewage secondary treatment.

pathogen
These are microorganisms (e.g., viruses, bacteria, protozoa and the ova of helminth parasites) that can cause disease in humans, animals and plants.

pathogenicity
Pathogenicity refers to the ability of an organism to cause disease (i.e., harm the host). This ability represents a genetic component of the pathogen and the overt damage done to the host is a property of the host-pathogen interactions. Commensals and opportunistic pathogens lack this inherent ability to cause disease. However, disease is not an inevitable outcome of the host-pathogen interaction and, furthermore, pathogens can express a wide range of virulence. Virulence, a term often used interchangeably with pathogenicity, refers to the degree of pathology caused by the organism. The extent of the virulence is usually correlated with the ability of the pathogen to multiply within the host and may be affected by other factors (.i.e, conditional). In summary, an organism (species or strain) is defined as being pathogenic (or not), and depending upon conditions, may exhibit different levels of virulence.

Pathogenicity is the quality or state of being pathogenic, the potential ability to produce disease. Virulence is the disease producing power of an organism, the degree of pathogenicity within a group or species.[47]

planning and scoping
This is the process that defines the purpose and scope of a risk assessment and focuses the issues and approach(es) involved in performing the assessment.

problem formulation
In ecological risk assessment, problem formulation is the initial stage of a risk assessment where the purpose of the assessment is articulated, assessment endpoints and a conceptual model are developed, and a plan for analyzing and characterizing risk is determined.
In microbial assessment, problem formulation is a systematic planning step that identifies the goals, breadth, and focus of the MRA, the regulatory and policy context of the assessment, and the major factors that will need to be addressed for the assessment.

qualitative risk assessment
Qualitative risk assessment uses verbal descriptors of risk and severity as well as uncertainty, and often involves the aggregation of expert opinions. The results are often

[47] For further discussion of these terms visit http://scienceblogs.com/effectmeasure/2006/06/pathogenicity_virulence_transm.php

stated in an estimated range, such as "there is a moderate to high risk of a certain outcome occurring."

quality-adjusted life year
This is a unit of health care outcomes that adjusts gains (or losses) in years of life subsequent to a health care intervention by the quality of life during those years. QALYs can provide a common unit for comparing cost-utility across different interventions and health problems. Other units for measuring health outcomes include DALYs and healthy-years equivalents (HYEs).

quantitative risk assessment
In quantitative assessments, the risk is expressed as a mathematical statement of the chance of illness or death after exposure to a specific hazard, and it represents the cumulative probabilities of certain events happening and the uncertainty associated with those events.

risk analysis
A process consisting of three components: risk assessment, risk management and risk communication.

risk assessment
In the context of human health, risk assessment is a systematic way to prepare and organize information and help establish programs, R&D, and regulatory priorities; the qualitative or quantitative characterization of the potential health effects of particular substances on individuals or populations; a scientifically based process consisting of the following steps: (i) hazard identification, (ii) hazard characterization, (iii) exposure assessment, and (4) risk characterization; the formal, scientifically based process to estimate the likelihood (probability) of exposure to a hazard and the resulting public health impact from this exposure. The product of the risk assessment is often a statement regarding the probability that populations or individuals so exposed will be harmed and to what degree (risk characterization).

risk characterization
In risk characterization, risk due to a particular exposure for a defined population is described in coherent, understandable, and informative conclusions about the microbiological risk to exposed humans in a way that is useful for decision makers as well as stakeholders in that risk. In all cases, major issues and uncertainty and variability associated with determining the nature and extent of the risk should be identified and discussed. The risk characterization should be prepared in a manner that is clear, transparent, reasonable, and consistent.

risk communication
This is the exchange of information about health or environmental risks among risk assessors and managers, the general public, news media, and other stakeholders. WHO considers risk communication to be the interactive exchange of information and opinions

concerning risks and risk management among risk assessors, risk managers, consumers, and other interested parties (WHO, 2000).

risk management

In the context of human health, risk management is a decision making process that accounts for political, social, economic and engineering implications together with risk-related information in order to develop, analyze and compare management options and select the appropriate managerial response to a potential chronic health hazard.

risk profile

Developing a risk profile involves an initial systematic collection of information, which is evaluated to determine what other actions (including a MRA) and resources may be needed. The risk profile is an overall summary of the context in which a risk is being analyzed, including: a description of the risk(s) considered, values threatened by the risk, social perception of the risk, who benefits from producing the risk, who benefits from managing the risk, and characteristics of the risk, the risk-producer and the risk-bearer, which are pertinent to successful management of the risk.

secondary transmission

This is the direct or indirect propagation of a pathogen from an infected person (with or without clinical illness) to additional people.

sensitivity analysis

Sensitivity analysis examines the relative influence and importance of a model's inputs on its output measuring the 'relative' influence. It is the process of changing one variable while leaving the others constant to determine its effect on the output. This procedure fixes each uncertain quantity at its credible lower and upper bounds (holding all others at their nominal values, such as medians) and computes the results of each combination of values. The results help to identify the variables that have the greatest effect on exposure estimates and help focus further information gathering efforts.

sequelae

These are abnormal conditions that arise following the acute phase of a disease. For example, kidney failure may follow acute *E. coli* O157:H7 infection.

stakeholder

A stakeholder is any organization, governmental entity, or individual that will be responsible for implementing, or financially affected by, new rules and regulations borne of the risk assessment, or may be impacted by a given decision based on the risk assessment.

subpopulation

This is a subset of the target population that has been identified for a specific purpose, usually requires the ability to estimate an attribute of the subpopulation.

susceptible, sensitive, vulnerable

These terms refer to individuals or populations that for varying reasons suffer more severe consequences than the general population as a result of exposure to a hazard. Although these terms are used interchangeably by many risk assessors and public health experts, the Interagency Risk Assessment Consortium Susceptible Populations Workshop considered the below definitions, but emphasized that when these terms are used, they should be defined.[48]

Susceptibility is: A capacity leading to higher risk at a given exposure level, due to biological (intrinsic) factors that can modify the effect of a specific exposure

Sensitivity is: A capacity for higher risk due to the combined effect of susceptibility (biological factors) and differences in exposure

Susceptibility - Includes intrinsic factors only; Characteristic of an individual; Defined by the host

Vulnerability - Includes intrinsic and extrinsic factors; Characteristic of an individual or a group; Defined by the host (behavior) and environment[49]

transparency

This is conducting a risk assessment in such a manner that all of the scientific analyses, uncertainties, assumptions, and science policies which underlie the decisions made throughout the risk assessment are clearly stated (i.e., made readily apparent). For risk assessment to be transparent, methods, and assumptions should be clearly stated and understandable to the intended audience, so that the audience is able to evaluate the adequacy of the data and methods.

uncertainty analysis

This is used to estimate the uncertainty associated with model inputs, assumptions, and structure/form and the process of interpreting the influence of uncertainty on the results of a risk assessment.

uncertainty factor

These are usually applied to accommodate for a lack of knowledge associated with inter-species extrapolation, high to low dose extrapolation (i.e., effect to no-effect), population variation (i.e., protection of sensitive subpopulations), and extrapolation across exposure durations (e.g., subchronic to chronic.) Although uncertainty factors are commonly applied in chemical risk assessment, much less information is available supporting the application of uncertainty factors to microbiological risk assessment.

uncertainty

Uncertainty is imperfect knowledge of the microbiological hazard (e.g., its virulence), environmental pathway/processes, or the human populations under consideration (from MRA)

[48] http://foodrisk.org/IRAC/events/2010-01-10/downloads/Concept_of_Susceptibility-R_Parkin.pdf

[49] In this document vulnerability is also used in the context of a "vulnerability assessment," which is not related to the definitions discussed in this set of terms. Refer to Section 2.5.3 for the definition of vulnerability with respect to the CARVER method.

Uncertainty represents a lack of knowledge about factors affecting risk assessments and can lead to inaccurate or biased estimates of risk and hazard. Some of the types of uncertainty include scenario uncertainty, parameter uncertainty, and model uncertainty.

variability

This refers to the observed differences attributable to true heterogeneity or diversity in a parameter. Examples include human physiological variation (e.g., natural variation in body weight, height, breathing rate, drinking water intake rate), weather variability, variation in soil types and differences in contaminant concentrations in the environment. Variability is usually not reducible by further measurement of study, but it can be better characterized.

virulence

This is the degree of intensity of the disease produced by a microorganism as indicated by its ability to invade the tissues of a host and the ensuing severity of illness. (see pathogenicity for comparison)

10. ABBREVIATIONS

ACSSuT	Ampicillin, chloramphenicol, streptomycin, sulfonamides, and tetracycline
AFLP	Amplified fragment length polymorphism
AIPH	Army Institute of Public Health
ALOP	Appropriate level of protection
AOAC	Association of Analytical Communities
APHIS	Animal and Plant Health Inspection Service (U.S. Department of Agriculture)
ASM	American Society for Microbiology
Bcc	*Burkholderia cepacia complex*
BenMAP	Environmental Benefits Mapping and Analysis program
BSE	Bovine Spongiform Encephalopathy
°C	degrees Celsius
CART	Classification and regression tree
CARVER	Criticality, Accessibility, Recuperability, Vulnerability, Effect, and Recognizability
CDC	Centers for Disease Control
CDER	Center for Drug Evaluation and Research (Food and Drug Administration)
CEA	cost-effectiveness analyses
CFSAN	Center for Food Safety and Applied Nutrition
CFR	Code of Federal Regulations
cfu	colony forming unit
DALYs	Disability-adjusted life years
DHS	Department of Homeland Security
DNA	Deoxyribonucleic Acid
DOD	Department of Defense
DT104	Definitive type 104 (p48)
ECSSC	European Commission Scientific Steering Committee
EPA	Environmental Protection Agency
°F	degrees Fahrenheit
FAO	Food and Agriculture Organization
FDA	Food and Drug Administration
FIRRM	Foodborne Illness Risk Ranking Model
FSIS	Food Safety and Inspection Service
GAO	U.S. Government Accountability Office
GRAS	generally recognized as safe
gyrB	DNA gyrase, subunit B
HACCP	Hazard Analysis Critical Control Point
HC	Hazard Characterization
HI	Hazard Identification
HIV	Human immunodeficiency virus
HYEs	Healthy-years equivalents
ID_{50}	Median Infectious dose
ILSI	International Life Science Institute
LD_{50}	Median lethal dose

LT2ESWTR	Long Term 2 Enhanced Surface Water Treatment Rule
MAC	Mycobacterium Avium-Complex
MCMC	Markov Chain Monte Carlo Simulation
MILYs	Morbidity Inclusive Life Years
MLGT	Multilocus genotype sequencing
MLST	Multilocus sequence typing
MRA	microbial risk assessment
MRM	microbial risk management
mRNA	messenger ribonucleic acid
NASA	National Aeronautics and Space Administration
NCEZID	National Center for Emerging and Zoonotic Infectious Diseases
NCRP	National Committee on Radiation Programs
NIOSH	National Institute for Occupational Safety and Health (Centers for Disease Control)
NN	Neural Networks
NOAEL	No observable adverse effect level
NRC	National research council
OAQPS	Office of Air Quality Planning and Standards
OECD	Organization for Economic Cooperation and Development
OMB	Office of Management and Budget
OPP	Office of Pesticide Programs (US Environmental Protection Agency)
OPPT	Office of Pollution Prevention and Toxics (US Environmental Protection Agency)
ORD	Office of Research and Development (US Environmental Protection Agency)
OSA	Office of the Senior Advisor (US Environmental Protection Agency)
OW	Office of Water (US Environmental Protection Agency)
P/CC	Presidential/Congressional Commission
PCR	Polymerase chain reaction
PCR-RFLP	Polymerase chain reaction-restriction fragment length polymorphism
PFGE	Pulsed field gel electrophoresis
$PM_{2.5}$	Particulate Matter with a diameter smaller than 2.5 microns
QALYs	Quality-adjusted life years
RAF	Risk Assessment Forum (US Environmental Protection Agency)
RAPD	Random amplification of polymorphic DNA
rDNA	Ribosomal deoxyribonucleic acid
REP-PCR	Repetitive element polymerase chain reaction
R&D	Research and development
SARS	Severe Acute Respiratory Syndrome
SDWA	Safe Drinking Water Act
SNP	Single nucleotide polymorphism
TAMU	Texas A&M University
TB	Tuberculosis
TCCR	Transparency, Clarity, Consistency, and Reasonableness
U.S.	United States
USDA	U.S. Department of Agriculture

VBNC	Viable-but-not-culturable
vCJD	variant Creutzfeldt-Jacob Disease
VOI	Value of information
WHO	World Health Organization

11. REFERENCES

Abromowitz, M. and I.A. Stegun. (1964) Handbook of Mathematical Functions with Formulas, Graphs, and Mathematical Tables. National Bureau of Standards Applied Mathematics Series – 55.

Airoldi, M. and A. Morton. (2009) Adjusting life for quality or disability: stylistic difference or substantial dispute? *Health Economics* 18(11):1237-1247.

Akkina, J.E., Hogue, A.T., Angule, F.J., Johnson, R., Peterson, K.E., Saini, P.K., Fedorka-Cray, P.J., and W.D. Schlosser. (1999) Epidemiologic aspects, control, and importance of multiple-drug resistant *Salmonella* Typhimurium DT104 in the United States. *Journal of American Veterinary Medical Association* 214:790-798.

Alexopoulos, C.J., C.W. Mims, and M. Blackwell. (1996) Introductory Mycology, 4th Edition. John Wiley & Sons, Inc, Hoboken, N.J.

Anderson, R.M. and R. May. (1991) Infectious Diseases of Humans: Dynamics and Control. New York: Oxford University Press.

Anderson, Y.S., Gillin, F.D., and Eckmann, L. (2006) Adaptive Immunity-Dependent Intestinal Hypermotility Contributes to Host Defense against *Giardia* spp. *Infection and Immunity* 74(4):2473–2476.
http://www.ncbi.nlm.nih.gov/pmc/articles/PMC1418922/pdf/1556-05.pdf

Association of Analytical Communities (AOAC International). (2007) Official Methods of Analysis. Eighteenth edition. Revision 2.

Armstrong, T.W. and C.N. Haas. (2008) Legionnaires' disease: Evaluation of a quantitative microbial risk assessment model. *Journal of Water and Health* 6(2):149-166.

Ashbolt, N.J., Schoen, M.E., Soller, J.A. and D.J. Roser. (2010) Predicting pathogen risks to aid beach management: the real value of quantitative microbial risk assessment (QMRA). *Water Research* 44(16) 4692-4703.

Bartrand, T.A., Weir, M.H., and C.N. Haas. (2008) Dose-response models for inhalation of Bacillus anthracis spores: Interspecies comparisons. *Risk Analysis* 28(4):1115-24.

Battista, J.R. and A.M. Earl. (2004) Mutagenesis and DNA Repair. In R.V. Miller and M.J. Day, Editors. Microbial Evolution: Gene Establishment, Survival, and Exchange. Washington, DC: ASM Press, pp. 3-11.

Batz. (2004) Identifying the most significant microbiological foodborne hazards to public health: a new risk-ranking model. Food Safety Research Consortium, Discussion Paper Series, Number 1, September 2004.
http://www.rff.org/RFF/Documents/FRSC-DP-01.pdf

Bernardo, J.M. and A.F.M. Smith. (1994) Bayesian theory. *Statistical Methods and Applications* 3(1):155-160. Also in paperback book (Bayesian Theory - Wiley Series in Probability and Statistics - April 24, 2000).

Berry, Donald A. (1996) Statistics: A Bayesian Perspective. Belmont, CA: Duxbury Press, Wadworth Publ. Co.

Bisson, I.-A., Marra, P.P., Burtt E.H., Sikaroodi, M, and P. M. Gillevet. (2007) A molecular comparison of plumage and soil bacteria across biogeographic, ecological, and taxonomic scales. *Microbial Ecology* 54:65-81.

Blaser, M.J. and D. Kirschner. (1999) Dynamics of *Helicobacter pylori* colonization in relation to the host response. *Proceedings of the National Academy of Sciences* 96(15):8359-8364.

Blaser, M.J., and D. Kirschner. (2007) The equilibria that allow bacterial persistence in human hosts. *Nature* 449(7164):843-849.

Boerlijst, M.C., Bonhoeffer, S., and M.A. Nowak. (1996) Viral quasi-species and recombination. *Proceedings: Biological Sciences* 263(1376):1577-1584.

Bollaerts, K., Aerts, M., Faes, C., Grijspeerdt, K., Dewulf, J., and K. Mintiens. (2008) Human salmonellosis: Estimation of dose-illness from outbreak data. *Risk Analysis* 28(2):427-440.

Bogosian, G., Morris, P.J.L., and J.P. O'Neil. (1998) A mixed culture recovery method indicates that enteric bacteria do not enter the viable but nonculturable state. *Applied Environmental Microbiology,* 64(5):1736-1742.

Bogosian, B.J., and E.V. Bourneuf. (2001) A matter of bacteria life and death. *EMBO Reports* 2(9):770-774.

Brynestad, S., Braute, L., Luber, P., and E. Bartelt. (2008) Quantitative microbiological risk assessment of campylobacteriosis cases in the German population due to consumption of chicken prepared in homes. *International Journal of Risk Assessment and Management* 8(3):194-213.

Carlin, B.P. and T.A. Louis. (2001) Bayes and Empirical Bayes Methods for Data Analysis. Second edition. New York: Chapman and Hall.

Center for Food Safety and Applied Nutrition (CFSAN). (2001) Bacteriological Analytical Manual (BAM). CFSAN, FDA. http://www.cfsan.fda.gov/~ebam/bam-toc.html

Center for Food Safety and Applied Nutrition (CFSAN). (2006) The Bad Bug Book: Foodborne Pathogenic Organisms and Natural Toxins Handbook. Washington, DC: FDA. http://vm.cfsan.fda.gov/~mow/intro.html

Cleaveland, S., Laurenson, M.K., and L.H. Taylor. (2001) Diseases of humans and their domestic mammals: pathogen characteristics, host range, and the risk of emergence, *Philosophical Transactions of the Royal Society* B 356:991–999.

Codex (Codex Alimentarius Commission). (1999) Principles and Guidelines for the Conduct of Microbial Risk Assessment, CAC/GL-30. http://www.who.int/foodsafety/publications/micro/cac1999/en/ www.codexalimentarius.net/download/standards/357/CXG_030e.pdf

Codex (Codex Alimentarius Commission). (2007a) Principles and Guidelines for the Conduct of Microbiological Risk Management. CAC/GL 63-2007 http://www.codexalimentarius.net/web/more_info.jsp?id_sta=10741

Codex (Codex Alimentarius Commission). (2007b) Working Principles for Risk Analysis for Food Safety for Application by Governments. CAC/GL 62-2007. ftp://ftp.fao.org/docrep/fao/010/a1550t/a1550t00.pdf

Coleman, M. and H. Marks. (2000) Mechanistic modeling of salmonellosis. *Quantitative Microbiology* 2:227-247.

Coleman, M.E., Marks, H.M., Golden, N.J., and H.K. Latimer. (2004) Discerning strain effects in microbial dose-response data. *Journal of Toxicology and Environmental Health, Part A: Current Issues* 67:667-685.

Covacci, A., and R. Rappuoli. (1998) *Helicobacter pylori*: Molecular evolution of a bacterial quasi-species. *Current Opinion in Microbiology* 1(1):96-102.

Cox, L.A. (2006) Quantitative Health Risk Analysis Methods: Modeling the Human Health Impacts of Antibiotics Used in Food Animals. New York: Springer Science.

Crabtree, K.D., Gerba, C.P., Rose, J.B., and C.N. Haas. (1997) Waterborne adenovirus: A risk assessment. *Water Science and Technology* 35(11-12):1-6.

Craun, G.F., Brunkard, J.M., Yoder, J.S., Roberts, V.A., Carpenter, J., Wade, T., Calderon, R.L., Roberts, J.M., Beach, M.J. and S.L. Roy. (2010) Causes of outbreaks associated with drinking water in the United States from 1971 to 2006. *Clinical Microbiological Reviews* 23(3), 507-528.

Cullen, A.C. (1999) Probabilistic Techniques in Exposure Assessment: A Handbook for Dealing with Variability and Uncertainty in Models and Inputs. New York: Springer, 352 pp.

de Koning, H.W. (1987) Setting Environmental Standards: Guidelines for Decision-Making. World Health Organization, Geneva, Switzerland.

Dennis, S.B., Kause, J., Losikoff, M., Engeljohn, D.L., and Buchanan, R. (2008) Chapter 5. Using Risk Analysis for Microbial Food Safety Regulatory Decision-Making, pp. 137-175 In: Microbial Risk Analysis of Foods, Series Editor: Michael P. Doyle; Editor: Donald Schaffner. ASM Press, Washington, DC (978-1-55581-461-8).

Disney, W.T., and M.A. Peters. (2003) Simulation modeling to derive the value of information for risky animal-disease import decisions. *Preventive Veterinary Medicine* 61:171-184.

Dobrindt, U., Hochhut, B., Hentschel, U., and J. Hacker. (2004) Genomic islands in pathogenic and environmental microorganisms. *Nature Reviews Microbiology* 2:414-424.

Dufour, A.P. (1984) Health Effects Criteria for Fresh Recreational Waters. U.S. Environmental Protection Agency, Cincinnati, OH. EPA-600/1-84-004. http://www.epa.gov/microbes/frc.pdf

Dwyer, J., Picciano, M.F, and D.J. Raiten. (2003) Future directions for the integrated CSFII-NHANES: What we eat in America–NHANES. *Journal of Nutrition* 133:576S-581S.

Ebel, E.D., Schlosser, W.D., Orloski, K., Kause, J., Roberts, T., Narrod, C., Malcolm, S., Coleman, M., and M. Powell. (2003) A Risk Assessment of *Escherichia coli* O157:H7 in Ground Beef. *In* Torrence, M.E. and Isaacson, R.E. (eds.) Microbial Food Safety in Animal Agriculture: Current Topics. Ames, Iowa: Iowa State Press.

Englehardt, J.D. (2004) Predictive Bayesian dose-response assessment for appraising absolute health risk from available information. *Human and Ecological Risk Assessment* 10:69-78.

Englehardt, J.D. and J. Swartout. (2004) Predictive population dose-response assessment for *Cryptosporidium parvum*: Infection endpoint. *Journal of Toxicology and Environmental Health, Part A: Current Issues* 67(8-10):651-666.

Englehardt, J.D. and J. Swartout. (2006) Predictive Bayesian microbial dose-response assessment based on suggested self-organization in primary illness response: *C. parvum. Risk Analysis* 26(2):543-554.

Environmental Protection Agency (EPA). (1989) National Primary Drinking Water Regulations: Filtration, Disinfection, Turbidity, *Giardia lamblia*, Viruses, *Legionella*, and Heterotrophic Bacteria. Final Rule. Federal Register 54(124):27486. [Also known as the surface water treatment rule (SWTR)]

Environmental Protection Agency (EPA). (1992) Framework for Ecological Risk Assessment. EPA/630/R-92/001.
http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=30759&partner=ORD_NCEA
Note: This Framework served as the foundation for, and has been superseded by, EPA's 1998 Ecological Risk Assessment Guidelines.

Environmental Protection Agency (EPA). (1994) Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimeter. EPA/600/8-90/066F.

Environmental Protection Agency (EPA). (1997a) Exposure Factors Handbook. Office of Research and Development, National Center for Environmental Assessment. Washington, DC. EPA/600/P-95/002Fa.

Environmental Protection Agency (EPA). (1997b) Guiding Principles for Monte Carlo Analysis. EPA/630/R-97/001.

Environmental Protection Agency (EPA). (1998a) Guidelines for Ecological Risk Assessment. EPA/630/R095/002F. EPA, Washington, DC.
http://www.epa.gov/raf/publications/pdfs/ECOTXTBX.PDF

Environmental Protection Agency (EPA). (1998b) *Giardia*: Human Heath Criteria Document. EPA-823-R-002.

Environmental Protection Agency (EPA). (1999) *Giardia*: Drinking Water Health Advisory. EPA-822-R-99-008.

Environmental Protection Agency (EPA). (2000a) Risk Characterization Handbook. EPA-100-B-00-002. EPA, Washington, DC. http://www.epa.gov/osa/spc/2riskchr.htm

Environmental Protection Agency (EPA). (2000b) Estimated Per Capita Water Ingestion in the United States. Publication No. EPA-822-00-008, 1-208. U.S. Government Printing Office, Washington, DC.
http://www.epa.gov/waterscience/criteria/drinking/percapita/index.html

Environmental Protection Agency (EPA). (2000c) Methodology for Deriving Ambient Water Quality Criteria for the Protection of Human Health. EPA-822-B-00-004.
http://www.epa.gov/waterscience/humanhealth/method/complete.pdf

Environmental Protection Agency (EPA). (2000d) EPA Science Policy Council Peer Review Handbook. Second Edition. Washington, DC. EPA-100-B-00-001.

Environmental Protection Agency (EPA). (2002a) Lessons Learned on Planning and Scoping for Environmental Risk Assessments. EPA Science Policy Council, Washington, DC.

Environmental Protection Agency (EPA). (2002b) Guidelines for Ensuring and Maximizing the Quality, Objectivity, Utility, and Integrity, of Information Disseminated by the Environmental Protection Agency. EPA/260R-02-008. http://www.epa.gov/quality/informationguidelines/documents/EPA_InfoQualityGuidelines.pdf

Environmental Protection Agency (EPA). (2003a) Framework for Cumulative Risk Assessment. Office of Research and Development, National Center for Environmental Assessment, Washington, DC. EPA/630/P-02/001F. http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=54944

Environmental Protection Agency (EPA). (2003b) Assessment Factors. Science Policy Council. EPA 100/B-03/001. http://www.epa.gov/spc/pdfs/assess2.pdf

Environmental Protection Agency (EPA). (2004a) An Expert Judgment Assessment of the Concentration-Response Relationship Between $PM_{2.5}$ Exposure and Mortality. Office of Air Quality Planning and Standards (OAQPS) Report and Peer Review of the Report. Report: http://www.epa.gov/ttn/ecas/regdata/Benefits/pmexpert.pdf Peer review of the report: http://www.epa.gov/ttn/ecas/regdata/Benefits/memo_7.30.04.pdf

Environmental Protection Agency (EPA). (2004b) An Examination of EPA Risk Assessment Principles and Practices. Office of the Science Advisor Staff Paper. EPA/100/B-04/001. http://www.epa.gov/OSA/pdfs/ratf-final.pdf

Environmental Protection Agency (EPA). (2004c) Water quality standards for coastal and great lakes recreation waters final rule. *Federal Register* 69(220):67217-43.

Environmental Protection Agency (EPA). (2004d) Risk Assessment Guidance for Superfund Volume I: Human Health Evaluation Manual (Part E, Supplemental Guidance for Dermal Risk Assessment) Final (RAGS-E). EPA/540/R/99/005.

Environmental Protection Agency (EPA). (2005a) Guidance on Selecting Age Groups for Monitoring and Assessing Childhood Exposures to Environmental Contaminants. National Center for Environmental Assessment, Washington, DC. EPA/630/P-03/003F. http://cfpub.epa.gov/ncea/CFM/recordisplay.cfm?deid=146583.

Environmental Protection Agency (EPA). (2005b) Rule to reduce interstate transport of fine particulate matter and ozone (Clean Air Interstate Rule); revisions to acid rain program; revisions to the NOX SIP call; final rule. *Federal Register* 70(91):25162-25405. http://www.epa.gov/air/interstateairquality/rule.html.

Environmental Protection Agency (EPA). (2006a) National primary drinking water regulations: Long term 2 enhanced surface water treatment rule - final. *Federal Register* 71(3):654-785.

Environmental Protection Agency (EPA). (2006b) Summary of NCEA Colloquium on Current Use and Future Needs of Genomics in Ecological and Human Health Risk Assessment. EPA/600/R-04/039F.
http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=149984.

Environmental Protection Agency (EPA). (2006c) EPA Science Policy Council Peer Review Handbook. Third edition EPA/100/B-06/002. Washington, DC.

Environmental Protection Agency (EPA). (2007a) Pesticides; Data Requirements for Biochemical and Microbial Pesticides. Federal Register 72 (207):60988-61025. October 26, 2007.

Environmental Protection Agency (EPA). (2007b) Thesaurus of Terms Used in Microbial Risk Assessment.
http://www.epa.gov/waterscience/criteria/humanhealth/microbial/thesaurus/

Environmental Protection Agency (EPA). (2007c) Compendium of Prior and Current Microbial Risk Assessment Methods for Use as a Basis for the Selection, Development, and Testing of a Preliminary Microbial Risk Assessment Framework. EPA/600/R-07/129. National Homeland Security Research Center.
http://www.epa.gov/NHSRC/pubs/600r07129.pdf.

Environmental Protection Agency (EPA). (2008) Scientific and Ethical Approaches for Observational Exposure Studies. EPA 600/R-08/062.
http://www.epa.gov/nerl/sots/SEAOES_doc20080707.pdf

Environmental Protection Agency (EPA). (2009a) Protocol for Microbial Risk Assessment. Draft July 30, 2009. Office of Science and Technology, Office of Water, Washington, DC. and EPA Science Advisory Board Review of EPA's Microbial Risk Assessment Protocol (EPA-SAB-10-008)
http://yosemite.epa.gov/sab/sabproduct.nsf/7FAA3A556A92CF21852576160064DEC2/$File/Draft+MRA+Protocol+July+30+2009+for+DWC+Sept+21-22+2009+Meeting.pdf
http://yosemite.epa.gov/sab/sabproduct.nsf/07322F6BB8E5E80085257746007DC64F/$File/EPA-SAB-10-008-unsigned.pdf

Environmental Protection Agency (EPA). (2009b) Using Probabilistic Methods to Enhance the Role of Risk Analysis in Decision-Making with Case Study Examples. EPA/100/R-09/001
http://www.epa.gov/osa/spc/expertelicitation/index.htm

Environmental Protection Agency (EPA). (2009c) DRAFT Expert Elicitation Task Force White Paper. January 6, 2009 External Review Draft.
http://www.epa.gov/osa/spc/expertelicitation/index.htm

Environmental Protection Agency (EPA). (2010) Integrated Risk Information System Glossary. Accessed October 1, 2010

http://www.epa.gov/ncea/iris/help_gloss.htm

European Commission, Scientific Steer Committee (ECSSC). (2003) Risk Assessment of Food Borne Bacterial Pathogens: Quantitative Methodology Relevant for Human Exposure. Final report. SSC Task Force Report on Harmonization of Risk Assessment Procedures.

Executive Order 12866. (1993) Executive Order 12866 of September 30, 1993, Regulatory Planning and Review. Federal Register 58 (190). http://www.reginfo.gov/public/jsp/Utilities/EO_12866.pdf

Farber, J.M., Ross, W.H., and J. Harwig. (1996) Health risk assessment of *Listeria monocytogenes* in Canada. *International Journal of Food Microbiology* 30(1-2):145-156.

Food and Agriculture Organization/World Health Organization (FAO/WHO). (1997) Risk Management and Food Safety. Report of a Joint FAO/WHO Consultation Rome, Italy, 27 to 31 January 1997, FAO Food and Nutrition Paper Number 65. ftp://ftp.fao.org/docrep/fao/w4982e/w4982e00.pdf

Food and Agriculture Organization/World Health Organization (FAO/WHO). (2003) Hazard Characterization for Pathogens in Food and Water, Guidelines. Microbiological Risk Assessment Series 3. http://www.fao.org/docrep/006/y4666e/y4666e00.htm http://www.cepis.org.pe/bvsacg/e/cd-cagua/guias/c.referencias/07.pathogen.pdf

Food and Agriculture Organization/World Health Organization (FAO/WHO). (2004) Risk Assessment of *Listeria monocytogenes* in Ready-to-Eat Foods. ftp://ftp.fao.org/es/esn/jemra/mra4_en.pdf

Food and Agriculture Organization/World Health Organization (FAO/WHO). (2006) Food Safety Risk Analysis: A Guide for National Food Safety Authorities. FAO: Food and Nutrition Paper 87. ftp://ftp.fao.org/docrep/fao/009/a0822e/a0822e00.pdf

Food and Agriculture Organization/World Health Organization (FAO/WHO). (2009) Risk Characterization of Microbiological Hazards in Food: Guidelines. Microbiological Risk Assessment Series17. ftp://ftp.fao.org/ag/agn/jemra/MRA17_05.10.09_f.pdf

Food and Drug Administration (FDA). (1999) A Defined-Risks Approach to the Regulatory Assessment of the Use of Neoplastic Cells as Substrates for Viral Vaccine Manufacture. Developed by Andrew M. Lewis Jr., Philip Krause, and Keith Peden for the Cell Substrate-Adventitious Agent Working/Interest Group. http://www.fda.gov/cber/minutes/brief091499.pdf

Food and Drug Administration (FDA). (2002) Initiation and Conduct of All "Major" Risk Assessments within a Risk Analysis Framework: A Report by the CFSAN Risk Analysis

Working Group. Center for Food Safety and Applied Nutrition (CFSAN). http://www.cfsan.fda.gov/~dms/rafw-toc.html

Food and Drug Administration (FDA). (2005) Quantitative Risk Assessment on the Public Health Impact of Pathogenic *Vibrio parahaemolyticus* in Raw Oysters. http://www.cfsan.fda.gov/~dms/vpra-toc.html

Food and Drug Administration (FDA). (2007) An Overview of the CARVER Plus Shock Method for Food Sector Vulnerability Assessments. http://www.fsis.usda.gov/PDF/Carver.pdf

Food and Drug Administration/U.S. Department of Agriculture/Centers for Disease Control and Prevention (FDA/USDA/CDC). (2003) Quantitative Assessment of Relative Risk to Public Health from Foodborne *Listeria monocytogenes* Among Selected Categories of Ready-to-Eat Foods. FDA Center for Food Safety and Applied Nutrition (CFSAN), USDA Food Safety and Inspection Service, and Centers for Disease Control and Prevention, Washington, DC. http://www.foodsafety.gov/~dms/lmr2-toc.html.

Ferson, S. (1996) What Monte Carlo methods cannot do. *Human and Ecological Risk Assessment* 2:990-1007.

Fischer-Le Saux, M., Hervio-Heath, D., Loaec, S., Colwell, R.R., and M. Pommepuy. (2002) Detection of cytotoxin-hemolysin mRNA in nonculturable populations of environmental and clinical *Vibrio vulnificus* strains in artificial seawater. *Applied and Environmental Microbiology* 68:5641-5646.

Fischhoff, B., Lichtenstein, S., Slovic, P., Derby, S.L., and R.L. Keene. (1981) Acceptable Risk. New York, NY: Cambridge University Press.

Frey, H.C., Mokhtari, A., and T. Danish. (2003) Evaluation of Selected Sensitivity Analysis Methods Based Upon Applications to Two Food Safety Process Risk Models. Prepared for: Office of Risk Assessment and Cost-Benefit Analysis USDA, Washington, DC. http://www.ce.ncsu.edu/risk/Phase2Final.pdf

Frey, H.C., Mokhtari, H., and J. Zheng. (2004) Recommended Practice Regarding Selection, Application, and Interpretation of Sensitivity Analysis Methods Applied to Food Safety Process Risk Models. Prepared for the Office of Risk Assessment and Cost-Benefit Analysis, U.S. Department of Agriculture. http://www.ce.ncsu.edu/risk/Phase3Final.pdf

Frost, F.J., Roberts, M., Kunde, T.R., Craun, G., Tollestrup, K., Harter, L., and T. Muller. (2005) How clean must our drinking water be: The importance of protective immunity. *Journal of Infectious Diseases* 191:809-814.

Furumoto, W.A. and R. Mickey. (1967) A mathematical model for the infectivity-dilution curve of tobacco mosaic virus: theoretical considerations. *Virology* 32(2):216-23.

Government Accountability Office (GAO). (2001) Chemical Risk Assessment: Selected Federal Agencies' Procedures, Assumptions, and Policies. GAO-01-810. Washington, DC: http://www.gao.gov/new.items/d01810.pdf

Gelman, A., Carlin, J.B., Stern, H.S., and D.B. Rubin. (2004) Bayesian Data Analysis, Second Edition. Boca Raton, Florida: Chapman and Hall/CRC.

Gerba, C.P., Rose, J.B., Haas, C.N., and K.D. Crabtree. (1996) Waterborne rotavirus: a risk assessment. *Water Research* 30:2929-2940.

Gilks, W., Richardson, S., and D.J. Spiegelhalter (eds.). (1996) Markov Chain Monte Carlo in Practice. London, UK: Chapman and Hall.

Gold, M.R., Stevenson, D., and D.G. Fryback. (2002) HALYs and QALYs and DALYs, oh my: similarities and differences in summary measures of population health. *Annual Review Public Health* 23:115-134.

Guzmán, E., Romeu, A., and S. Garcia-Vallve. (2008) Completely sequenced genomes of pathogenic bacteria: A review. *Enfermedades Infecciosas y Microbiologia Clinica* 26:88-98.

Haas, C.N., Rose, J.B., Gerba, C., and S. Regli. (1993) Risk assessment of virus in drinking water. *Risk Analysis* 13(5):545-552.

Haas, C.N., Crockett, C.S., Rose, J.B., Gerba, C.P., and A.M. Fazil. (1996) Assessing the risks posed by oocysts in drinking water. *Journal of the American Water Works Association* 88(9):131-136.

Haas, C.N., Rose, J.B., and Gerba, C.P. (1999) Quantitative Microbial Risk Assessment. New York: John Wiley and Sons.

Haimes, Y.Y. (2004) Risk Modeling, Assessment, and Management. Second edition. New York: John Wiley & Sons.

Hethcote, H. (1976) Qualitative analyses of communicable disease models. *Mathematical Biosciences* 28:335-356.

Hethcote, H.W. (2000) The mathematics of infectious diseases. *SIAM Review* 42:599-653.

Hoeting, J.A., Madigan, D., Raftery, A.E., and C.T. Volinsky. (1999) Bayesian model averaging: A tutorial. *Statistical Science* 14(4):382–417.

Hopkins, R.S., Gaspard, G.B., Williams, F.P., Karlin, R.J., Cukor, K.G., and N.R. Blacklow. (1984) A community waterborne gastroenteritis outbreak: evidence for rotavirus as the agent. *American Journal of Public Health* 74:263-265.

Hirshliefer, J. and J. Riley. (1992) The Analytics of Uncertainty and Information. Cambridge: Cambridge University Press.

Humber, J.M.; R.F. Almeder. (1986) Quantitative Risk Assessment. Biomedical Ethics Reviews. Clifton, New Jersey: Humana Press.

Huq, A., Rivera, I.N.G., and R.R. Colwell. (2000) Epidemiological Significance of Viable But Nonculturable Microorganisms. *In:* Colwell, R.R. and Grimes, D.J. (eds.) Nonculturable Microorganisms in the Environment. Washington, DC: ASM Press.

Hurd, H.S. and J.B. Kaneene. (1993) The application of simulation models and systems analysis in epidemiology: A review. *Preventive Veterinary Medicine* 15:81-99.

International Life Sciences Institute (ILSI). (2000) Revised Framework for Microbial Risk Assessment. Washington, DC: ILSI Press.
http://rsi.ilsi.org/Publications/MRAWorkbook.htm

Interagency Risk Assessment Consortium (IRAC). (2000) Public Meeting on Food Safety Risk Analysis Clearinghouse Data Quality Objectives. December 5, 2000.
http://foodrisk.org/IRAC/events/2000-12-05/index.cfm

Jameel, S. (1999) Molecular biology and pathogenesis of hepatitis E virus. *Expert Reviews in Molecular Medicine* (6 December):1-16.
http://www-ermm.cbcu.cam.ac.uk/99001271h.htm

Kaplan, S. and B.J. Garrick. (1981) On the quantitative definition of risk. *Risk Analysis* 1:11-27.

Kaplan, S. (2000) Combining probability distributions from experts in risk analysis. Letter to the editor. *Risk Analysis* 20(2):155-156.

King, A.A., Ionides, E.L., Pascual, M., and M.J. Bouma. (2008) Inapparent infections and cholera dynamics. *Nature* 454:877-880.

Knapp, S., Hacker, J., Jarchau, T., and W. Goebel. (1986) Large, unstable inserts in the chromosome affect virulence properties of uropathogenic *Escherichia coli* 06 Strain 536. *Journal of Bacteriology* 168:22-30.

Ko, G., Thompson, K.M., and E.A. Nardell. (2004) Estimation of tuberculosis risk on a commercial airliner. *Risk Analysis* 26(2):379-388.

Kodell, R.L., Kang, S.-H., and J.J. Chen. (2002) Statistical models of health risk due to microbial contamination of foods. *Environmental and Ecological Statistics* 9:259-271.

Kosa, K.M.; Cates, S.C.; Karns, S.; Godwin, S.L.; and D. Chambers. (2007) Consumer knowledge and use of open dates: Results of a web-based survey. *Journal of Food Protection* 70(5):1213-1219.

Labbe, R.G. and S. Garcia. (2001) Guide to Foodborne Pathogens. Hoboken, NJ: John Wiley & Sons.

Lammerding, A.M., and A. Fazil. (2000) Hazard identification and exposure assessment for microbial food safety risk assessment. International Journal of Food Microbiology 58(3):147-57.

Law, A.M., and W.D. Kelton. (2000) Simulation Modeling and Analysis, Third Edition. New York: McGraw-Hill Companies.

Lave, L., and T. Romer. (1981) A Survey of Safety Levels in Federal Regulation. Nuclear Regulatory Commission. NUREG/CR-2226.

Li, X.J., and H.H. Wang. (2010) Tetracycline resistance associated with commensal bacteria from representative ready-to-consume deli and restaurant foods. *Journal of Food Protection* (73):1841-1848.

Lerat, E., and N.A. Moran. (2004) The evolutionary history of quorum-sensing systems in bacteria. *Molecular Biology and Evolution* 21:903-913.

Liu, D. (2006) Identification, subtyping, and virulence determination of *Listeria monocytogenes*, an important foodborne pathogen. *Journal of Medical Microbiology* 55:645-659.

Lowrance, W.W. (1976) Of Acceptable Risk: Science and the Determination of Safety. Los Altos, CA: William Kaufmann, Inc.

Lundgren, R. and A. McMakin. (1998) Risk Communication: A Handbook for Communicating Environmental, Safety, and Health risks. Second edition. Columbus, OH: Battelle Press.

Lunn, DJ, Thomas, A., Best, and N.D. Spiegelhalter. (2000) WinBUGS - A Bayesian modeling framework: Concepts, structure, and extensibility. *Statistics and Computing* 10(4):325-337.

Mahenthiralingam, E., J. Bischof, S. K. Byrne, C., Radomski, C., Davies, J.E., Av-Gay, Y., and P. Vandamme. (2000) DNA-based diagnostic approaches for identification of *Burkholderia cepacia* complex, *Burkholderia vietnamiensis*, *Burkholderia multivorans*,

Burkholderia stabilis, and *Burkholderia cepacia* genomovars I and III. *Journal of Clinical Microbiology* 38(9):3165-3173.

Manning, S.D., Motiwala, A.S., Springman, A.C., Qi, W., Lacher, D.W., Ouellette, L.M., Mladonicky, J.M., Somsel, P., Rudrik, J.T., Dietrich, S.E., Zhang, W., Swaminathan, B., Alland, D., and T.S. Whittam. (2008) Variation in virulence among clades of *Escherichia coli* O157:H7 associated with disease outbreaks. *Proceedings of the National Academies of Science* 105:4868-4873.

McBride, G.B., Till, D., Ryan, T., Ball, A., Lewis, G., Palmer, S., and P. Weinstein. (2002) Freshwater Microbiology Research Programme: Pathogen Occurrence and Human Health Risk Assessment Analysis, Technical Publication, Ministry for the Environment, Wellington, 93 pp. http://www.mfe.govt.nz/publications/water/freshwater-microbiology-nov02/freshwater-microbiology-nov02.pdf

Mead, P.S.., Slutsker, L., Dietz, V., McCaig, L.F., Bresee, J.S., Shapiro, C., Griffin, P.M., and R.V. Tauxe. (1999) Food-related illness and death in the United States. *Emerging Infectious Diseases* 5(5):607-625. http://www.cdc.gov/ncidod/EID/vol5no5/pdf/mead.pdf

Medema, G.J., Teunis, P.F., Havelaar, A.H., and C.N. Haas. (1996) Assessment of the dose-response relationship of *Campylobacter jejuni*. *International Journal of Food Microbiol*ogy 30(1-2):101-11.

Messner, M.J., Chappell, C.L., and P.C. Okhuysen. (2001) Risk assessment for *Cryptosporidium*: A hierarchical Bayesian analysis of human dose-response data. *Water Research* 35(16):3934-3940.

Moon, H., Kim, H-J., Chen, J.J., and R.L. Kodell. (2005) Model averaging using the Kullback Information Criterion in estimating effective doses for microbial infection and illness. *Risk Analysis* 25(5):1147-1159.

Morgan, M.G. and M. Henrion. (1990) Uncertainty: A Guide to Dealing with Uncertainty in Quantitative Risk and Policy Analysis. Cambridge: Cambridge University Press.

Morgan, G.M., Fischhoff, B., Bostrom, A., and Atman, C.J. (2002) Risk Communication A Mental Models Approach. New York: Cambridge University Press.

National Advisory Committee on Microbiological Criteria for Foods (NACMCF). (1997) Hazard Analysis and Critical Control Point (HAACP) Principles and Application Guidelines. Adopted August 14, 1997. http://www.fda.gov/Food/FoodSafety/HazardAnalysisCriticalControlPointsHACCP/HACCPPrinciplesApplicationGuidelines/default.htm

Namata, H., Aerts, M., Faes, C., and P. Teunis. (2008) Model averaging in microbial risk assessment using fractional polynomials. *Risk Analysis* 28(4):891-905.

Nauta, M.J. (2002) Modeling bacterial growth in quantitative microbiological risk assessment: Is it possible? *International Journal of Food Microbiology* 73:297-304.

Nauta, M.J. (2005) Microbiological risk assessment models for partitioning and mixing during food handling. *International Journal of Food Microbiology* 100:311-322.

National Committee on Radiation Programs (NCRP). (1996) A Guide for Uncertainty Analysis in Dose and Risk Assessments Related to Environmental Contamination. NCRP, Scientific Committee 64-17, Washington, DC. NCRP Commentary No. 14 [as cited in EPA, 1997b, page 14].

Newsome, R., Tran, N., Paoli, G.M., Jaykus, L.A., Tompkin, B., Miliotis, M., Ruthman, T., Hartnett, E., Busta, F.F., Petersen, B., Shank, F., McEntire, J., Hotchkiss, J., Wagner, M., and D.W. Schaffner. (2009) Development of a risk-ranking framework to evaluate potential high-threat microorganisms, toxins, and chemicals in food. *Journal of Food Science* 74(2):R39-R45.

National Research Council (NRC). (1983) Risk Assessment in the Federal Government: Managing the Process. Washington, DC: National Academy Press. http://www.nap.edu/catalog.php?record_id=366

National Research Council (NRC). (1994) Science and Judgment in Risk Assessment. Washington, DC: National Academies Press. http://www.nap.edu/catalog.php?record_id=2125.

National Research Council (NRC). (1996) Understanding Risk: Informing Decisions in a Democratic Society. Washington, DC: National Academies Press. http://www.nap.edu/catalog.php?record_id=5138

National Research Council (NRC). (2003) Scientific Criteria to Ensure Safe Food. Washington, DC: National Academies Press. http://books.nap.edu/catalog.php?record_id=10690

National Research Council (NRC). (2005) Reopening Public Facilities After a Biological Attack: A Decision-Making Framework. Washington, DC: National Academies Press. http://www.nap.edu/catalog.php?record_id=11324

National Research Council (NRC). (2008) Public Participation in Environmental Assessment and Decision Making. Washington, DC: National Academies Press. http://www.nap.edu/catalog.php?record_id=12434

National Research Council (NRC). (2009) Science and Decisions: Advancing Risk Assessment. Washington, DC: National Academies Press. http://www.nap.edu/catalog.php?record_id=12209

O'Donoghue, P. (1995) *Cryptosporidium* and cryptosporidiosis in man and animals. *International journal for Parasitology*: 25:2:139-195.

World Organization for Animal Health (OIE). 1999. Chapter 4. Import risk analysis In: OIE Animal Health Code. World Organisation for Animal Health, Paris, France.

Okhuysen, P.C., Chappell, C.L., Crabb, J.H., Sterling, C.R., and H.L. DuPont. (1999) Virulence of three distinct *Cryptosporidium parvum* isolates for healthy adults. *Journal of Infectious Diseases* 180(4):1275-1281.

Office of Management and Budget (OMB). (2002) Guidelines for ensuring and maximizing the quality, objectivity, utility, and integrity of information disseminated by federal agencies; republication. *Federal Register* 67 (36)8452-8460. http://www.whitehouse.gov/sites/default/files/omb/fedreg/reproducible2.pdf

Office of Management and Budget (OMB). (2003) Circular A-4, Regulatory Analysis (09/17/2003). http://www.whitehouse.gov/sites/default/files/omb/assets/omb/circulars/a004/a-4.pdf

Office of Management and Budget (OMB). (2004) Revised Information Quality Bulletin for Peer Review, April 2004. http://www.whitehouse.gov/omb/inforeg/peer_review041404.pdf

Office of Management and Budget (OMB). (2007a) Final Bulletin for Agency Good Guidance Practices. Bulletin No. 07-02. http://www.whitehouse.gov/omb/memoranda/fy2007/m07-07.pdf.

Office of Management and Budget (OMB). (2007b) M-07-24 Memorandum for the Heads of Executive Departments and Agencies, Subject: Updated Principles for Risk Analysis, September 19, 2007. http://www.whitehouse.gov/sites/default/files/omb/assets/omb/memoranda/fy2007/m07-24.pdf

Office of Management and Budget (OMB). (2010) Agency Checklist: Regulatory Impact Analysis. October 28, 2010. http://www.whitehouse.gov/sites/default/files/omb/inforeg/regpol/RIA_Checklist.pdf

Oscar, T.P. (2005) Validation of lag time and growth rate models for S*almonella typhimurium*: acceptable prediction zone method. *Journal of Food Science* 70(2):M129-M137.

Office of Science and Technology Policy (OSTP). (2010). Memorandum for the Heads of Executive Departments and Agencies, Subject: Scientific Integrity. December 17, 2010. http://www.whitehouse.gov/sites/default/files/microsites/ostp/scientific-integrity-memo-12172010.pdf.

Ouchi, F. (2004) A Literature Review on the Use of Expert Opinion in Probabilistic Risk Analysis. World Bank Policy Research Working Paper, Report Number WPS3201. http://econ.worldbank.org

Parkin, (2008) Foundations and Frameworks for Human Microbial Risk Assessment. http://www.epa.gov/raf/files/epa_mra_fw_comparison_report_0609.pdf

Pedersen A.B., Altizerb, S., Poss, M. Cunningham, A.A., and C.L. Nunn. (2005) Patterns of host specificity and transmission among parasites of wild primates. *International Journal for Parasitology* 35(6):647-657.

Peter, J.B. (ed). (1998) Use and Interpretation of Laboratory Tests in Infectious Disease. 5th Edition. Santa Monica, CA: Specialty Laboratories.

Pinsky, P.F. (2000) Assessment of risks from long term exposure to waterborne pathogens. *Environmental and Ecological Statistics* 7:155-175.

P/CC (Presidential/Congressional Commission on Risk Assessment and Risk Management). (1997) Risk Assessment and Risk Management in Regulatory Decision-Making, Volume 2. Washington, DC: Government Printing Office. http://www.riskworld.com/riskcommission/Default.html

Pouillot, R.; Lubran, M.B; Cates, S. C; S. Dennis. (2010) Estimating parametric distributions of storage time and temperature of ready-to-eat foods for U.S. households. *Journal of Food Protection* 73(2):312-321.

Presidential Memorandum. (2009) The White House, Office of the Press Secretary, Memorandum for the Heads of Executive Departments and Agencies, Subject: Scientific Integrity. March 9, 2009. http://www.whitehouse.gov/the-press-office/memorandum-heads-executive-departments-and-agencies-3-9-09

Regli, S., Rose, J.B., Haas, C.N., and C.P. Gerba. (1991) Modeling risks for pathogens in drinking water. *Journal of the American Water Works Association* 83(11):76-84.

Rice, G., Heberling, M.T., Rothermich, M., Wright, J.M., Murphy, P.A., Craun, M.F. and G.F. Craun. (2006) The role of disease burden measures in future estimates of endemic waterborne disease. *Journal of Water and Health* 4 (Suppl 2):187-199.

Riley, S., Fraser, C., Donnelly, C.A., Ghani, A.C., Abu-Raddad, L.J., Hedley, A.J., Leung, G.M., Ho, L.M., Lam, T.H., Thach, T.Q., Chau, P., Chan, K.P., Lo, S.V., Leung, P.Y., Tsang, T., Ho, W., Lee, K.H., Lau, E.M., Ferguson, N.M., and R.M. Anderson. (2003) Transmission dynamics of the etiological agent of SARS in Hong Kong: Impact of public health interventions. *Science* 300:1961-1966.

Rose. J.B., Haas, C.N., and S. Regli. (1991) Risk assessment and control of waterborne giardiasis. *American Journal of Public Health* 81:709-713.

Rose, J.B., and M.D. Sobsey. (1993) Quantitative risk assessment for viral contamination of shellfish and coastal waters. *Journal of Food Protection* 56(12):1043-1050.

Rose, J.B., and C.P. Gerba. (1991) Use of risk assessment for development of microbial standards. *Water Science and Technology* 24:29-34.

Ross, T. and McMeekin, T.A. (1994) Predictive microbiology. *International Journal Food Microbiology* 23:241-264.

Ross, T. and T.A. McMeekin. (2003) Modeling microbial growth within food safety risk assessments. *Risk Analysis* 23(1):179-197.

Ross, T. (2008) Chapter 3. Translating Knowledge of Microbial Ecology into Risk Assessment Models, pp. 51-97 *In:* Microbial Risk Analysis of Foods, Series Editor: Michael P. Doyle; Editor: Donald Schaffner. ASM Press, Washington, DC (978-1-55581-461-8).

Rowman, N.J. (2004) Viable but non-culturable forms of food and waterborne bacteria: Quo vadis? *Trends in Food Science and Technology* 15:462-467. http://www.sciencedirect.com/science?_ob=ArticleURL&_udi=B6VHY-4CVX484-5&_user=5941288&_coverDate=09%2F01%2F2004&_rdoc=1&_fmt=&_orig=search&_sort=d&view=c&_acct=C000050221&_version=1&_urlVersion=0&_userid=5941288&md5=a7b4ed126d6e80d28143b03d4b0ebbca

Schaffner, D.W. (2008) Microbial Risk Analysis of Food. Series Editor: Michael P. Doyle. ASM Press, Washington, DC 270 p. (978-1-55581-461-8).

Schmidt, H. and M. Hensel. (2004) Pathogenicity islands in bacterial pathogenesis. *Clinical Microbiology Reviews* 17:14-56.

Schoen, M.E. and N.J. Ashbolt. (2010) Assessing pathogen risk to swimmers at non-sewage impacted recreational beaches. *Environmental Science and Technology* 44(7):2286-2291.

Sellnow, T.L., Ulmer, R.R., Seeger, M.W., and R.S. Littlefield. (2008) Effective Risk Communication A Message-Centered Approach. *In* Series: Food Microbiology and Food Safety. New York: Springer Publishing.

Sen, K. and N.J. Ashbolt. (2011) Environmental Microbiology: Current Technology and Water Applications. Portland, OR: Caister Academic Press.

Smith, B. and J.D. Oliver. (2006) In situ and in vitro gene expression by *Vibrio vulnificus* during entry into, persistence within, and resuscitation from the viable but nonculturable state. *Applied Environmental Microbiology* 72:1445-1451.

Soller, J.A., Eisenberg, J.N., and A.W. Olivieri. (1999) Evaluation of Pathogen Risk Assessment Framework. Oakland, CA: Eisenberg, Olivieri and Associates.

Soller, J.A., Seto, E.Y., and A.W. Olivieri. (2007) Application of Microbial Risk Assessment Techniques to Estimate Risk Due to Exposure to Reclaimed Waters. WateReuse Foundation, Final Project Report WRF-04-011.

Soller, J.A., and J.N. Eisenberg. (2008) An evaluation of parsimony for microbial risk assessment models. *Environmetrics* 19:61-78.

Soller, J.A. (2009) Potential implications of person-to-person transmission of viral infection to US EPA's Groundwater Rule. *Journal of Water Health* 7(2):208-223

Soller, J.A, Bartrand, T., Ashbolt, N.J., Ravenscroft, J., and T.J. Wade. (2010) Estimating the primary etiologic agents in recreational freshwaters impacted by human sources of faecal contamination. *Water Research* 44:4736 -4747.

Teunis, P.F., van der Heijden, O.G., van der Giessen, J.W.B., and A.H. Havelaar. (1996) The Dose-Response Relation in Human Volunteers for Gastro-Intestinal Pathogens. RIVM (National Institute of Public Health and the Environment) Report No. 284550002.

Teunis, P.F.M., and A.H. Havelaar. (1999) *Cryptosporidium* in Drinking Water: Evaluation of the ILSI/RSI Quantitative Risk Assessment Framework. RVIM Report No. 284 550 006. Bilthoven, The Netherlands.

Teunis, P.F.M., and A.H. Havelaar. (2000) The beta-Poisson model is not a single hit model. *Risk Analysis* 20(4):513-520.

Teunis, P.F., Chappell, C.L., and P.C. Okhuysen. (2002) *Cryptosporidium* dose-response studies: Variation between isolates. *Risk Analysis* 22(1):175-183.

Teunis, P., Takumi, K., and K. Shinagawa. (2004) Dose response for infection by *Escherichia coli* O157:H7 from outbreak data. *Risk Analysis* 24(2):401-407.

Teunis, P.F.M., van den Brandhof, W., Nauta, M., Wagenaar, J., van den Kerkhof, H., and w. Van Pelt. (2005) A reconsideration of the *Campylobacter* dose-response relation. *Epidemiology and Infection* 133:583-592.

Teunis, P.F.M., Ogden, I.D., and N.J.C. Strachan. (2008a) Hierarchical dose response of *E. coli* O157:H7 from human outbreaks incorporating heterogeneity in exposure. *Epidemiology and Infection* 136(6):761-770.

Teunis, P.F.M., Moe, C.L., Liu, P., Miller, S.E., Lindesmith, L., Baric, R.S., Pendu, J.L., and Calderon, R.L. (2008b) Norwalk virus: How infectious is it? *Journal of Medical Virology* 80(8):1468-1476.

U.S. Army Center for Health Promotion and Preventive Medicine (USACHPPM). (2009) Technical Guide 316 – Microbial Risk Assessment for Aerosolized Microorganisms. U.S. Army Center for Health Promotion and Preventive Medicine.

U.S. Department of Agriculture (USDA). (2003) Risk Analysis at FSIS: Standard Operating Procedures. Washington, DC: USDA, FSIS. http://www.fsis.usda.gov/OPPDE/rdad/FRPubs/RASOPs.pdf

U.S. Department of Agriculture (USDA). (2008) Microbiology Laboratory Guidebook. http://www.fsis.usda.gov/science/Microbiological_Lab_Guidebook/index.asp

Vandamme, P., Holmes, B., Vancanney, M., Coenye, T., Hoste, B., Coopman, R., Revets, H., Lauwers, S., Gillis, M., Kersters, K., and J.R.W. Govan. (1997) Occurrence of multiple genomovars of *Burkholderia cepacia* in cystic fibrosis patients and proposal of *Burkholderia multivorans* sp. nov. *International Journal of Systematic Bacteriology* (47):1188–1200.

Vandamme, P., Mahenthiralingam, Holmes, B., Coenye, T., Hoste, B., De Vos, P,. Henry, D., and D.P. Speert. (2000) Identification and population structure of *Burkholderia stabilis* sp. nov. (formerly *Burkholderia cepacia* Genomovar IV). *Journal of Clinical Microbiology* 38:1042-1047.

Vatn, J. (2004) A discussion of the acceptable risk problem. Norwegian University of Science and Technology. http://www.ntnu.no/ross/reports/acceptable_risk.pdf; http://www.ntnu.no/ross/info/notes.php. Posted on February 18, 2004.

Vose, D.J. (2008) Risk Analysis: A Quantitative Guide. Third Edition. Chichester, England: John Wiley & Sons Ltd.

World Health Organization (WHO). (2000) The Interaction between Assessors and Managers of Microbiological Hazards in Food. WHO/SDE/PHE/FOS/007 http://www.who.int/foodsafety/publications/micro/en/march2000.pdf

World Health Organization (WHO). (2001) Water Quality: Guidelines, Standards and Health: Assessment of Risk and Risk Management for Water-Related Infectious Disease. Fewtrell, L., Bartram, J. (eds.) Published on behalf of IWA Publishing, WHO and Swedish Institute for Infectious Disease Control. http://www.who.int/water_sanitation_health/dwq/whoiwa/en/index.html

World Health Organization (WHO). (2005) Water Safety Plans, Managing Drinking-Water Quality from Catchment to Consumer. WHO/SDE/WSH/05.06. http://www.who.int/water_sanitation_health/dwq/wsp0506/en/index.html

World Health Organization/Food and Agriculture Organization (WHO/FAO). (2008) Microbiological Risk Assessment Series 7 - Exposure Assessment of Microbiological Hazards in Food – Guidelines. http://www.fao.org/docrep/010/a0251e/a0251e00.htm

Williams, R.A., and K.M. Thompson. (2004) Integrated analysis: Combining risk and economic assessments while preserving the separation of powers. *Risk Analysis* 24(6):1613-1623.

Wooldridge, M. (2008) Chapter 1. Qualitative Risk Assessment, pp. 1-28 In: Microbial Risk Analysis of Foods, Series Editor: Michael P. Doyle; Editor: Donald Schaffner. ASM Press, Washington, DC (978-1-55581-461-8.)

Xiao, L. (2010) Molecular epidemiology of cryptosporidiosis: an update. Exp Parasitol 124:80-89.

Yakota, F. and K.M. Thompson. (2003) Value of information (VOI) analysis in environmental health risk management (EHRM): Past, present and future. *Risk Analysis* 24(3):635-650.

Yoon, S.H., Park, Y.K., Lee, S., Choi, D., Oh, T.K., Hur, C.G., and J.F. Kim. (2007) *Nucleic Acids Research* 35:D395-D400.

Zhao, P., Zhao, T., Doyle, M.P., Rubino, J.R. and J. Meng. (1998) Development of a model for evaluation of microbial cross-contamination in the kitchen. *Journal of Food Protection* 61:960-963.

Appendix A Example Assumptions

This appendix contains a list of types of assumptions that you may encounter during MRA. Many of these assumptions are due to data gaps. As data become available in the future, these assumptions could be different. Whether any of these assumptions are justifiable and adequate for a given risk assessment can be decided on a case-by-case basis. For example the assumptions about the immune status of the population may be simple (no one has immunity) or complex (based on actual immunity data).

For all assumptions, the strengths, limitations, and implications of the assumption should be fully explored and documented in the risk assessment. This level of transparency helps peer reviewers decide if they agree with the basis of the assumptions and whether the results of the risk assessment seem credible.

Some assumptions are related to the scope of the risk assessment and are less subject to challenge on a scientific basis. For example the choice to evaluate a single agent instead of a mixture of agents is a policy decision about scope. In addition most MRA is limited in scope to estimating risk as a result of a single exposure event. In chemical risk assessment the exposure duration is often a 70-year life span.

General Assumptions:

MRA's focus on known pathogens that contribute significantly to the human disease burden or emerging pathogens for which the potential for disease is recognized, but the disease burden is unknown.

MRA's related to food-borne infections typically focus on a food-pathogen pair. This assumes that a specific pathogen is associated with a specific food. In reality, more than one pathogen could be associated with a specific food (for example, *Salmonella* and *Campylobacter* in chicken) and also different types of foods could be associated with a specific pathogen (for example, *Salmonella* in chicken and pork).

A given MRA's scope reflects the regulatory jurisdiction of the sponsoring agency and the statues behind regulations. For example EPA risk assessments in water media are divided into ambient water which is regulated by the Clean Water Act and drinking water which is regulated by the Safe Drinking Water Act. EPA has not to date sponsored a MRA that examined the risks of one pathogen in all water media. MRA for foods categories that are regulated by multiple agencies have required interagency collaborations.

A mathematical model is assumed to adequately represent complex biological phenomenon and ecological relationships.

Available data are assumed to be representative of the parameter. In practice, this assumption can lead to an overestimate or underestimate. Although MRA's routinely

incorporate uncertainty (and/or variability) about the parameters of interest, the underlying assumption is that data included in a risk model is representative of the pathogen and target population of interest. Many examples illustrate the ubiquitous nature of this assumption. Data from experiments using animal models are commonly used rather than data from human infections. Certain related bacteria are considered to be surrogates for the pathogen of interest. Data based on short-term exposures are extrapolated to model the chronic effects of prolonged exposures. Public health surveillance data is considered to be reflective of the actual disease burden experienced by the human population under surveillance. Surveys of foods reflect the true prevalence and distribution of pathogens in that food. Surveys that focus on individual's food consumption during a limited time period reflect the long-term consumption patterns of the individual, and the study population is representative of the population of interest.

It is assumed that it is appropriate to pool data derived from a variety of sources. This assumption can result in an overestimate or underestimate of true risk**.** Estimated prevalence and cell number distribution of pathogens is generally determined by combining results available in published literature, government's surveillance reports and industry reports. These data have inherent variability and uncertainty resulting from the variety of methods used to obtain them. It is assumed that pooling data from multiple sources (with or without weighting each observation with respect to a quality score) will provide a valid estimate of the parameter of interest with appropriate limits of uncertainty.

Normal distributions or triangle distributions are often assumed for parameters. Point estimate based on 50^{th}, 75^{th}, or 95^{th} percentile are sometimes assumed and may not be correct or appropriate.

Assumptions concerning the Agent:

It is generally assumed that a minimum unit of the microbiological hazard is necessary to cause disease. For example, a risk model could be based on an underlying assumption that a single cell of a pathogenic bacterium has the potential to cause disease. Alternatively, it could be assumed that some minimum concentration of a bacterial toxin is required to cause disease.

Most models assume that each microbial unit acts independently, and that a single bacterial cell or viral particle has the potential to cause disease.

It is usually assumed that the chance of contracting a disease once an individual is infected is independent of the ingested dose, i.e., once infected, a particular individual contracts the disease regardless of ingested dose. A higher dose of pathogen would not cause more severe symptoms.

It is often assumed that factors intrinsic to the pathogen are the primary determinant of the agent's ability to cause infection/disease. Extrinsic factors (related to both the environment and the potential host) also impact the affect the pathogen's

ability to cause infection/disease, and in some cases may be a more important determinant of the health outcome than the agent itself.

Certain assumptions must be stated regarding the variability among pathogen subtypes with respect to multiple characteristics that influence the development and/or detection of infection and/or disease. Many characteristics intrinsic to a specific pathogen ultimately influence both the probability that a specific subtype of a pathogen will result in an infection (or development of disease) and that this infection/disease will be detected and subsequently reported. These characteristics must be considered and prioritized according to their relative importance in impacting the health outcome of interest. In some cases, data is available that permits differentiation among subtypes with respect to these factors during the risk assessment process. For example, certain *Salmonella* serotypes appear to be more pathogenic to humans than others, and their relative pathogenicity could be defined to vary between serotypes in risk models. In other cases, these data are not available and it becomes necessary to assume that all subtypes are equal with respect to the defined characteristic. For example, certain *Salmonella* serotypes could be assigned the same relative pathogenicity in risk models when information that discriminates among the serotypes is unavailable. It is common to assume all isolates in the scenario are equally pathogenic.

Certain assumptions must be stated regarding the variability among pathogens and pathogen subtypes with respect to survival and growth in a variety of matrices. For food matrices in particular, many characteristics intrinsic to a specific pathogen ultimately influence the ability of the pathogen to survive and/or grow in that matrix. These characteristics must be considered and prioritized according to their relative importance in impacting the role of a given food as a vehicle. Assumptions concerning these characteristics are often necessary. For example, the pathogen may not be able to proliferate in the food, but it can survive and cause an adverse effect when consumed by a susceptible host. Some pathogens can form spores to ensure survival during adverse environmental condition and other may produce toxins under certain circumstances.

It is assumed that available growth kinetics models are adequately representative of all pathogen subtypes and are appropriate across different food matrices/environmental conditions. This assumption can lead to an overestimate or underestimate of risk.

Assumptions concerning the Host:

Certain assumptions are required with respect to the variability of susceptibility of individuals to the development of infection/disease. For example, it is frequently assumed that certain groups of people are more susceptible to infection/disease than others (i.e., young, elderly, pregnant woman, immunocompromised individuals).

Target population is assumed not to be vaccinated or immune due to previous exposures. The probability of infection or illness resulting from exposure is independent of previous exposures; also, the probability of infection or illness resulting from

secondary transmission is also independent of previous exposures. This ignores the possibility of temporary or permanent immunity.

Assumptions concerning the Environment:

Risk assessment models typically assume that microbes are homogeneously distributed throughout the specified matrix. While this may be a reasonable assumption for certain foods (e.g., milk, juices) and air, and water, it is unlikely to hold true for most food categories or soil matrices.

MRA models typically assume that the quantitative levels of contamination (i.e. microbiological counts) are best represented by a log normal distribution.

Assumptions are stated concerning the analytical methods used to detect the presence/quantitative level of the pathogen in a matrix. It is typically assumed that viable pathogen can be detected (e.g., culture, bioassay, serological test, polymerase chain reaction [PCR]). It is necessary to indicate the limit of detection, analytical specificity, epidemiologic sensitivity, and epidemiologic specificity of a described method when interpreting the results obtained when testing to identify a particular pathogen in a defined matrix. It is often assumed that all subtypes of a pathogen are equally likely to be detected using a particular analytical method; however, this may not be valid. Similarly, certain methods are applied across a variety of matrices. In some situations, characteristics of the matrix itself may interfere with pathogen detection.

Assumptions concerning the geographic and temporal (seasonal) distribution of pathogens are required. For example, it is believed that *E. coli* O157:H7 exhibits a seasonal distribution in cattle, ground beef, and ambient waters. You could assume that this pathogen is more prevalent during May – September than in October through March when modeling potential exposures or you could assume that prevalence is uniform throughout the year. You could make similar assumptions concerning the geographic distribution of a pathogen.

The complex series of environmental events that impact the survival and/or growth of microbiological hazards are typically simplified in the context of risk models. Considerable variability exists with respect to environmental conditions (i.e., time, temperature, pH) over time. For example, conditions associated with storage, refrigeration, product formulation, and batching process will vary greatly. Risk models assume that a single situation (or perhaps a limited number) occurs and that this factor occurs consistently over time.

Assumptions Concerning the Exposure Scenario:

The exposure time span of interest is usually specified in the scope. Whether the exposure time span is a specified event (e.g., meal, trip to the beach) or a lifetime it is still an assumption that must be transparent.

A specific food is the vehicle of transmission for a given pathogen. In reality, multiple foods may serve as a vehicle. Further, assumptions regarding the relative importance of potential exposures (i.e., food-borne; direct contact with animals, wildlife, insect vectors, or the environment; human-to-human transmission; water-borne exposures, recreational exposures) are necessary.

Certain assumptions are required with respect to individuals' exposures to pathogens of concern and the variability in exposures among individuals. These assumptions may lead to an underestimate or overestimate of risk. Consumer behaviors are diverse (e.g., by region, ethnicity, religion, food preparation, eating practices, packaging methods, manufacturing production practices, food production practices, local conditions, sanitation). Also, foods are not going to be consumed with the same frequency by the same people over an entire year. Factors such as seasonal availability of certain foods and changing eating habits may be appropriate to consider in a national scale MRA. It is not feasible or advisable to try to break out every possible behavior that may influence the exposure scenario. Often an average behavior is assumed to be representative at a population level. For example the number of servings consumed by each person or the number of contaminated servings.

Appendix B Hazard Identification Questions

This appendix contains examples of specific hazard identification questions that may be useful for the risk assessor's consideration. These are not all the questions risk assessors might consider.

General Questions:

1. What is the hazard in question and what is the specific media or food of concern? (e.g. *Campylobacter* spp. in poultry).
2. What are the common routes of exposure associated with the hazard?
3. Are there indicator organisms or surrogates that can allow for an indirect evaluation of this pathogen in the absence of data?
4. Which pathogens are of concern to public health? Which are regarded as being of *greatest* concern, and why?
5. Are any media closely associated with, or often linked to, specific illnesses?
6. What agents are present in the media that may cause adverse health effects?
7. Prioritize and tabulate all the pathogens in terms of their degree of severity.
8. How is the media linked to the illness associated with the pathogen (epidemiologic evidence? Laboratory evidence?)
9. Are there available epidemiological data and microbiological data to associate what type of pathogen is associated with the media of interest?
10. Are there adequate public health data to substantiate the occurrence of pathogenic microorganisms in the media in question?
11. Are there any established standards/guidelines regarding the pathogen of interest? (assuming there are answers)
12. Is there data relevant to support the hazard? scientific literature? databases from industry, government agencies, international organizations? expert opinion? clinical studies, epidemiological studies, surveillance studies, outbreaks (domestic and international)? laboratory animal studies, investigations of the characteristics of the food-borne pathogen? microbial ecological studies? microorganism's behavior studies? sensitive populations (high risk, elderly, prenatal)?
13. For food media, is there a list of generic processes for the normal exposure of the food product in question in the food chain? retail, restaurant, HACCP plan systems?

Questions concerning the Agent:

1. What type of pathogen is this organism (Bacteria, Virus, Parasite, Fungus, Prion, noninfectious toxin)?
2. What are other taxonomic/strain considerations that influence ability to cause disease? Does the pathogen have particular strains that differ in ability to cause disease? If so, what is the strain of interest?
3. What intrinsic properties influence this agent's ability to cause disease?
4. What is the "life cycle" of this agent?

5. Does the pathogen produce a toxin? If so, is it the toxin that presents the hazard, or the pathogen?
6. Are there indicator organisms or surrogate species that can allow for an indirect evaluation of this agent in the absence of data?
7. How is the pathogen identified?
8. What factors influence the spatial distribution of this agent (clumping, aggregation, particles, clustering)?
9. Is the pathogen an anaerobic, gram-positive, spore- forming rod that produces a toxin?
10. Is the pathogen a microaerophilic organism?
11. What are the biochemical/taxonomic characteristics to identify the pathogen?
12. What is the subtyping of the pathogen?
13. What are the survival characteristics? heat resistance? susceptibility to antimicrobial food additives? acid resistance? sensitivity to disinfectants or desiccation? sensitivity to radiation? UV? ionizing?
14. Are there phenotypic characteristics that influence virulence and/or pathogenicity?
15. Are there genotypic characteristics that influence virulence and/or pathogenicity?
16. Are the spores heat resistant? Can it survive in treatment processes?
17. Is there a microbiological testing/identification method for the pathogen in human clinical samples? Is there a specific method for testing for this pathogen?
18. What methods are available for detecting and quantifying the agent?
19. What are the cultural characteristics?
20. What are the detection methods to identify the food-borne pathogen?
21. What are the sampling and enrichment techniques to identify the food-borne pathogen?
22. What are the growth characteristics? free-living vs. obligate parasite? growth requirements? temperature? pH? water activity? oxygen? Relative humidity?
23. What is the main pathogenic strain of this hazard, and what other specie(s) within the identified genus are humans susceptible to? (e.g., *Campylobacter jejuni* and *coli*).
24. Are there phenotypic characteristics that influence this agent's virulence and/or pathogenicity?
25. Are there genotypic characteristics that influence this agent's virulence and/or pathogenicity?
26. How does the agent cause pathology and/or disease?
27. Does the microorganism produce a toxin while growing in the intestinal tract?
28. How infectious/toxigenic is the pathogen?
29. What is the incubation period?
30. What is the pathogenic microorganism's virulence?
31. What is the mechanism of action for infection and illness?
32. Is secondary transmission possible?
33. What is the target organ?
34. Can the pathogen replicate in the media of concern?
35. Is the pathogen's ability to cause disease restricted to specific strains with identifiable virulence characteristics?

36. What are the virulence-associated characteristics to identify the pathogen?
37. What is the pathogenicity? dis. char. and diagnosis? sequelae? host range? infectious dose? subpopulations at risk? animal models?
38. Describe the contagiousness of the infection (is or is not)
39. Identify the degree of severity for each pathogen

Questions concerning the Host:

1. Who is the susceptible population and what makes them susceptible? (e.g. children, adults, pregnant women)
2. What types of hosts can be infected by this agent? Is there host specificity?
3. Are any practices/behaviors closely associated with, or often linked to, specific illnesses?
4. Is illness host-specific, i.e. more likely to affect susceptible populations?
5. Are there subpopulations at increased risk for this pathogen?
6. What are the demographics associated with this pathogen?
7. What are the general socioeconomic trends associated with this pathogen?
8. What is/are the characteristic(s) of the infection caused by the above hazard? (e.g. gastroenteritis).
9. What is the nature of identified cases of infection? (e.g., sporadic, small/family related outbreaks).
10. What types of pathology and/or diseases are caused by this agent?
11. What are the typical symptoms of illness, and how do these differ between different pathogens?
12. What are the symptoms?
13. Is the microbe presenting itself as a gastrointestinal symptom? Vomiting, watery diarrhea? inflammatory diarrhea?
14. Does the pathogen produce all or any of the following symptoms: abdominal cramps, nausea, vomiting, diarrhea, fever, dehydration?
15. Does the microbe usually have no manifestations of gastrointestinal illnesses associations? neurologic? systemic? hepatitic?
16. What is the incubation period within susceptible host, before the onset on an infection?
17. Is the pathogen found in the large intestinal tract only?
18. Does it shed in the feces?
19. What is the associated morbidity/mortality?
20. How might one define immunocompromised?
    a) Are there specific quantifiable biomarkers of immunocompetency?
    b) Can one define such biomarkers?
21. How would one account for genetic/ethnic/cultural differences within or between populations?
22. What is the definition of biomarkers for malnutrition?
23. What are the age groupings? How do they relate to susceptibility?
24. Should age be defined with respect to chronology or physiology?
25. What would one do if the definitions [for different biomarkers] overlap?
26. What are the definitions for chronic ailments?

27. What are multiple concurrent factors?
28. How is immune status defined?

Questions concerning the Environment:

1. What are the environmental conditions of the pathogen in question: contamination, growth, inactivation, elimination, survival, for each part of the exposure scenario?
2. Does the pathogen have any specific survival characteristics that may promote its survival through the exposure scenario?
3. How do various environmental conditions (temperature, nutrients, pH) influence this agent's growth, survival, and/or death?
4. Are there certain environmental conditions and/or control processes that this agent can survive (or develop resistance)?
5. Are there seasonal, geographic or climatic factors that affect the occurrence of this agent?
6. What are the time factors involved in identifying the pathogen?
7. What are the temperature factors involved in identifying the pathogen?
8. What is the geographical range and seasonality of the pathogen?
9. Is the organism found in warmer months?
10. What is the geographic location?
11. What season does it occur?
12. If carried by another species, what is the geographic range of that species?
13. Is there a reservoir for the pathogen?
14. What are the ways in which the pathogen can contaminate the media?
15. Is that pathogen zoonotic?
16. Where (in what media?) is the pathogen commonly found?
17. Is the pathogen endemic?
18. Has it been identified in other countries recently?
19. What is the reservoir and/or environmental niches for this agent? Are there other important ecologic factors that influence this agent?
20. How can the media become contaminated with this pathogen? Are there any specific practices/behaviors that could promote survival of this pathogen?
21. For food, did contamination occur during growing, harvesting, processing, storing, shipping, or final preparation?
22. Does the pathogen grow in the media?
23. What is the route of contamination and location of the pathogen in the media?
24. What is the microbial ecology to identify the pathogen?
25. What are the technologies associated with identifying the pathogen?
26. What are the consumer trends associated with identifying the pathogen?
27. For food, what is the effect of food processing and preparation on their survival?
28. What is the consumption patterns associated with this pathogen?
29. What are the marketing and preparation practices associated with this food-borne pathogen?
30. What are the globalization trends associated with this pathogen regarding the media of interest?

31. Is the pathogen's presence in contaminated media the result of an error or breakdown in normal controls?
32. What are the consumer behavior and consumption practices associated with this pathogen and media?
33. For food, identify and list (document) all the associated places (poultry (chicken, turkey), slaughter house (bovine, swine) , ready to eat, …), locations/residing critical control points, and the time that certain pathogens are present.
34. What is the principal reservoir of this hazard, and where is it also commonly found?
35. What are common environmental media and the expected concentration of this agent?
36. Is the pathogen found in humans or animals or both?
37. Where is the source found? humans? domestic wild animals? poultry/eggs? swine? cattle? rodents? pets such as turtles, chicks, dogs and cats.
38. Is the pathogen found in cat, rodent or bird feces?
39. Is the food-borne pathogen found in soil? dust? sewage? and intestinal tracts of animals and/or humans?
40. What is the pathogen's reservoir in nature?
41. Is the pathogen found in the intestinal tract?
42. Identify and list all the potential sources (reservoir) for all of these pathogens.

Questions concerning Transmission:

1. What is the mode of transmission? (e.g., direct contact with contaminated animals/carcasses, ingestion of contaminated food or water).
2. Identify all the potential routes/pattern of transmission (direct or indirect) of microbial infection (disease) for each pathogen
3. Is the pathogen found in putting something in the mouth that has been contaminated with the stool of an infected person or animal; direct contact with the droppings of infected animals?
4. Is the pathogen a person-to-person transmission such as in child daycare settings?
5. For food media, what are the eating habits associated with the pathogen?
6. How does this agent infect a susceptible host? What are the typical routes of infection and/or portals of entry?
7. Is there potential for secondary spread?
8. What is the infectious dose?