This is a "Post-Print" accepted manuscript, which has been published in "International Journal of Food Microbiology".

Please cite this publication as follows:

Havelaar, A.H., Brul, S., De Jong, A., De Jonge, R., Zwietering, M.H., Ter Kuile, B.H. (2010) Future challenges to microbial food safety. International Journal of Food Microbiology 139 (Suppl 1), S79-S94.

You can download the published version at:
http://dx.doi.org/10.1016/j.ijfoodmicro.2009.10.015

Future Challenges to Microbial Food Safety

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Abstract

Despite significant efforts by all parties involved, there is still a considerable burden of foodborne illness, in which microorganisms play a prominent role. Microbes can enter the food chain at different steps, are highly versatile and can adapt to the environment allowing survival, growth and production of toxic compounds. This sets them apart from chemical agents and thus their study from food toxicology. We summarize the discussions of a conference organized by the Dutch Food and Consumer Products Safety Authority and the European Food Safety Authority. The goal of the conference was to discuss new challenges to food safety that are caused by microorganisms as well as strategies and methodologies to counter these. Management of food safety is based on generally accepted principles of Hazard Analysis Critical Control Points and of Good Manufacturing Practices. However, a more pro-active, science-based approach is required, starting with the ability to predict where problems might arise by applying the risk analysis framework.

Developments that may influence food safety in future occur on different scales (from global to molecular) and in different time frames (from decades to less than a minute). This necessitates development of new risk assessment approaches, taking the impact of different drivers of change into account. We provide an overview of drivers that may affect food safety and their potential impact on foodborne pathogens and human disease risks. We conclude that many drivers may result in increased food safety risks, requiring active governmental policy setting and anticipation by food industries whereas other drivers may decrease food safety risks.

Monitoring of contamination in the food chain, combined with surveillance of human illness and epidemiological investigations of outbreaks and sporadic cases continue to be important sources of information. New approaches in human illness surveillance include the use of

molecular markers for improved outbreak detection and source attribution, sero-epidemiology and disease burden estimation.

Current developments in molecular techniques make it possible to rapidly assemble information on the genome of various isolates of microbial species of concern. Such information can be used to develop new tracking and tracing methods, and to investigate the behavior of microorganisms under environmentally relevant stress conditions. These novel tools and insight need to be applied to objectives for food safety strategies, as well as to models that predict microbial behavior. In addition, the increasing complexity of the global food systems necessitates improved communication between all parties involved: scientists, risk assessors and risk managers, as well as consumers.

Keywords: food safety, microbial hazards, future trends, monitoring, surveillance, risk assessment, tracking and tracing, communication

1. Introduction

The microbiological aspects of food safety have been studied intensively for many decades. In the Netherlands, the standard of food safety has increased in the last decades (Van Kreijl *et al.*, 2006), and political attention is shifting to other food-related problems such as obesity and unhealthy diets. However, even in industrialized countries, there is still a considerable burden of foodborne illness. For example, in the Netherlands there are an estimated 700.000 cases of illness and 80 deaths per year. The burden of foodborne disease for this country is at least 3800 Disability Adjusted Life Years and 65 million Euro per year (Havelaar *et al.*, 2008). Also other industrialized countries report a continuing burden of foodborne illness (Flint *et al.*, 2005, Hall *et al.*, 2005, Adak *et al.*, 2005, Anonymous 2007a, Jones *et al.*, 2007). Foodborne outbreaks appear to be on the rise again in some industrialized countries, with a shift from traditional problems with foods from animal origin to fresh foods such as produce (Anonymous, 2008a), shellfish (Pontrelli *et al.*, 2008) and dry products and ingredients (e.g. peanuts, Anonymous, 2009). Furthermore, new threats continue to be identified. The attention for viruses is more recent, but no less relevant. New risks are being encountered because of changing characteristics of the relevant micro-organisms, changing production methodologies, changes in the environment and the ecology, and an increase of the global trade of foodstuffs. In addition, demands on food safety increase steadily. Due to the nature of microbes and our food chain, measures to ensure food safety have to be implemented on a global scale, necessitating a global approach.

To discuss the challenges that microbes pose to food safety on the longer term, the Dutch Food and Consumer Products Safety Authority (VWA) and the European Food Safety Authority (EFSA) organized a conference on “Future Challenges to Food Safety” (Wolfheze, the Netherlands, 9-12 June 2008). The goal of the conference was to discuss new challenges to food safety that are caused by microbes and strategies and methodologies to counter these.

It was aimed at achieving conceptual breakthroughs through an imaginative combination of recent developments in microbiology, epidemiology, mathematical modeling and expert knowledge; using these to propose new approaches to analyze and control food safety issues in the future. Such tools should enable risk assessors to pro-actively address imminent problems before they cause harm. This paper provides an overview of the major themes identified during the conference. The paper starts with a discussion of food safety from the risk management perspective. A systems approach to identify and structure future developments that may help to develop a pro-active approach to food safety is introduced, followed by a more detailed discussion of relevant developments with special attention for the interaction between micro-organisms and their environment, and for microbial evolution. Despite the need for pro-active approaches, surveillance and monitoring are discussed as important cornerstones of food safety policy and new developments in the available methodology are discussed. Finally, needs related to communication between all actors in the food chain are outlined.

2. Risk management

Food safety demands

Management of microbial food safety is a balancing act involving disparate factors. A high level of safety can be achieved by rigorously heat-sterilizing the food, thereby destroying taste and nutritious value. Irradiation would be another method for virtually absolute control of microbial risks, but in addition to being expensive, it is not acceptable to the public at large in many countries. Furthermore, some bacterial and fungal toxins are not inactivated by currently used irradiation doses. The consumer demands fresh, tasty, healthy and wholesome food products. Nevertheless, safety is in this framework considered an absolute requirement; placing unsafe food on the market is not an option in the consumer's mind. Food laws everywhere are very clear on this point. For example, the EU General Food Law (Anonymous, 2002) states that: “a high level of protection of human life and health should be assured in the pursuit of Community policies”. Still, placing chicken contaminated with *Salmonella* or *Campylobacter* on the market is tolerated, because the consumer can circumvent this risk by cooking the meat properly and taking adequate precautions against cross-contamination, illustrating that responsibility for food safety is distributed over the entire chain. Nevertheless, there is an increasing pressure on producers to reduce contamination levels of fresh meat as far as possible and economically feasible. It has been demonstrated that it is very difficult to modify consumer behavior by education campaigns (Nauta *et al.,* 2008).

Microbial food safety differs fundamentally from chemical food safety. While chemical residues and additives typically enter the food chain at more or less predictable steps, microbes can enter at any step. They grow and die and interact with the food in ways that are at best empirically described, but less understood in detail. The effects are also of a different nature. Chemical contaminants, such as dioxins, can accumulate in the human body over the years and still exert influence long after ingestion. Microbial pathogens can in some cases be dormant for a certain time, but usually cause disease in a matter of days or weeks. The public perception of microbial and chemical risks is also different. Residues of pesticides cause public outcries if they exceed the norms, but usually will not have any noticeable detrimental effect, while foodborne microbial and viral diseases are generally more accepted as facts of life, as long as death or permanent harm do not occur (Hansen *et al.*, 2003)

One of the challenges for managers of microbial food safety risks is to put in place effective controls, without unnecessarily increasing costs or reducing taste and nutritional value. Microbial hazards can be introduced at any step in the production chain and the most effective opportunity for controlling those hazards can very well be a different step. Microbial risk management therefore requires a thorough understanding of the entire food production chain. Monitoring the presence of pathogens in the end product usually is an inefficient approach to hazard control, because it is impossible to test sufficient samples to obtain the necessary degree of statistical power to detect contaminants at levels that may create unacceptable health risks. Furthermore, by the time the potential presence of pathogens has been confirmed, the optimal moment to take measures may have passed. Therefore, a pro-active approach is required, starting with the producer ensuring a safe product and process design, and predicting where problems might arise, rather than detecting them after they have occurred.

At present HACCP (Hazard Analysis Critical Control Point) programs and GMP (good manufacturing practice) are mainly used to manage microbial hazards in foods. While these systems have proven very effective for the control of food safety (Van Der Spiegel *et al.*, 2004, Arvanitoyannis and Traikou, 2005), it must be realized that they are designed on the basis of known hazards, and do not necessarily take potential future developments in consideration. For innovations, new validations and verifications are necessary. Furthermore, it should be realized that the implications of microbial adaptability are not sufficiently taken into account (McMeekin and Ross, 2002, McMeekin *et al.*, 2006). Although documentation is a very important aspect of HACCP and GMP procedures, the real confidence in control comes for the validity of the effectiveness of the written guidelines and the adherence to them. Producers and handlers of foodstuffs are more likely to adhere to the prescribed HACCP procedures if they recognize these as useful and implementable (Taylor and Taylor, 2004).

Science-based risk-management

Within the food safety discipline the terms “risk manager” and “risk management” are not unequivocal and are used to indicate several functions and persons. As a consequence there are several persons with responsibility as risk managers, each at a different step of the food chain. Formally “the” risk manager is the government’s minister of public health and/or his/her colleague of agriculture, as they set the standards to which food producers must adhere. Furthermore, within government supervisory agencies the persons in charge of the enforcement branch are often called risk managers. Often, however, the person responsible for compliance to procedures within a food manufacturing company is called a “risk manager” as well, as this term in fact very well describes his/her day-to-day activities. Although these functions are less clearly defined in smaller operations and in primary production, the role should be fulfilled in any food operation. Risk assessors, on the other hand, have a different function. They provide the risk manager with science-based advice on the magnitude of risks and cost-effective ways to reduce these. This advice enables the different risk managers to take decisions on measures to control risks, by setting standards, by implementing in-plant control measures and/or enforce existing regulations.

A challenge to food microbiologists in their role of risk assessors is to translate complex scientific problems in such a way as to help a risk manager to make a simple yes/no decision. The risk manager wants to know what standards to set, when to interfere in the production process, prevent a batch from reaching the market or take another measure. Under the WTO Agreements, and in particular the Agreement on the Application of Sanitary and Phytosanitary Measures, considerations of food safety and animal and plant health are the only legitimate reason for trade restrictions. The type of decisions in these cases is also of the yes/no kind. The underlying science will have to provide the decision maker with a solid rationale that will stand up in the international courts. This implies that data on the microbiological status of the foods concerned must be communicated in terms of public health risk with a limited margin of error, requiring a predictive power from food microbiology. As zero-risk is unattainable, this approach also implies that the risk manager defines a level of acceptable (tolerable) risk. In international trade, this is called the Appropriate Level of Protection, equivalent to the currently realized risk level under the food safety system of the importing country. Food safety managers may wish to achieve a higher level of protection in future by stating additional public health targets (Anonymous 2006, Anonymous 2007b).

Food safety risk-management in the EU

Public concern about food safety increased sharply as a result of the food scandals in the last decade of the twentieth century and confidence decreased in parallel. To counter these sentiments national governments and the EU established food laws and regulations that strictly separate risk management from risk assessment. The idea behind this was to create transparency by having the risk assessor provide advice to the risk manager completely in the open. The risk assessor operates independently, based on the best available science and free from influence by politics, industry or any other stakeholder. The risk assessor should give objective advice, based on science while taking account of other considerations that the risk manager has indicated. The risk manager can then incorporate other factors such as public concern or political preferences into the decision making process.

The exact procedures for risk assessment differ considerably in the different member states. The EU itself has mandated the European Food Safety Authority (EFSA) to carry out risk assessments, either at the request of the Commission, a member state or the European Parliament, or on its own initiative. The EFSA in turn has handed this task to 10 scientific panels and the Scientific Committee, which it finances and supports scientifically and otherwise. These panels write opinions which are passed on unchanged to the EU Commission and published on the EFSA website. When strong public interest is expected a press release is issued as well. The panels are comprised of scientists who operate independently from risk management, and almost all come from EU-member states, though this is not a formal requirement. The scientists are chosen in an EU-wide application procedure on the basis of their expertise and prominence in their scientific fields, and are appointed for a period of three years at a time.

The advice of EFSA's panels is presented as a scientific opinion to assist in the formulation of risk management measures by the European Commission. Measures may be decided upon by the members states in a consensus procedure, allowing political, economic and other considerations to influence the decision making process.

Interaction between scientists and regulators

Researchers, in their role as risk assessors, need to provide the regulators, in their role as risk managers, with advice that can be implemented in a practical manner. Regulators may or may not have a scientific background and thus results of risk assessments have to be communicated in a way that can be understood by fully informed laypersons. The process of risk analysis, as defined by Codex Alimentarius, consists of risk assessment, risk management and risk communication. The very first step in the process, hazard identification, is a component of risk assessment. The hazard identification can come from any source. In practice it is often the risk manager, who "identifies" the risk by asking for a risk assessment. For a good risk assessment, the problem at stake needs to be well understood by the assessor. Therefore, correct phrasing of the request is essential for a successful risk assessment procedure. The regulator needs to articulate his question so that it will not be misunderstood by the scientist. In short, they need to "speak each others language". This seems a trivial point, but experience proves that it is not.

The roles of the risk assessor and the risk manager need not only to be formally separated, but also with respect to substance. A risk manager has to consider more issues than science only, such as stakeholder interests, public concerns and political pressure. The risk assessor needs to present a science-based advice, but may anticipate the risk manager to take other than scientific factors into account when selecting the risk management options.

3. A systems approach to analyzing future challenges to food safety

In addressing the present or future state of food safety one should investigate effects both on large scales of space and time as well as on small scales (Table 1). Certain aspects slowly change at a global scale, like climate change while others such as point mutations or the acquisition of a plasmid by a micro-organism, occur on a molecular scale and on very short time scales. In addition, certain aspects occur over longer time scales, but on a molecular level, like subsequent adaptation of micro-organisms, or on a small time scale but on a larger spatial scale, like the spread of a virulent micro-organism due to the large traffic of people or goods. Furthermore these changes occur in micro-organisms and humans, in habitats and in the environment. To accurately describe and predict processes in all these different organisms and locations, on these very different spatial scales and time scales is virtually impossible, the more because all these aspects interact. The risk framework of the UK Foresight project on infectious diseases (Tait *et al.*, 2006) offers a useful starting point for the development of scenario analysis in relation to food safety. The project has developed the following definitions, which are cited literally here:

- Disease sources/emerging hazards: phenomena or biological events that: give rise to potential new diseases; enable existing diseases to become more harmful; enable existing diseases to infect new hosts; or enable existing diseases to spread to new areas.
- Pathways: mechanisms or routes by which a disease organism can transfer from one host to another, within or between species.

- Drivers: social, economic or physical factors that affect disease outcomes by changing the behavior of disease sources or pathways.
- Outcomes: diseases of plants and animals at the individual, community and ecosystem or farming system level, and diseases of humans at individual and societal levels.

A basic risk framework (Figure 1) shows the links between these different factors.

The PERIAPT project on emerging risks (Noteborn *et al.*, 2005) identified 8 major categories of drivers, based on expert surveys. Later, we will present a tabular representation of these drivers of change in the food system, and a qualitative analysis how they affect sources, pathways and outcomes. To better explore these complex interrelationships among different drivers, mathematical models may be helpful.

Mathematical models are a representation of the essential aspects of an existing system (or a system to be constructed), presenting knowledge of that system in a usable form. A mathematical model usually describes a system by a set of numerical variables and a set of equations that establish relationships between the variables. The variables represent some properties of the system, obtained by measurements or by expert opinion. The actual model is the set of functions that describe the relations between the different variables. The purpose of modeling is to increase our understanding of the world. The usefulness of a model rests not only on its fit to empirical observations, but also on its ability to extrapolate to situations or data beyond those originally described in the model (from Wikipedia, March 21, 2008).

Some examples of models that are currently used to analyze and to support decision making on food safety:

- Microbial risk assessment (MRA) models (hazard identification, exposure assessment, hazard characterization (including dose-response), risk characterization).

  Used to understand the relationships of pathogen occurrence (both prevalence and *concentration) in different steps of the food chain, to predict health risks associated with pathogens in food, and the expected public health effects of interventions and the setting of risk-based standards for food production.*

- Predictive microbiology (growth/death/survival).
  Used to understand the growth or death of micro-organisms in relation to their implicit properties and interactions, and the intrinsic properties of the food and the extrinsic factors of the (processing) environment. It is an important component of MRA models, and also used for prediction of shelf-life and intrinsic safety of foods.
- Dynamic infectious disease models.
  Used to understand the spread of diseases in human or animal populations, depending on contact patterns and mode of spread of the pathogens, in relation to the development of protective immunity.
- Risk factor models (analytical epidemiology).
  Used to relate the observed occurrence of responses (e.g. illness) to the occurrence of potential predictive factors.
- Attribution models.
  Used to estimate the contribution of putative sources to the observed occurrence of responses (e.g. illness).
- Multi-criteria analysis models.
  Used to support decision makers in making evaluations of different options, based on a combination of variables of a different nature (health, economic, societal, ….) with value-based weights.

All models are based on a set of (simplifying) assumptions and have specific data needs. They also differ in their form (linear or non-linear, deterministic vs. stochastic, static vs. dynamic) which impacts on their use for specific purposes. It is noted that all models described above work at relatively low levels of aggregation, looking at specific (parts of) food chains in relation to public health effects. The challenge is to develop models that are able to capture the impact of different drivers on foodborne risks at higher levels of aggregation.

? The overall aim of this coupling of various aggregation levels is to better understand how drivers affect the evolution of foodborne illness and to determine the most important drivers. This can help to react with a timely and adequate response, and support a pro-active approach, targeted at future events. Generally one needs to decrease complexity in the more detailed parts and move upwards to more global aspects, but also *vice-versa*, since after identifying certain important aspects on a global scale one might need to change focus to the more detailed level. For this, intensified linking to models in other domains is necessary, like biosphere models, geospatial modeling, catastrophe modeling, climate models, remote sensing, network science, statistical physics, and data mining.

For these models, a huge amount of data is needed such as information of food production sites and global product flows (volume, origin) for high risk products (e.g. fresh meat, fresh produce, shellfish), risk maps, global atlases of food consumption and production, food categories with different levels of risk. Connecting them in dynamic systems can help to identify rates of change. However, it is necessary to begin with low granularity, with more detail based on sensitivity analysis.

In current risk evaluations considerable uncertainties already prevail, so, when modeling future risks even larger uncertainties will exist. However, important insights can still be gained using the available information, making the best informed decisions at a given point in time, that later can be detailed if more and better information becomes available. Scenario analysis will be particularly important to better understand the impact of different factors, their interrelatedness and their uncertainties. It is a process of analyzing possible future events by considering alternative possible future developments and outcomes (scenarios), and their likelihood. These insights can then be used to develop a range of contingency plans to address the most likely or most serious scenarios. The analysis is designed to allow improved decision-making by providing more complete consideration of outcomes and their implications. Typically, scenario analysis starts with the identification of possible important drivers of change, and subsequently assigning a preliminary ranking of their importance.

4 Trends and future developments

In this section, we present an attempt to collect and structure available information on the current and future aspects of microbial food safety. Table 2 gives an overview of factors identified so far, according to the model presented by Tait *et al.*, (2006). We interpret the sources category as referring to the pathogens; the pathways category is split into the three major stages of the farm-to-fork pathway (farm, processing and consumption), and outcomes are defined at the public health level. Note that there are complex interrelationships between different drivers, sources, pathways and outcomes that are difficult to visualize in a two-dimensional table, and hence the information must be interpreted with care. Nevertheless, important insights can be gleaned from this analysis, which provides a general background against which specific situations can be analyzed. Currently, it is only possible to discuss the impact of drivers of change on sources and outcomes in general, qualitative terms and so an attempt has been made to indicate the anticipated direction of change for specific sources and overall for drivers. It is clear from Table 2 that many drivers are expected to result in an increased risk to food safety, although there are also favorable exceptions, such as the lesser consumption of meat due to higher food prices. Controlling such threats is a challenge to governments as the only drivers assumed to result in reduced threats relate to government and policies. A second important challenge is to food industries that can modulate the effects of many drivers in such a way that a neutral or even positive effect on food safety can be expected (see drivers under science, technology and industry).

Further elaboration of the crude framework sketched in this chapter requires considerable inputs. It is suggested that to further develop the framework, historical examples be analyzed. This will provide more detailed insight regarding relevant drivers and sources, and their interaction as well as data to validate the approach. Such examples include the BSE epidemic, different meatborne zoonoses (e.g. VTEC O157), shellfish poisoning and more recent outbreaks in fresh produce.

In order to improve future responsiveness, signals about changes or breakdowns in the food safety system must be received and processed in time. This implies a pro-active approach. Process or production failures, including fraud and terrorist action need specific attention. Risk mapping (on a global scale) can be a helpful tool. This includes observation and systematically analyzing consumption patterns, processing / production changes, knowledge of international production chains (trade), and data & knowledge sharing. Where necessary, available data can be supplemented with expert opinions (global and multidisciplinary).

Trends in food processing

The trend for mildly preserved foods comes with a range of approaches being investigated, mostly using minimal heating, natural preservatives and non-thermal treatment as technologies and often combined preservation/hurdle technology as the principle in designing the overall treatment. Such processes need to be well controlled through adequate product and process design and proper implementation and monitoring through HACCP. This places a responsibility on industry, including small enterprises. There are more weak links, and overall the processes are less robust and more accident prone. As the scale of operation of food businesses continues to increase, errors may have a bigger impact.

Consumers need to be aware of the criticality of the formulations and of the need to treat manufactured foods either as perishable products needing proper refrigeration or requiring specific conditions of preparation (for instance non-Ready-To-Eat products that need to be cooked properly before consumption even though they may appear to be cooked). The concern is that many consumers do not habitually read labels and are unaware of shelf-lives and preparation requirements.

Can these new product types offer niches for concurrent, old, emerging or new microbial hazards? Classical examples are refrigeration and the niche created for *Yersinia* and *Listeria*, and for sporeformers by non-thermal treatments at the pasteurization level. As in many of these foods spoilage organisms have been removed or suppressed, there is increased opportunity for the growth of pathogens that recontaminate treated products – in the absence of the "normal" spoilage signal. Noroviruses show prolonged survival during cold storage and even freezing. The probability and extent of survival and of post-process contamination, rather than pathogen growth opportunity, may then determine the level of consumer risk.

While there would be a benefit from (and indeed a need for) for irradiation technology for certain applications (e.g. it is "safe", "invisible", no microbial issues of resistance, or of recontamination when done in-pack), consumer concerns towards the technology itself and about misuse to make spoiled food marketable, prohibit its wider use in practice. This implies considerable communication challenges, should the technology prove to be the treatment most relevant for certain applications.

While reducing packaging is a laudable initiative where the packaging is for "cosmetic" or bulk-transport purposes, industry and consumers should be aware of those situations where the packaging has a preservative function by minimizing growth and/or recontamination of microorganisms.

Consumer behavior

Several changes in food composition related to consumer health are foreseen. These may also have an impact on bacterial growth. For instance, components like salt and sugar are often used to inhibit the growth of organisms, both by their water activity lowering effect, and additionally solute specific effects. Their concentration cannot be safely reduced for health or other non-safety reasons without adapting the product design. On the other hand, fat can be seen as a vehicle for better stomach survival of pathogens, so less fat might reduce risks. Lower fat may also increase the water activity of a product by having more diluted solutes in the aqueous phase (Senhaji, 1977).

Considering the present increased level of general health of the population and better medical treatments, proportionally more elderly will be present in society. These elderly often are more vulnerable to foodborne diseases partly, as a result of a weakened immune system increasing the risks of complications and even death. Other defence systems, such as stomach acid may be impaired (achlorhydria), augmented by medication.

Due to changes in eating habits, certain risks might change in magnitude. For example an increased consumption of fish for health benefits may result in increased microbial risks. The increasing trend for fresh, pre-packaged produce or other foods that are consumed without additional heating by consumers also increases consumer risk. Exotic and ethnic foods are now trendy in the market, but do we understand the underlying preservation system? When we change these products (adapted for new markets; altered ingredients), are we clear on how this may affect safety? More and more animal species are used for food production and there is little knowledge about zoonotic risks of such foods (e.g. reptile meat, Magnino *et al*., 2009). New culinary techniques, such as molecular gastronomy involve more and more technical creativity and exotic ingredients to improve quality and consumer acceptance, which may result in unexpected risks. In the case of small restaurants, catering establishments and street vendors the scale of production is small and often, knowledge and sufficient technology may be lacking, resulting in avoidable errors which may lead to serious consequences for consumers.

Improving animal welfare (e.g. by increased outdoor access) and organic food production may lead to the re-introduction of pathogens with wildlife reservoirs such as *Trichinella spiralis* and *Toxoplasma gondii* and increase the prevalence of other hazards such as *Campylobacter* spp. but may reduce the prevalence of others such as *Salmonella* spp. (Gebreyes et al., 2008). Reduced usage of antimicrobial agents may have a positive impact on resistance development (Van der Giessen *et al*., 2007; Hoogenboom *et al*., 2008). Trends towards continuously increasing herd size in intensive bio-husbandry may also lead to increased public health risks by increased numbers of contacts between food animals, including purchasing of animals from more suppliers. The potential for infections to spread increases with herd size, and if such farms are concentrated in particular regions, there is also an increased risk of spread between farms. On the other hand, establishment of larger, newly designed farms may improve conditions for biosecurity and farm management, potentially reducing zoonotic risks (Kornalijnslijper *et al*., 2008).

Price

It is well known that cost is a very important consideration for the consumer when selecting foods, and that profits on food products are generally quite low. Both aspects make it difficult to be critical towards food safety and furthermore might occasionally result in fraud. Also, costs and conservatism may lead to resistance against implementing reasonable interventions or to interpret existing regulations liberally (e.g. use of approval of sick animals for consumption). Such non-compliance may increase consumer risk. The Law Enforcement Department of the Netherlands Food and Consumer Safety Authority experienced during the 2008-9 economic downturn that food producers and retailers more frequently violate regulations relating to cleaning and maintenance due to cost cutting (J. van der Kooij, personal communication). Increasing food prices are likely to compromise food security (in terms of food availability) on a global scale. For industrialized countries, food security is less at risk, but consumers may choose less costly alternatives. This may lead to less consumption of animal proteins (also driven by animal welfare and environmental considerations), leading to other health-related issues. Higher food prices may cause consumers to use food more frequently past its shelf life, and may increase recycling of food.

Global aspects

Due to the increase of international travel, organisms can be spread easily and quickly over the globe, and people come in contact with organisms and specific strains to which they have not been exposed earlier, increasing the risk of illness. Global trade will result in a longer transit distances and durations in the food chain, possibly increasing risk. Furthermore, complex food chains with stakeholders in many different countries will make the management of safety more difficult, especially at the initial stages of the food chain, the primary production as it consists of many small farms and is increasingly global in nature. On the other hand more powerful stakeholders (trade companies, supermarkets) will have the intention to influence these complex chains in order to guarantee food safety. Several large retailers that operate internationally have organized the GlobalG.A.P. quality control system (www.globalgap.org) that aims to supervise the primary production process of all agricultural products. Sourcing food from various climate areas means more variation in hazards. Border controls are effective in regard to the control of only a small proportion of imported foods and are less effective than hygiene controls imposed in the country of origin. Furthermore, global food chains may be more vulnerable to terrorist attacks.

More and more harmonization will occur in international regulation of food safety by the activities of e.g. the Codex Alimentarius Commission and the European Union. As a positive effect, fairness in trade will increase but on the other hand the same level of contamination in food may result in very different health risks in different parts of the world, due to differences in e.g. demography, immune status, food preparation and consumption habits, and relevance of various routes of infection.

Climate change

Climate change is considered to be one of the greatest current challenges to mankind, affecting all sectors of society, including nutrition, food security and food safety (Anonymous, 2008b). Due to climate change various risks may change, as a result of changed ecological conditions on various places on the earth. Changing ecology is expected to affect the distribution of plant and animal diseases. Water shortages may lead to limited quantities or quality problems with irrigation water, process water or ingredient water. This may lead to shifts in production areas and cultured crops, as well as an increased use of agrochemicals. This trend may be increased by competition for land-use, e.g. for biofuels or for settlements. Flooding may lead to increased contamination of crops in the field, or increased exposure of food animals to zoonotic agents. Control of cold chains may be impeded by rising ambient temperatures. Humidity may increase production of mycotoxins, whereas certain foodborne pathogens may thrive better under warm conditions. The mechanisms by which climate change affect food safety are highly complex and interrelated with many other societal factors, The outcome of these changes strongly depends on the adequateness of societal responses, both of a technical and a political nature.

Science

More and more public health risks, physiological and ecological traits of foodborne pathogens, routes of contamination, effects of interventions, will be investigated and quantified, making it possible to better balance risks, and evaluate optimal interventions to control risks to an appropriate level. That is, science will have a greater role in setting criteria both nationally and in international trade agreements.

The genomics revolution will facilitate easier and faster detection and identification methods, and can in particular lead to a better mechanistic understanding of the behavior of micro-organisms, both their physiology as well as their ecology. Furthermore new pathogens can be uncovered, for example better detection of injured and thus less easily culturable organisms is possible and more advanced methods to investigate cases and outbreaks become available.

Antimicrobial resistance

Usage of antimicrobial agents, both in the agricultural sector and in human health care settings, contributes to the emergence of resistant microbes. While resistance in the agricultural sector might be considered an economic problem with limited other consequences, it is developing into a cause of growing concern for human health care (see Newell et al., in an accompanying paper in this Special Issue). The overall use of antimicrobial agents in food animals is high and in certain countries largely exceeds the human use. The ban on usage of antimicrobials as growth promotors has, in some countries, barely had an influence, as it has been replaced by increased use for therapeutic purposes (Mevius and Van Pelt, 2006). Micro-organisms have an immense diversity and can easily transfer genetic information, making the emergence of new hazards, and adaptation to previously effective intervention methods possible.

At present, it is not clear what proportion of resistance encountered in human pathogens originates from selection in and transfer from animal reservoirs. It is not known if measures to reduce usage in the agricultural setting will lead to a rapid reduction of resistance, as this can also contribute in other ways to overall fitness of the micro-organism. The benefits to human health care of such measures are not quantified, as resistance may also develop due to other factors. In spite of these uncertainties, the development of antimicrobial resistance in agricultural settings is a cause of growing concern to public health and prudent use is advocated.

5. Epidemiology and surveillance

Monitoring of contamination in the food chain, combined with surveillance of human illness and epidemiological investigations of outbreaks and sporadic cases continue to be important sources of information to evaluate the success of current food safety management systems and to identify new hazards. Surveillance is defined as “the ongoing and systematic collection, analysis, and interpretation of data about a disease or health condition; used in planning, implementing, and evaluating public health programs” (Anonymous, 2000a). Surveillance can be aimed at outbreaks or sporadic cases of foodborne disease, and continues to be a cornerstone of food safety management (see the accompanying paper by Tauxe *et al.*, in this issue).

Outbreak surveillance primarily aims to stop the outbreak by identifying incriminated products and taking them from the market. Furthermore, investigations may aim to prosecute those responsible, or to learn from outbreaks so as to avoid future outbreaks by identifying unsafe practices that had led to the outbreak. Outbreak investigations have and will continue to be an important instrument for identifying new pathogens (e.g. *Cyclospora cayatenensis*, Herwaldt, 2000), new vehicles for known pathogens (e.g. *Salmonella* Tennessee in peanut butter, Anonymous, 2007c), new disease syndromes associated with known pathogens (e.g. febrile gastroenteritis associated with *Listeria monocytogenes*, Dalton *et al.*, 1997), and the re-emergence of problems that were thought to be under control (e.g. botulinum toxins in canned foods, Ginsberg et al., 2007). They are important sources of data for establishing the economic impact of foodborne illness on populations and may provide dose-response information for microbial risk assessment (Teunis *et al.*, 2008).

Many countries have surveillance systems for outbreaks of foodborne illness and data are reported at an aggregated level annually (Anonymous, 2007a) or over a number of years (Wang *et al.*, 2007, Cretikos *et al.*, 2008). New tools are becoming available to detect international outbreaks for foodborne viruses (Verhoef *et al*., 2009). Supranational agencies such as EFSA and ECDC in Europe present regular reports on foodborne outbreaks in a larger region or globally (Anonymous, 2007a). Outbreak summary reports provide important insights in current and emerging food safety problems but it is essential that such summaries are based on systematic surveillance activities. Reports in the peer-reviewed literature may suffer from publication bias and overestimate the impacts of milk/milk products, miscellaneous foods (e.g. sandwiches) and desserts while underestimating those of poultry, fish and shellfish, red meat/meat products and eggs/egg products (O'Brien *et al.*, 2006).

Molecular tools identifying causative agents in environmental and clinical samples, and molecular typing techniques identifying nucleotide sequences of single genes (i.e. *fla*-typing), techniques identifying sets of genetic elements (MLST, MLVA) and various restriction techniques (i.e. PFGE) have proven to be very useful aids in the epidemiology of foodborne illness. Developments in genotyping of pathogens and informatics have enabled the recognition of diffuse or multinational outbreaks which were previously unnoted (Gerner-Smidt *et al.*, 2006, Kirk *et al.,* 2004, Kroneman *et al*., 2008).

Estimating the incidence of sporadic cases of foodborne illness is more complex. Most existing surveillance systems are based on either notifiable disease reporting or laboratory surveillance. Both systems are passive in nature, and record only a minor proportion of all cases in the population. To estimate the true incidence of diseases that can be transmitted by food, active surveillance is necessary and more accurate estimates are needed for under-reported illness. The UK (Wheeler *et al.,* 1999, Tompkins *et al.,* 1999) and the Netherlands (De Wit *et al.,* 2001a, De Wit *et al.,* 2001b, De Wit *et al.,* 2001c) have carried out population-based prospective studies of infectious gastro-enteritis, combined with laboratory diagnostics to assess the proportion of cases due to specific pathogens. Currently, the UK has launched the second IID study (http://www.iid2.org.uk). Even in these large-scale projects, in a large proportion of cases (60%) it was not possible to identify a causal pathogen. However, it appears to be possible to reduce this diagnostic gap by the application of molecular methods (Amar *et al*., 2007). Population-based studies are expensive and time consuming, and several countries have attempted to develop less costly alternatives. These include FoodNet in the USA (Jones *et al.,* 2007), OzFoodnet in Australia (Kirk *et al.,* 2008), and the International Collaboration on Enteric Disease Burden of Illness Studies (Flint *et al.*, 2005, Roy *et al.,* 2006, Thomas *et al.,* 2006). As laboratory-based surveillance only detects a fraction of all illness occurring in the population, modeling approaches have been used to reconstruct the surveillance pyramid (Michel *et al*., 2000, Voetsch *et al*., 2004, Majowicz *et al*., 2005). Serosurveillance is now being explored as a new tool to provide internationally comparable estimates of the exposure of populations to foodborne pathogens (Simonsen *et al.,* 2008).

Although most surveillance activities are focused on gastro-intestinal illness, other symptoms are also commonly associated with foodborne illness. These may be more serious or of longer duration than GI illness. Furthermore, most pathogens that can be transmitted by food may also be transmitted by other pathways such as water, direct human and animal contact. Therefore, there is a need for source attribution to quantify the proportion of all cases that is foodborne, and the food vehicles that are most frequently associated with illness (Batz *et al.*, 2005). Molecular typing has successfully been used for source attribution of salmonellosis (Van Pelt *et al*., 1999, Hald *et al*., 2004) and more recently for campylobacteriosis (Wilson *et al*., 2008). Other methods being explored include case-control studies, outbreak studies, risk assessment modeling, natural or deliberate intervention studies at population level and expert elicitation. Each method is subject to specific biases, and may attribute illness to different points in the food chain. Therefore, interpreting the results from attribution studies should be done with care (Pires et al., 2009).

The World Health Organization has recently launched a new initiative to estimate the burden of foodborne illness on a global scale (Stein *et al.,* 2007). This initiative is advised by experts of the Foodborne Epidemiology Reference group (FERG), which assembles and appraises global evidence on foodborne disease epidemiology. This action is considered necessary in view of globalization, and to contribute towards meeting the Millennium Development Goals[1]. Results will be a basis for action at the global scale. Virtually no data on morbidity and mortality exist in large areas of the world, and even more data gaps are expected for attribution. Therefore, systematic reviews will be carried out and extrapolation will be necessary. As an example approach, the estimates of death from diarrhea for children under 5 from the Childhood Epidemiology Reference group (CHERG) will be used (Boschi-Pinto *et al.,* 2008); complemented with other methods, including expert opinion.

In addition to surveillance of human illness, systematic food chain surveillance is necessary to inform food safety decision making. Recent EU-wide baseline studies on the prevalence of zoonotic pathogens have illustrated the benefits of such standardized sampling and analytical approaches. For example, in the baseline survey on the prevalence of *Salmonella* in slaughter pigs, which took place between October 2006 and September 2007, it was demonstrated that approximately one out of every ten slaughter pigs in the European Union was infected with *Salmonella* in the lymph nodes, while one out of twelve pig carcasses was contaminated with Salmonella. The survey also indicated large differences between Member States (Anonymous, 2008c). These data will be the basis for risk assessment and cost-benefit analysis of

[1] http://www.un.org/millenniumgoals/

Salmonella control in the slaughter pig chain to support decision making by European risk managers.

The previous sections have described a highly complex set of interrelated factors affecting future trends in food safety. Predicting the impact of these factors is highly complex and surrounded by uncertainties. Hence, to be able to respond timely and appropriately, active, real-time surveillance in both the human and food system and communication to professionals responsible for infection control is of utmost importance.

6. Methodology

Molecular Methods for complex food analysis

One of the key challenges in food microbiology that has always been around and can now be addressed is to assess what molecular mechanistic processes underlie the observed physiological behavior of pathogens in food (see e.g. McMeekin *et al.*, 2007). Much of this work relies on a proper identification of (a) the microorganisms in the food at hand and (b) the food components that are relevant in determining the microbial stability of such foods. The latter range from small molecules (flavor-like molecules, food preservatives and other organic molecular) to the macro-ingredients i.e. proteins (peptides), sugar (polymers) and fats.

In many foods the microbes that are to be analyzed for are non-uniformly dispersed throughout the product. This is the case for many ready-to-eat products from the chilled food chain and is equally so for liquid products such as sauces and soups in which particles, as putative sources of microorganisms, may be non-uniformly mixed.

Analysis of microorganisms in foods may be done with two objectives in mind. On the one hand it may be a direct assessment related to production processes or inspection, on the other hand it may be research-oriented in which physiological inferences are made from molecular data. Rapid analysis techniques for use in industrial practice have to be easy to perform, low cost, optimally selective and must demonstrate reproducible sensitivity and specificity. Such methods need to be validated and written down in standardized protocols, preferably being able to provide quantitative data of use in risk assessment and in food safety management.

Currently they are generally based on DNA detection systems, either specific for ribosomal genes, or in the more advanced systems for specific sequences that occur along the entire genome (see e.g. Wattiau *et al.*, 2008; Scaria et al., 2008). Comparative genome sequencing is certainly at hand nowadays. Thus, in the case of relevant, closely related bacterial isolates it is increasingly easy to identify unique sequences (see e.g. the discussions in Earl *et al.*, 2008 and Medini *et al.*, 2008). Many of these may then be used to derive sequences amenable to use in DNA chip and / or PCR based detection platforms to the benefit of the safety assessment of food processing.

Although these methods are fast, highly specific and relatively sensitive, the application of molecular-based techniques in the control of food safety seems to be limited as they suffer from some serious drawbacks. The development of a horizontal method is seriously hampered by the fact that food products may contain interfering components. The development of horizontal methods becomes even more difficult due to a constant introduction of new matrices. While they are very sensitive, low copy numbers are difficult to detect when the sample size is very small. Introduction of an enrichment step preceding DNA-detection is a solution, but this makes results qualitative, rather than quantitative unless cumbersome MPN techniques are used. While this may not be problematic for quality assurance purposes, quantitative results may be necessary for risk assessment studies. Sample preparation needs close attention. Preferably, such sampling needs to be rapid and as homogeneous as possible. Innovative strategies focus on the use of magnetic beads coated with cell-recognizing molecules, on physical methods such as floatation, and on lysis of whole food matrices (Wagner and Dahl, 2008). The latter was described originally by Hein and co-workers who obtained enough bacteria from a complex set of food matrices in one-step approach taking only a few hours to be able to recover DNA for further study (Rossmanith *et al.,* 2007). It has yet to be established that such a procedure will also be effective for determining the concentration of bacterial spores. A limitation of currently available molecular techniques is also that they fail to discriminate between viable and inactivated organisms. Recent research may provide future practical solutions to this as transcriptional activity around bacterial cell survival / death reveals molecular markers for cell viability (Kort *et al*., 2008).

Finally, assays based on detection of multiple virulence genes can still give ambiguous results if there is a mixed culture to begin with. This may be for instance the case for the detection of the Shiga like toxin (stx) and Intimin (eaeA) genes from *Escherichia coli* in direct molecular analyses on food samples (e.g. Monday et al. 2007). The outcome will be positive with one strain having both or two strains, each having one of the virulence genes.

The issues discussed above corroborate the notion that it is necessary to properly validate such newly developed techniques. How do they compare to standard reference culture techniques described in ISO-protocols, and which controls must be used? Information is available on the efficacy of protocols using spiked samples, but little information is available on the efficacy of developed protocols in case of naturally contaminated samples. Without multi-laboratory validation, protocols for molecular techniques can be used for in-house purposes, but to be used as a standard method for the detection of pathogens, molecular techniques have to be validated, using a multi-laboratory approach, according to the ISO-16140 protocol.

In analyzing the composition of foods it is also more and more possible to detail comprehensively the full chemical spectrum of the compounds observed. To this end tools such as liquid chromatography (LC) or gas chromatography (GC) coupled to mass spectrometry (MS) are increasingly successfully used (reviewed in Hounsome *et al.,* 2008). A full analysis provides valuable information on product quality as well as the environmental parameters that can be most relevant to microbial survival (Beckmann et al., 2007). The analysis may also be used to detect the presence of microbial spoilage (Ellis et al., 2007). Pattern analysis to identify relevant compounds is the area where developments are rapid. While the costs of detection at the DNA level are increasingly reducing and interpretation of the data can now be automated to a large extent, this is not always as straightforward with the measurements of small to medium sized (mostly) organic molecules (see e.g. review by Hounsome *et al.,* 2008).

In the research area, molecular techniques are widely used. They can be used for the identification of organisms, for behavioral studies or for studying genes involved in (the regulation of) virulence and stress in response to different environmental conditions, while they can also be used in evolutionary studies. This approach marks the second objective, i.e. the use of genomics data to underpin physiological observations and to mechanistically 'explain' them. Here the analysis platform used need not be restricted to the cheaper methods, focused on biomarkers only; in fact the approach should be wider in nature while costing marginally more (though cost-effectiveness remains an important parameter). To pinpoint which type of compound has the most effect on the microorganisms at hand it is useful to analyze the genome-wide expression pattern and use the data obtained as a bioassay to identify the physiologically most sensitive environmental cues. Translation of molecular data into a biological meaning remains an essential subject for future studies. Application of techniques from other disciplines like ecology and medicine will be very useful.

Mining molecular data for new threats; metabolic capability models

New methods to screen sequenced genomes with the aim of understanding the physiological capability of microbes have led to the identification of major differences at the genome level between common laboratory strains of e.g. *E. coli* K12, enterohemorragic *E. coli* and uropathogenic *E. coli* (Brzuszkiewicz *et al.*, 2006; Fraser-Ligget, 2005; see also Perna *et al.*, 2001). Extending the analysis with state of the art rapid sequencing technique to other pathogens such as bacilli and certain streptococci has led to the realization of the so-called 'pan' and 'core' genome concepts (see e.g. Hiller *et al.*, 2007, Ara *et al.*, 2007 and the discussion in Medini *et al.*, 2008). In this classification the pan-genome is seen as being composed of three elements: the core genome, a set of non-essential genes shared by several isolates (strains) of the species, and a set of genes unique to an isolate. The size of each can significantly differ from species to species. The core genome generally gives the basic metabolic requirements of a certain species whereas the genetic plasticity of strains is generated by the other sequences (Medini *et al.*, 2008 for general concepts and Earl *et al.*, 2008 for specifics regarding bacilli). Data on the core genome aid significantly in defining the metabolic potential of an organism. Flux Balance Analysis is then often used as a modeling approach to find the possible steady states that the organism can attain (Schilling, 2000). In a next step such models may be detailed further to incorporate molecular signaling data at various levels of complexity (see Ropers *et al.*, 2006 for a highly detailed model of carbon starvation in *Escherichia coli*). Such extension should at all times be subject to scrutiny though in assessing its use given the investment of effort needed versus the (food) microbiological problem at hand needs to be considered.

7. Interaction between micro-organisms and their environment (foods) and microbial evolution

Understanding short-term adaptations of microbes

The recent genomics revolution has facilitated the interpretation of the molecular basis of microbial behavior. Examples stem from many fields and range from bacteria to filamentous fungi. The response to high-end temperature stress conditions often characteristic of the manufacturing process of savoury products is nowadays studied at the molecular level. This is most relevant to aerobic bacterial spore formers. Various strains of bacilli produce spores resistant to temperatures up to and well above those of classical sterilization at 121ºC (Oomes *et al.*, 2007). Keijser *et al.*, (2007) showed that spores express specific stress response genes during germination, some of which are likely responsible for repair of incurred thermal damage (see also Setlow 2006). In order to aim at understanding spore behavior after a thermal stress, in particular mechanisms of heat damage repair, we now have the possibility of utilizing the genome information for *Bacillus subtilis* 168 (Kunst *et al.*, 1997).

Cells subjected to acidic food conditions or to low water activity environments have been studied extensively in the context of food microbiology. Specific examples of food preservative stresses are those where cells have to respond to the antimicrobial action of a weak-organic acid such as sorbic acid. The latter is the most widely used food preservative. The common view is that the cells initially use energy driven pumps to extrude the acid from the cytosol while upon full adaptation they induce the synthesis of pumps specific for lipophilic weak acids (discussed for yeast extensively in Mollapour *et al*., 2008; for recent original research on sorbic acid stress response in vegetative bacteria see Ter Beek *et al*., 2008).

Stress adaptation of microorganisms in foods or upon being exposed to food processing conditions may lead to the induction of survival systems and could even induce virulence in pathogens. Many phenomena i.e. resistance to preservatives, oxidizing agents and natural extracts in foods are as important for successful infection as are mechanisms operative in 'in host', actual infection, and survival. Erickson and Doyle (2007) have illustrated this extensively for Shiga toxin-producing *Escherichia coli* and its survival on fresh produce, meat and in unpasteurized juices. Successful activation of stress response systems by some but not all strains may be instrumental in letting some strains adapt to the ‘adverse’ conditions in the food chain.

Presence and development of microbial virulence traits in non-human environments

The following section will provide some selected examples of microbial virulence traits of relevance to man of organisms present in non-human environments.

Foodborne pathogens may survive well in the animal production chain. Classical examples include *Campylobacter jejuni*, an organism not pathogenic to avian species but highly pathogenic to man (reviewed by Poly and Guerry, 2008). The organism is widely spread, as was demonstrated again by, for instance, the studies of Fearnley *et al.* (2008). These authors demonstrated the occurrence of hyperinvasive *Campylobacter* strains in isolates both from poultry and from human sources. There is not much data yet on the molecular basis of infection, be it that information on the intracellular signal transduction cascades of the organism becomes more and more available (Boyd *et al.*, 2007).

Salmonella species are well-known pathogens for animals and man. Callaway *et al.* (2008) have recently described their occurrence in various types of cattle. Sternberg et al. (2008) described an outbreak of *Salmonella* infection in a Swedish dairy herd. Schmidt et al. (2008) reported on *Salmonella enterica* infections in Swiss cattle in the summer of 2008. The various serovars have meanwhile been characterized at the molecular level (Edwards *et al.*, 2002). The use of such data can be in two non mutually exclusive directions. On the one hand the data provide information for use in quality control settings and epidemiological surveillance. On the other hand the gathered information can be used as a starting point in the formulation of novel research questions such as the molecular physiological mechanisms behind the observed microbial ecology. Studies aiming at answering such questions will require next to kinetic data on microbial metabolism measured at the population level, also quantitative data on cell - cell variation in microbial stress response in order to allow incorporation in next generation predictive food microbiology models (McMeekin et al., 2007). Both for non-pathogenic *B. subtilis* and for pathogenic *B. cereus*, spore formation of strains attached to naturally occurring biofilms is a well known phenomenon (Lindsay *et al*., 2006). It has also been documented nicely that spore-formation in a biofilm-like environment, a complex colony, leads to spores with a higher thermal resistance than that observed in spores originating from liquid cultures (Veening *et al.*, 2006). The (thermal) stress resistance of spores is again not a direct virulence trait but is does contribute to survival in the animal chain as well as transfer to the human food chain (Huck *et al.*, 2008). As such it is a crucial determinant of the likelihood of intoxication of the host. Other such virulence characteristics of e.g. *Bacillus cereus* include the resistance of the spores (and vegetative cells) to acid facilitating the 'settlement' and toxin production of the organism in the intestine (Wijnands, 2008; see also Stenfors Arnesen *et al.*, 2008).

Predictive modeling

Crucial is the conversion of molecular physiological 'analogue' data at the population level to data at the level of single cells relevant to the prediction of the behavior of low-numbers exposed to stressful environments (discussed in McMeekin *et al.*,, 2007; see for original research amongst others Den Besten *et al*., 2007). This provides insight into the link between the genome, gene-expression, protein and metabolic functional cellular units (see for the original physiology data Balaban *et al.*, 2004).). Koutsoumanis (2008) provides a clear example of the variability in growth limits of individual *Salmonella* Enteritidis cells subjected to NaCl stress. Another highly relevant example is the quantification of the germination and outgrowth processes operative in bacterial endospores (Stringer *et al*., 2005; Smelt *et al*., 2008). Both examples do not yet include a mechanistic analysis e.g. at the level of inclusion of genome-wide expression data. The initial challenge for future research is to do just that i.e.

study at single cell / spore level the molecular physiology in order to enable the generation of mechanistic models that describe the cellular heterogeneity in genetically homogeneous microbial populations. Phenotypic heterogeneity in microbial populations mediated by bi-stable signaling networks is a much discussed topic in general microbiology (e.g. Veening et al., 2008, Locke and Elowitz, 2009). To be of relevance to (predictive) food microbiology this will require performing experiments under the relevant physiological (food) conditions and as much as possible with the relevant food isolates.

Taxonomy

Regulators in the US use the GRAS approach (generally recognized as safe) to assess the safety of microbes used in food production, while the EFSA employs the QPS (qualified presumption of safety) system (Anonymous, 2007d). Basically it is assumed that a micro-organism that has been used for a considerable time in food manufacturing without causing problems can be considered “safe”. The difficulty in establishing these regulatory systems is the breadth of the taxonomic unit for which QPS or GRAS status can be conferred. If it only can be conferred at strain level, a full risk assessment will still have to be performed for any other strain, even those that are closely related to the ones having GRAS or QPS status. If it is applied at an overly high level, e.g. genus, it can happen that pathogenic cousins of a safe strain are wrongly considered harmless. The taxonomic units concerned are defined in the QPS list and the list is reviewed every year to take into account changes in the taxonomy and other considerations. If the history of safe use and the body of knowledge concerns only one strain, this property cannot be generalized to apply to the entire species, (an example is the *Enterococcus* spp). That is, if there is a risk that closely related strains of the safe strains are pathogenic, the taxonomic unit, species or genus, is not given QPS status, unless the pathogenic strains can be specifically identified, for instance by the presence of virulence factors. In the latter case the unit can be QPS, but with additional qualification. This is, for instance, the case for some *Bacillus* species.

Guidelines for the selection of “safe” cultures for biopreservation exist in the area of feed, but not for food (apart from the probiotics area). For general biopreservation, screening of cultures for virulence factors or other genes coding for undesirable properties would be relevant as well as studies of cultures possibly acquiring resistance.

For the approval of specific biopreservation agents (e.g. bacteriocins such as nisin) governmental as well as academic and industry views on the criteria to be applied differ around the world. Inconsistencies can cause problems, especially where criteria for safety evaluation or for an agent’s effectiveness are too lenient or because they do not take sufficient account of ill-informed use of such agents. The inverse, when a harmless strain is considered pathogenic is less problematic as the only consequence is that the strain undergoes an unnecessary safety assessment. Safe and robust use would need to follow general guidelines on the steps to be taken, as defined in the safe design of the product or the process.

8. Risk communication and education

Collaboration/communication between scientists

Historically, food systems used to be fairly simple; most foods were produced and eaten locally. Nowadays, a large share of our diet is produced in another country, and not uncommonly, several food ingredients come from different parts of the world (Käferstein *et al*., 1997). Furthermore, we prefer to eat the food as fresh as possible (Doyle and Erickson, 2008). This trend increases the need for worldwide food safety systems and thus collaboration and communication between all players in the food chain. As food chains extend or expand from local chains into worldwide chains, more and different factors may affect food safety. This implies that for food safety management knowledge or information from different scientific disciplines needs to be combined. Furthermore, the format in which information is made available needs to be standardized.

Food safety starts at primary production. To reduce the risk of foodborne gastroenteritis, especially for foods to be eaten raw, such as fresh produce and shellfish, knowledge about contamination routes and preventive measures is of great importance. In certain cases interventions could be effective early in the primary production phase, including the production environment. For fresh produce, grown in the open field, *Escherichia coli* (in particular the verocytotoxin producing strains VTEC) is, amongst others, a food safety hazard. This is underlined by a massive foodborne infection outbreak in the USA in 2006 (Anonymous, 2007e) caused by baby spinach eaten raw. The probable source of the VTEC in this outbreak was either irrigation water, or feces from cattle or wild boar. Cattle are a known source of this pathogenic bacterium (Hussein and Sakuma, 2005), yet cow manure continues to be used as a main soil fertilizer in organic farming (Anonymous, 2000b). Preventive measures could be a change of feeding diet in order either to reduce numbers of VTEC shed by cattle (Diez-Gonzalez *et al.*, 1998; Synge, 2000) or to reduce their survival in manure-amended soils (Franz *et al.*, 2005). Thus, produce safety can be increased in this case by combining agricultural science and (food) microbiology.

In other outbreaks, preventive measures are straighter foreword. In 2008, a *C. jejuni* outbreak in Alaska was linked to the consumption of raw peas contaminated on the field by Sandhill crane feces. The outbreak investigation identified a lack of chlorine residual in pea-processing water, which could have been easily prevented (Gardner and McLaughlin, 2008). Introducing buffer zones, set-back distances and fences to restrict wildlife access to the production environment of produce such as leafy greens may prevent problems with feral swine or deer (Atwill, 2008). Although these solutions seem sometimes fairly simple, they could only be taken due to a proper outbreak investigation that elucidated the (probable) source of the food contamination.

In order to stop an ongoing outbreak, such outbreak investigations should be carried out quickly, which relies on close collaboration between different (scientific) disciplines such as microbiologists, epidemiologists, wildlife control specialists, risk communicators, etc, often represented by different organisations or institutes, which may hamper the investigation. Although not standing on its own, the lack of proper communication between different scientific disciplines and/or institutes/departments during an outbreak investigation is shown in the 2009 U.S.A. *Salmonella* Saintpaul outbreak caused by raw jalapeño and serrano pepper in which more than 1400 cases were registered. This outbreak continued to spread due to malfunctioning at the level of policy, the public-health system’s organization and outbreak response, and its communications with the media and the public as concluded by the post-mortem investigation into this outbreak (Anonymous, 2008). <u>This clearly shows that collaboration of scientific disciplines is eminent to increase the safety of our food system</u>.

Other examples that demonstrate the added value of combining different expertises to limit the risk of foodborne illness are for instance *Vibrio* spp. in shellfish and mycotoxins in grain (products). The quality of shellfish depends on the quality of the water they are grown in. Pathogenic bacteria of importance in these types of products are *Vibrio* spp. The number of vibrios present in the water is positively related to the water temperature (Motes *et al.*, 1998) and models that predict ocean water temperatures can be used to predict the level of these pathogens in shellfish, thus combining (food) microbiology and oceanography, Ford et al., 2009). This knowledge was used by Californian lawmakers in order to ban the sale of raw oysters from certain waters during the warmer months of the year (Anonymous, 2003). The presence of mycotoxins on grain products is affected by weather conditions during growth and harvest. As humidity increases, growth increases of the moulds that produce mycotoxins on these products. Thus meteorological data are a reliable indicator of the risk of the concentration of mycotoxins on grain products (Schaafsma and Hooker, 2007; Van der Fels-Klerx *et al.*, 2008).

To determine which intervention strategy is the best or most cost effective to be taken to reduce risk of foodborne illness, a detailed risk assessment needs to be conducted and combining with a proper economic analysis may prove necessary. This requires a highly multidisciplinary approach, involving microbiologists, epidemiologists, risk modelers, economists and social scientists (Havelaar *et al.*, 2007a). An example of this approach is the CARMA project, carried out in the Netherlands, to evaluate options to reduce the risk of campylobacteriosis due to consumption of chicken meat (Havelaar *et al.*, 2007b).

For risk assessment studies, many data are needed like prevalence and numbers of foodborne pathogens in food. However, these data are not always available or are not in the correct format. For instance, data on the occurrence of *Salmonella* in food is mainly available as “presence or absence in 25 g of product” as legislation requires testing based on this criterion (Anonymous, 2005a). For risk assessment studies, however, the exact level of contamination is important (Malorny *et al.*, 2008). Even when data are available in the preferred format, problems may arise with nomenclature of foods or lack of other necessary details. For instance, a meatball can be either made from beef or pork or a combination of both. And problems increase with multiple-ingredient products, such as in lasagna. Uniformity in nomenclature of foods is, therefore, of great importance and a tool such as LanguaL, a food description thesaurus (www.langual.org), can be very useful.

In many food consumption surveys foods are categorized as, for example “beef with or without sauce”, whereas the degree of cooking of the meat is of greater microbiological relevance. To improve the use of data generated in food consumption surveys, closer collaboration between risk assessors, food microbiologists and nutritionists is needed.

In the Netherlands, the Dutch Food and Consumer Product Safety Authority (VWA) works closely together with the National Institute for Public Health and the Environment (RIVM) to improve the quality and usefulness of the data obtained from routine monitoring programs of the microbiological quality of foods. Some recent studies focused on the relative microbiological risk to consumers associated with the consumption of fresh vegetables (Pielaat and Wijnands, 2008) and prevalence of potentially pathogenic *Bacillus cereus* in food (Wijnands *et al.*, 2006). A simple, spreadsheet-based tool is being developed to assess consumer risks associated with such products using available data (Evers and Chardon, 2008).

These examples clearly show the benefits of inter- and intra- scientific collaboration. Close personal collaboration may not always be necessary when data can be shared by other means. In the scientific literature, many data are published that can be used by others. However, translation of these data to a uniform data set is time consuming. In order to improve sharing of data on microbial growth and inactivation, the ComBase Initiative was established, a collaboration between the Food Standards Agency and the Institute of Food Research from the United Kingdom, the USDA Agricultural Research Service and its Eastern Regional Research Center from the United States and the Australian Food Safety Centre of Excellence (Combase Consortium, 2008). ComBase is a combined database of microbial responses to food environments and data can be used for predictive modelling. Recently, Combase started a collaboration with the Journal of Food Protection, which request authors to submit their data to the Combase database. A clearinghouse of interdisciplinary data is offered by Foodrisk.org, an initiative of the Joint Institute for Food Safety and Applied Nutrition (JIFSAN), and which is a collaboration between the University of Maryland (UM) and the Food and Drug Administration (FDA). The clearinghouse provides data and methodology on food safety risk analysis offered by the private sector, trade associations, federal and state agencies, and international sources (www.foodrisk.org).

In conclusion, although interdisciplinary collaboration sounds very promising, it must be noted that it is generally not straightforward as, for instance, the data thus made available may be limited or be in the wrong format. Despite this, with some extra effort more progress may be achieved than would otherwise be the case.

Education

New trends in food consumption patterns, e.g. the consumption of raw foods, and the introduction of a wide variety of newly developed food products with each their specific way of preparation (e.g. ready-to-eat; ready-to-heat), in combination with an increase in the number of vulnerable people, require clear communication about food safety aspects, communication between industry, consumer and government.

Risk managers should be aware and understand public concerns about food safety as this must be the basis of a risk management strategy (Frewer, 2004). Whether such a strategy will be judged as effective by the public, will depend on the expertise of food risk managers (Van Kleef *et al*, 2007) and cultural variation: what is effective in one country, is not always as effective in another (Van Dijk *et al*, 2008).

Industry

When introducing new food products or new food preparation techniques, it is crucial to provide pertinent information concerning safe food handling and preparation. The food industry can contribute to food safety in several ways. Labeling, providing information about correct storage conditions and ways of preparation, can contribute to food safety, although the addition of more information is at odds with providing clear food labels (Mills *et al*, 2004). Icons can be used. The food industry can further contribute to food safety by educating professionals working along the food production chain. As an example, the efforts of the public-private partnership formed by the Industry Council for Development with FAO and WHO may be noteworthy (Motarjemi, 2006).

Consumers

Presence of pathogenic bacteria on raw food materials, such as meat and fresh produce is in most cases not totally avoidable, therefore intervention strategies are also needed in subsequent steps of the food chain to reduce the risk of foodborne illness for the consumer. At the other side of the farm-to fork continuum the consumers also play an important role in maintaining food safety. Different information campaigns therefore focus on improving home hygiene (Anonymous, 2008d, 2008e). However, the impact of such campaigns is often not evaluated. A combined research project undertaken by social scientists, food microbiologists and risk assessors showed the limited effect of such campaigns on reducing the actual level of bacteria present in a meal, and on the associated risk of human illness (Nauta *et al.,* 2008).

Slovic (1987) developed a psychometric paradigm, which demonstrated that psychological factors determine a person’s response to different hazards, including those in the area of food safety. Do consumers know what they should do in order to prepare a safe meal, in particular how to avoid cross-contamination and proper heating of reused food (Fischer *et al*, 2007)? According to Nauta *et al* (2008), consumers already possess the necessary knowledge regarding hygiene practices; this knowledge only needs to be activated. How to achieve this? What can we learn in this respect from other education campaigns, (e.g. smoking, alcohol, and fat), and from communication of medical product risks (Goldman, 2004), or from programs focusing on disease prevention and control (O’Loughlin *et al.*, 1995; Sarraf-Zadegan *et al.*, 2003).

Health related behaviors are often differentially distributed across socioeconomic groups. Close collaboration with social scientists therefore seems logical. Communication with different consumer groups requires different approaches and media. Information should be targeted to specific groups at risk, like single households, pregnant women or elderly people. This is because different groups have different food preparation and cooking habits and therefore are exposed to different levels of risk. Kornelis *et al.* (2007) showed that different consumers prefer different information sources when posing questions about food safety. Two-thirds of all consumers prefer information from either institutional or social sources. *Your life*, a free magazine containing articles about fashion, lifestyle and entertainment published by the UK National Health Service, is an illustrative example of such a social source. Members of the lower socioeconomic groups are more likely to respond to information from their direct social environment (Weenig and Midden, 1997). Apparently, communication with different consumer groups requires different approaches and media. The importance of educating children through the school system cannot be emphasized enough.

Government

Food preparation and cooking practices are based on habits. This goes for consumers and often also for food professionals in small food establishments, such as food services and restaurants. Since such behavior is difficult to change, education should be a life long learning process on general aspects of food safety as hygiene and contamination routes, starting at a young age, making use of all types of media, including video-gaming. Education in food safety aspects could be combined with nutritional information. Concurrent with the development of education programs, strategies should be developed to measure the impact of such programs (Nauta *et al.*, 2008). In addition, governmental organizations should consider introducing and supporting specific education programs for consumers and professionals in small food preparation enterprises as well as for producers, especially for producers of fresh produce, as all have their responsibility with regard to food safety.

Finally, while education might be expected to improve food safety in the developed world, in developing countries, economic growth rather than education might be the best way to minimalize food-related mortality and morbidity amongst children.

Acknowledgements

The authors wish to thank all participants of the conference for their willingness to share their insights in plenary and group discussions. Special thanks are due to the plenary speakers (see contributions elsewhere in this Special Issue), the chairs and rapporteurs of breakout groups and the members of the Scientific Committee. These include, apart from the authors, Andrea Ammon, József Baranyi, Bob Buchanan, Patrice Buche, John D. Collins, Paul Cook, Mike Doyle, Aamir Fazil, Leon Gorris, Colin Hill, Marta Hugas, Marion Koopmans, Hilde Kruse, Hein van Lieverloo, Pia Makelä, Tom McMeekin, Diane Newell, Wim Ooms, Tom Quested, Peter Raspor, Tom Ross, Moez Sanaa, Don Schaffner, Claudia Stein, Mark Tamplin, Rob Tauxe, Mieke Uyttendaele, Yvonne Van Duynhoven, Didier Verloo, and Martin Wagner.

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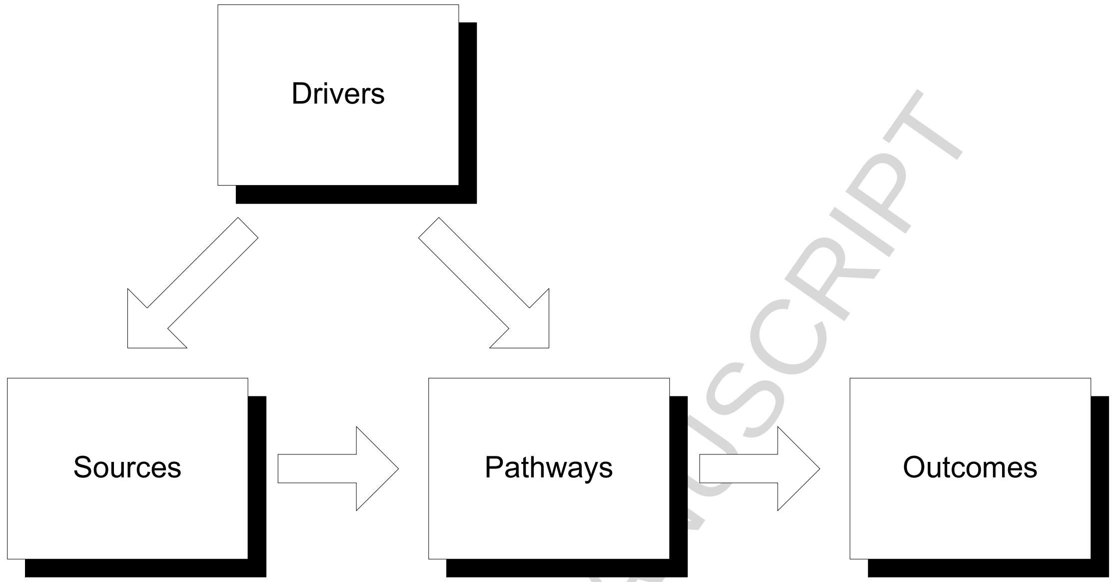
1438 Figure 1. Basic risk framework for infectious diseases (from Tait *et al.*, 2006)



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1441 **Table 1. Different aggregation levels for evaluation of food safety.**

MICROORGANISM-RELATED FACTORS	HUMAN-RELATED FACTORS
Ecosystems	Global systems
Food chains	Regions
Food products	Countries
Food products	Consumer
Populations of micro-organisms	Human populations
Individual cells of micro-organisms	Human individuals
Cellular and molecular processes	Cellular and molecular processes

Table 2. A systems approach to food safety.

Bold red font: Source increases risk to food safety;

Blue, normal font: Effect on risk to food safety unclear or neutral;

Green italics font: Source reduces risk on food safety.

DRIVERS	SOURCES	PATHWAYS			OUTCOMES
	Pathogens	*Farm*	*Processing/distribution*	*Preparation/Consumption*	*Public health*
Economy					
Globalization	**Reduced geographical barriers to spread (of new variants)**	**Inadequate sanitation: higher pathogen loads** **Global sourcing** **Intensified contact structures**	**Long and complex supply chains** **Varying hygiene levels**		**Increased risk**
Food price / income level		**Less profit margins; decreased investment in food safety**		Preference for cheaper alternatives (e.g. less meat and butter; discounters; home brands)	Risk not clear
Science and technology and industry					
Minimal processing	**Adaptation**		Less kill steps		**Increased risk if not well controlled**
Innovation		**New food animal species**	Step change food innovation *Smart packaging* *Bacteriophages*	*Smart labels*	Risk not clear

DRIVERS	SOURCES	PATHWAYS			OUTCOMES
	Pathogens	*Farm*	*Processing/distribution*	*Preparation/Consumption*	*Public health*
Laboratory methods	**Discovery of new pathogens or variants** **Omics approaches**				**Increased observed risk**
Culture and demography					
Population growth		Polluted environments		**Increased demand**	**Increased risk**
Migration				New food habits	
Age structure				**Increase in elderly** **More premature babies**	**Increased risk**
Nature and environment					
Climate change and regional differences	Changing ecology	Droughts, floods Competition for land resources Movement of farms to new areas		**Population displacement** **Increased difficulties to maintain cold chain**	Changing spatial patterns of risk
Water, waste and energy		**Irrigation water quality** **Waste recycling**	Water/energy savings cleaning, process and ingredient water quality		**Increased risk**
Evolution	**Emergence and transfer of virulence factors** **Antimicrobial resistance**	**New reservoirs**	**Increased survival**	**Increased infectivity**	**Increased risk**
Population contact structures	**Species jumps (spill-over from epizootics or exploitation of new**	**Contact zoonoses (MRSA, Q-fever)**			**Increased risk**

DRIVERS	**SOURCES**	**PATHWAYS**			**OUTCOMES**
	Pathogens	*Farm*	*Processing/distribution*	*Preparation/Consumption*	*Public health*
	agricultural areas)				
Consumer behavior					
Food choice	**Psychrotrophs** **Re-emerging pathogens**	Exotic/ethnic foods Regional products	**No or mild processing, less heat treatment** Increased pre-processing and -packaging	**Convenience foods** **Year round availability** Healthy foods (fish, vegetables & fruits) Less fat/salt/sugar Eating outside home	**Increased risk**
Food handling Technologies Attitudes/education			**No acceptance of irradiation**	**Storage: inadequate time/temp control**	**Increased risk**
Information					
Surveillance	**Identification of new pathogens** **Detection of unexpected events**		*Effectiveness of current controls*	Changes in consumption patterns: who, what, where, why?	**Increase in observed risk**
Education		*Professional education*		Hygiene campaigns Attitude changes to accept safe technologies	*Reduced risk*
Government and policies					
Regulations		*Standardisation* *Ban of antibiotic growth promoters*	*GHP/HACCP* **Fraudulent behavior**		Reduced risk

DRIVERS	**SOURCES**	**PATHWAYS**			**OUTCOMES**
	Pathogens	*Farm*	*Processing/distribution*	*Preparation/Consumption*	*Public health*
Risk (-benefit) assessment		*Targets for pathogen reduction*	*Risk-based targets*		*Reduced risk*
Food defense		Agro/bioterrorism			Increased risk
Agriculture					
Animal friendly and organic production	*Reduced AMR*	**Re-emergence (Trichinella, Toxoplasma)** **Higher (*Campylobacter*)** or *lower prevalence (Salmonella)*			Risk not clear
Aquaculture		More farmed fish			Risk not clear
Antimicrobial use	**Resistance development**	**Increased therapeutic use**			**Increased risk**