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Microbiological and chemical risk assessment and its respective exposure modelling

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Introduction

Different chemical (Fig. 1) and microbiological (Fig. 2) hazards may be present in foods. Most of them are readily controlled by adequate actions along the whole food supply chain from farm to fork within established food safety systems. Over the course of time, various concepts relating to either the hygiene or chemical and microbiological safety of foods have been developed. Nowadays, these concepts relating to the safety of food have to be linked to public health. Mainly international organizations like FAO/WHO and Codex Alimentarius strongly support the development of a harmonized proceeding for the analysis of different risks such as chemical, microbiological and physical impurities. Thus, existing international guidelines should be aligned to result in measurable and comparable outcomes of health risks. This process is carried out by the so called risk analysis process, which is a systematic, disciplined framework. Within the risk analysis process, the scientific and consistent base of risk assessment leads to the estimation of risk. It comprises the components hazard analysis, hazard characterization, exposure assessment and risk characterization. Risk assessment for food is always complex when executed from farm to fork because of many levels of the food supply chain, numerous variables involved and restricted data availability. Moreover, the amount of food consumed as well as the amount of chemical and microbiological contamination ingested vary considerably and depend on region, climate, product and processing parameters. Over all, this situation leads to a not exactly predictable, but measurable or calculable (by computer simulation) ingestion of the different contaminations. Important differences exist between the different classes of hazards: certain chemical hazards, as for example pesticides and antibiotic residues are strictly controlled. Their presence in food is not desired and only allowed in concentrations considered to be safe. The presence of carcinogenic chemicals and toxins in food should be avoided. These substance classes should not be brought into the environment or the food supply chain. On the other hand, microbiological hazards often are living microorganisms also present in the environment and capable of reproducing in production facilities as well as in foods. This means, that slightly contaminated food, e.g. soft cheese, may develop considerable amounts of (pathogenic) microorganisms and / or toxins until the time of consumption.

Fig. 1:
Aflatoxin B_1 as an example of a chemical hazard

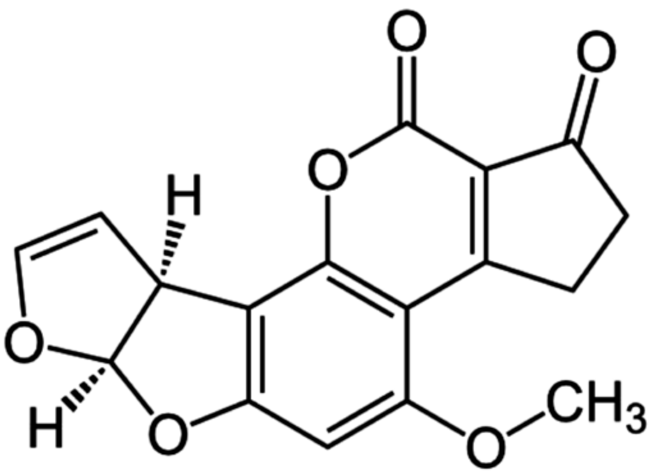
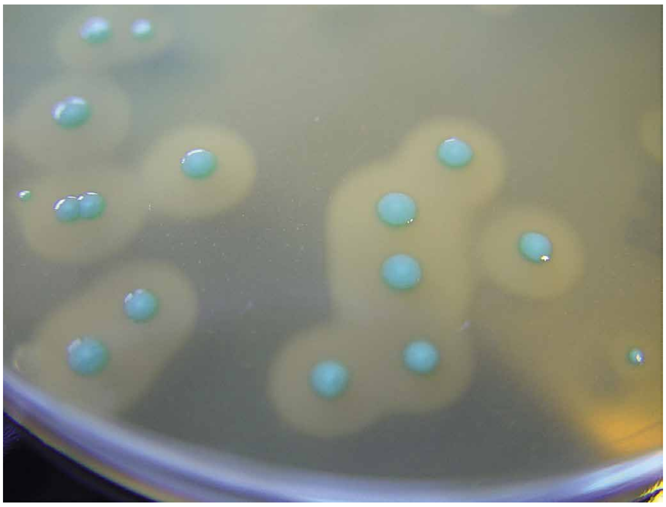


Fig. 2:
Listeria colonies on an Agar culture plate as an example of a microbiological hazard



Microbiological risk assessment

Risk based food safety covers the complete food chain starting with raw materials at farm level. It follows the processes they undergo in production, transportation and storage of the finished products at retail level as well as private transport and storage in the homes of consumers. Finally, it ends up with the expiring date, the end of shelf life or the preparation and ingestion by consumers. The link between microbiological food safety and public health is achieved by means of the Appropriate Level of (Consumer) Protection (ALOP) and the Food Safety Objective (FSO). Hereby, the ALOP expresses the maximally tolerated illness cases per 100'000 inhabitants and the FSO represents, within this given ALOP, the maximal possible exposure of consumers to a certain microbiological hazard. The exposition to a hazard takes place with the time of ingestion of a food. Therefore, producers must guarantee the safety of their products during shelf life and they need to fulfil performance objectives (PO) more stringent than the FSO. For that reason, they demand for substantial reductions of eventually present pathogenic bacteria by sharp process performance criteria (PC; Fig. 3).

The basic microbiological risk metrics are given by the formula (1):

$$Ho - R + I \leq FSO$$

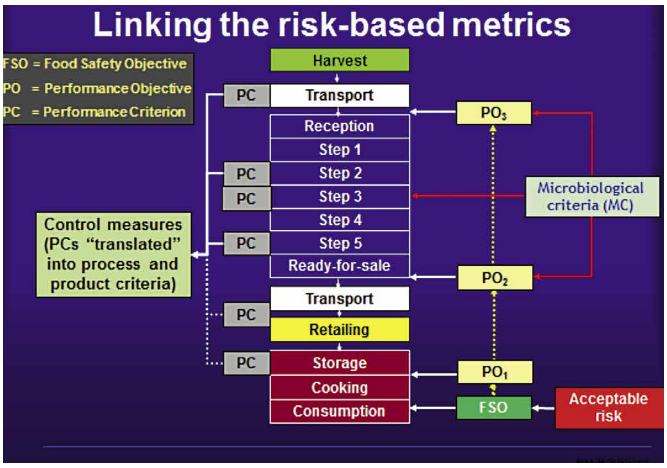
where: FSO = Food Safety Objective

Ho = Initial level of the hazard

R = Total (cumulative) reduction of the hazard

I = Total (cumulative) increase of the hazard

Fig. 3:
The linking of risk-based metrics between microbiological food safety and Public health (Reproduced with cordial permission of Claus Heggum, Danish Dairy Board, 2014)



In words: the microbiological hazard of the primary product minus the sum of effects reducing the hazard plus the sum of effects increasing the hazard must be smaller or equal to the FSO (at the time of consumption, within shelf life).

This risk metrics applied along the whole food supply chain allow for different proceedings and measures to be taken in order to reach an established FSO. For this reason, only limited information is gained when performing all microbiological controls of an (intermediate) product at one stage of the food supply chain. Better knowledge about the behaviour of a microbiological contamination and its propagation along the levels of the food supply chain allows for better definition of PO`s and PC`s for a certain product with its specific history and intended use. Where the propagation of a microbiological hazard along the food supply chain is unknown, e.g. due to a lack of reliable data, the prevention of multiplication of the microorganisms is preferable to the control of growth.

Of course, traditional and artisanal cheese production for example strongly relies on prevention of microbiological contamination, i.e. on minimal microbiological contamination in the initial farm and bulk milk. Subsequently, control of excessive growth exceeding the FSO is aimed. The microbiological contamination at single farm level mainly is composed of the frequency and extent of microbiological contamination in herd animals and in their environment. Both types of contamination might grow during milk storage and transportation. The contaminations are substantially imposed by the presence of udder inflammation causing shedding of pathogenic microorganisms, the hygiene of production places and environmental contamination. The growth of contamination is mainly controlled by short periods of milk storage at low storage temperatures.

On the other hand, industrial production often applies a process step eliminating (almost) all present pathogenic microorganisms. Subsequently, recontamination of the product is avoided and growth prevented with permitted food additives and the control of intrinsic parameters (pH, a_w). Finally, only few hazards eventually present in milk pose a health risk to consumers of traditional and industrial produced cheese varieties.

Available surveillance and modelling systems for pathogens

The surveillance of foodborne diseases in humans and of diseases in animals helps to safeguard food safety and therefore public health. Most systems actively or passively report illness cases in order to: a) estimate the burden of foodborne disease and monitor trends; b) identify priorities and priority setting in the control and prevention of foodborne diseases; c) detect, control and prevent foodborne disease outbreaks; d) identify emerging food safety problems; and e) evaluate foodborne disease prevention and control strategies (2).

Internationally, data is available e.g. on FoodNet, a collaboration between CDC, Food and Drug Administration, and the United States Department of Agriculture (3).

European surveillance on 52 communicable diseases and conditions may be found at the European Center for Disease Prevention and Control (ECDC) (4) and at the European Food Safety Authority (EFSA) (5). Laboratory surveillance systems are e.g. the pulseNet coordinated by the US-CDC (6) and the WHO Global Foodborne Infections network (7). Different National Public Health institutions maintain data on foodborne diseases due to pathogens, e.g. Canada (8) and France (9).

The safety of foods in production, distribution and consumption is of special interest for the food industry. Some modeling systems allow for the prediction of microbiological contamination over time with constant or changing intrinsic and extrinsic factors, e.g. pH, time, temperature, the presence of salt or acids, water activity at aerobic or anaerobic conditions. Such expert systems with user-friendly software and free access are the USDA`s Pathogen Modeling Program (10; Fig 4) and ComBase (Fig. 5) - a combined data base for quantitative and predictive microbiology. Its main components are: a database of observed microbial responses to a variety of food-related environments and a collection of relevant predictive models. ComBase is managed by the ComBase Consortium consisting of the Institute of Food Research (IFR) in the United Kingdom, the USDA Agricultural Research Service (USDA-ARS) in the United States, and the University of Tasmania Food Safety Centre (FSC) in Australia. Other partners are Mexico, Japan and Greece as well as the international food industry (11). SymPrevius, another

Fig. 4:
Screen shot of the Internet opening site of the Pathogen Modelling Program

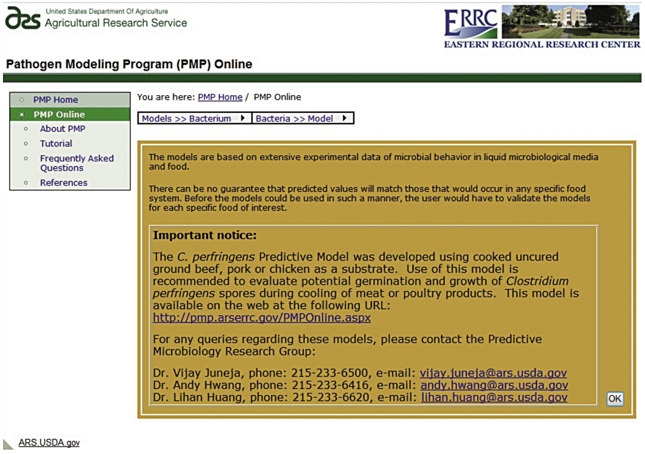
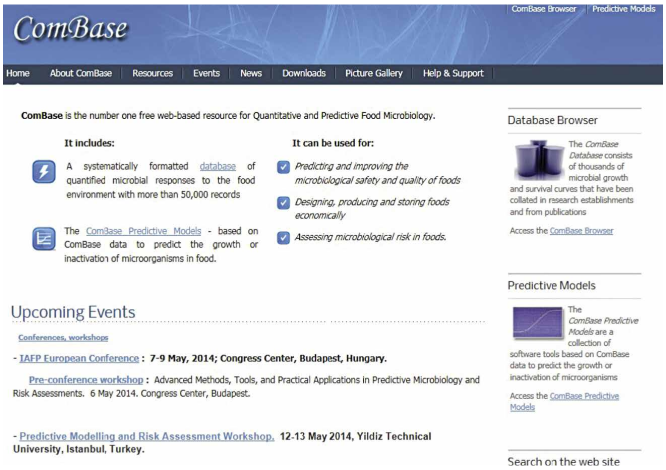


Fig. 5:
Screen shot of the Internet opening site of the ComBase model system

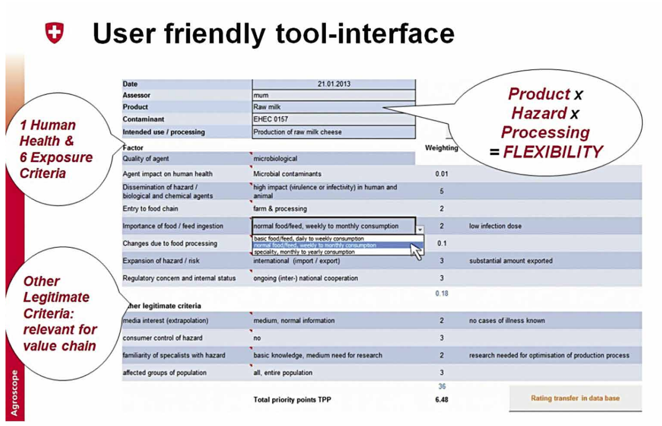


software is available in France for an annual fee (12; Fig. 6). In general, the models help to elucidate expected growth, survival or decline of pathogenic microorganisms in different foods or broths and at different conditions during production and storage. Therefore, the models help to optimize food processing and food formulation.

Recently, the risk assessment based iRISK 1.0 model with the purpose of ranking microbiological and chemical risks has been released by the FDA. It has a need for qualified assumptions to be taken on dose-response as well as consumption models and on severity of the health outcomes. The model helps comparing the health risk arising from microbiologically and chemically contaminated foods (13). A practical risk ranking framework with criteria and sub criteria remaining within an expectable horizon of knowledge and experience of risk managers in general has been presented and explained by Agroscope (14; Fig. 7). The model aims to rank the different classes of hazards, i.e. the (micro-)biological, chemical, physical and nutritional hazards in food and feed with their related research needs.

Fig. 6:
Screen shot of the Internet opening site of the Sym`Previus model system

Fig. 7:
Screen shot of Agroscope`s user friendly risk prioritization tool-interface



Risk assessment of chemicals

Undesired substances in foods such as pesticides, their residues and metabolites, environmental contaminants, contaminants in plants and food additives may enter the feed and food chain on numerous routes: air, soil and water may contaminate food and feed on the field, plants may form toxins, pesticides are applied for plant or crop protection, moulds grow on plants and crops, farming animals are treated with medicines including antimicrobial agents, food additives help in the production and storage of foods and contaminants may be formed in heat treating processes during food production and food preparation. Chemical contaminations gained attention with the wider use of plant protection products and the considerable amount of chemicals produced internationally.

International agreements on the Application of Sanitary and Phytosanitary Measures (SPS Agreement) and international bodies like the European Food Safety Authority (EFSA) now develop and apply risk assessment systems and measures to guarantee safe foods from plant and animal origin. Of special importance to feed and food risk assessment are: a) data in mammals on absorption, distribution, metabolism and excretion of a chemical (ADME) providing insight in its metabolic behaviour and the kinetics of excretion, b) data on acute and long term toxic effects in laboratory animals, namely mice, rats, farming animals and dogs, c) the toxicological mechanism of action, d) in vivo and in vitro testing of carcinogenicity, genotoxicity, neurotoxicity, reproductive and developmental toxicity, e) the (cumulative) exposure to or intake of a chemical through food and feed, and f) access to data from human epidemiological studies.

For compounds that are considered to have a threshold of toxicity like e.g. plant protection products, acceptable (ADI) and tolerable daily intakes (TDI) are defined (15, 16). Hereby, the ADI stands for compounds intentionally entered to a food and the TDI for compounds unintentionally present in foods, e.g. (environmental) contaminants. The ADI/TDI represents an estimate of the amount of a compound that can be ingested every day and all life long without measurable health risk. To derive these values, the lowest dose without effect in the most susceptible species tested is identified (no observable adverse effect level (NOAEL) and the lowest observable adverse effect level (LOAEL)). Finally, to derive the ADI, the NAOEL is divided by a safety factor (typically 100). The safety factor roughly is composed by 10 for the extrapolation of animal data to humans multiplied with 10 for variability among humans. Moreover, each safety factor is subdivided in order to allow for differences in toxicokinetics (delivery of the chemical to its site of action/toxicity) and toxicodynamics (extent of effect or response due to the presence of the chemical). Contaminants with a long half life time or accumulating in humans are referred to as (provisional) tolerable weekly (PTWI) or tolerable monthly intake (TMI), both of them, as for ADI/TDI, expressed at the basis of body weight. For compounds with the capacity to cause acute effects after only a single dose or after a short time of intake, an acute reference dose (ARfD) is derived. This value generally is similarly derived as the ADI but the basis usually is a NOAEL for acute effects.

In the process of setting safe maximum residue levels (MRL) for compounds in food and feed, the MRL should be set in such a way that the expected exposure due to the consumption of the commodity with the compound at its MRL should not exceed ADI, TDI or ARfD.

In the past, contaminants relied on the "as low as reasonably achievable" – principle (ALARA), which rather represents a risk management tool and does not refer to available toxicological data.

Genotoxic and cancerogenic compounds are believed to be non-thresholded, because for genotoxic effects current paradigms assume that no safe doses (e.g. ADI or ARfD) can be determined. Currently, such compounds without a threshold of toxicity are assessed by the margin of exposure (MOE). The MOE represents the ratio between a dose of a compound inducing tumors at a certain incidence in animals (or humans) and the human exposure to this compound. For the calculation, basically an effect dose with known tumor incidence is needed. Hereby, typically the mathematical modelling of the dose-response relationship of effects at 10 % tumor incidence is used as Bench mark dose (BMD) and BMDL refers to the corresponding lower limit of a one-sided 95 % confidence interval on the BMD. So, a MOE of 100 based on a theoretical BMDL10 would mean, that the expected exposition would result in 1 tumor per 1`000 persons. If the same MOE of 100 is based at a BMDL0.1, the expected exposition would result in 1 tumor per 100`000 persons. So, the smaller the dose from exposure is, the larger results the MOE and the compound under consideration is of minor immediate priority. Thus, the MOE provides an instrument for the comparison and prioritization of compounds, a major advantage for risk comparison.

Available modelling systems for chemicals

As an alternative to the LOAEL and NOAEL extrapolation approach, the BMD/L software is more sophisticated and takes into account all existing dose-response data of a chemical substance. The program is used by the United States Environmental Protection Agency for pollutants. The model considers the mode of action of a substance and whether the effects of concern are likely to be linear or nonlinear at low doses. The incidence of a health response at a certain dose of a substance is identified (17). The EFSA recommended some changes, so that the thereof derived BMR model or the sophisticated Dutch model PROAST for statisticians - in contrast to the simple BMD software - also considers the input data quality and the uncertainty (18, 19). Both models estimate a health response at any dose of a substance. All 3 models find applications in food safety, chemical risk assessment, Plant Protection Products, biocides, occupational safety, environmental contaminants and carcinogens.

The PRIMo model, rev. 2 (Pesticide Residue Intake Model) incorporates all national EU diets. With this model, acute and chronic exposition to pesticide residues of consumers in foods are calculated (20). Initially, the model was developed in order to assess the risk of provisional maximum residue levels in foods.

The exposure to chemical substances in consumer products and consumer articles may be assessed by the ConsExpo model initiated by the Dutch Government. The model covers inhalation, oral and dermal routes of exposure. It`s software and fact sheets of input data are used for the assessment of (semi-) volatile substances within REACH as well as for biocides, cosmetics, toys and sprays. The model allows for subsequently more sophisticated analysis from simple and quick worst cases with low information input towards refined assessments with higher quality input data (21).

The exposure of operators, bystanders and residents by oral, dermal and inhalation routes is elucidated by a variety of models like UK POEM and the German model for outdoor applications and EUROPOEM for indoor uses. Different models are used for the assessment of operator exposure to plant protection products within EU member states (22).

Recently, a number of modern in vivo and in vitro methodologies and in silico tools have been developed to investigate toxicokinetic (TK) and toxicodynamic (TD) processes of chemicals, i.e. Mode of Action (MoA)/Adverse Outcome Pathway (AOP) at different levels of biological organisation (organism, organ, cellular and molecular level). These methodologies provide the opportunity to move towards a mechanistic understanding of toxicity (23), but there remains a lot of research to be done within the above mentioned methodologies and tools.

Zusammenfassung

In Lebensmitteln können verschiedene chemische und mikrobiologische Verunreinigungen auftreten. Risikobasierte Lebensmittelsicherheit deckt die ganze Lebensmittelkette ab, beginnend mit den Rohmaterialien auf dem Niveau der Primärproduktion. Sie folgt den Produkten durch die Veränderungen, die diese während der Verarbeitung, dem Transport und der Lagerung der fertigen Produkte bei Herstellern und Verteilern wie auch beim privaten Transport und der Lagerung in den Haushalten der Konsumenten erfahren. Fragen der Lebensmittelsicherheit enden erst mit dem Ablaufdatum oder mit der Zubereitung und dem Verzehr des Produktes durch den Konsumenten. Aus diesem Grund werden die orale Aufnahme von Verunreinigungen durch Konsumenten, aber auch die Aufnahme durch Inhalation und der Hautkontakt von Anwendern von Chemikalien berechnet. Um diesem Ziel zu entsprechen, führt die vorliegende Veröffentlichung einige Datenbanken über mikrobiologische Lebensmittelsicherheit und öffentliche Gesundheit auf. Die grundsätzlichen Berechnungsweisen und verbreitete computergestützte Systeme zur mikrobiologischen und chemischen Risikoberechnung werden kurz vorgestellt. Moderne Systeme zur Darstellung der Vermehrung und Abnahme von pathogenen Mikroorganismen sind zum Beispiel das PMP, ComBase sowie das SymPrevius Modellsystem. Für Chemikalien existieren zum Beispiel das BMD-Modell, die ConsExpo und die PRIMo Datenbasis wie auch das PROAST Modell für Statistiker. Darüber hinaus werden zwei Vorgehensweisen zum Vergleich von verschiedenen Risiken und Risikoklassen - wie sie zum Beispiel mikrobiologische und chemische Risiken darstellen, erwähnt: Einerseits ein praktisches Modell, welches mit indirekten Risikomerkmalen innerhalb vom Wissensstand von Risikomanagern arbeitet, andererseits ein der komplexen Risikobewertung angelehntes Modell für Spezialisten. Heutzutage wird bei Chemikalien deren toxischer Wirkungsweise vermehrt Beachtung entgegengebracht. Daraus resultierende moderne Vorgehensweisen haben das Potential, die Toxikologie hin zu einem mechanischen Verständnis zu führen, allerdings muss dafür noch viel Forschungsarbeit geleistet werden.

Résumé

Les denrées alimentaires peuvent être contaminées par diverses impuretés chimiques et microbiologiques. La sécurité alimentaire basée sur le risque couvre l'ensemble de la chaîne alimentaire, de la production primaire des matières premières jusqu'à la consommation des produits finis. Elle tient compte des modifications que les produits subissent pendant la transformation, le transport et l'entreposage chez les fabricants et les distributeurs, mais aussi pendant le transport et l'entreposage par les ménages privés. La sécurité alimentaire ne prend fin qu'avec l'échéance de la date de consommation ou la préparation et la consommation du produit par le consommateur. Raison pour laquelle on tient compte, dans les calculs, de l'ingestion orale d'impuretés par les consommateurs, mais aussi de l'ingestion par inhalation et par contact avec la peau des utilisateurs de produits chimiques. Pour répondre à cet objectif, cette publication présente quelques banques de données sur la sécurité alimentaire et la santé publique. Les méthodes de calcul de base et les systèmes assistés par ordinateur pour le calcul du risque chimique et microbiologique y font l'objet d'une brève présentation. Certains systèmes de modélisation de la multiplication et du déclin des microorganismes pathogènes sont par exemple les modèles PMP et ComBase de même que le système de modélisation SymPrevius. Pour les produits chimiques, il existe par exemple le modèle BMD, le ConsExpo et la banque de données PRIMo de même que le modèle PROAST pour les statisticiens. Par ailleurs, deux méthodes de comparaison des divers risques et classes de risque – comment par exemple elles classifient les risque microbiologiques et chimiques – sont présentées: d'une part un outil qui se base sur des critères de risque indirects, connus des personnes chargées de la gestion du risque en entreprise et, d'autre part, un modèle qui s'appuie sur l'évaluation complexe du risque, destiné quant à lui aux spécialiste. De nos jours, on porte davantage d'attention au mode d'action des produits chimiques. Les méthodes modernes qui en résultent ont le potentiel de décrypter le mécanisme toxicologique des produits chimiques, mais de nombreux travaux de recherche sont encore nécessaires.

Summary

Different chemical and microbiological hazards may be present in foods. Risk based food safety covers the complete food chain starting with raw materials at farm level. It follows the processes they undergo in production, transportation and storage of the finished products in retailers as well as private transport and storage in the homes of consumers. Finally, it ends up with the expiring date, the end of shelf life or the preparation and ingestion by consumers. Therefore, especially the oral route of exposure to consumers, but also the inhalation and dermal routes of exposure of users are quantified. To reach this aim, the present work names some microbiological databases of food safety and public health. For both, microbiological and chemical risk assessment, the basic metrics as well as the most common modelling systems are shortly presented. Model systems on the microbiological growth and decline of pathogens include the PMP and ComBase models as well as SymPrevius. For chemicals, these are the Benchmark Dose modeling, ConsExpo and PRIMo databases as well as the PROAST model for statisticians. Moreover, two models are referenced, one a practical tool within expectable knowledge of managers, the other a complex risk assessment based tool for specialists. Both models intend to rank different classes of risks, e.g. chemical and microbiological risks. Nowadays, the mode of action of chemical substances is focused. Such modern methodologies will move toxicology towards a mechanistic understanding, but a lot of research remains to be done.

Literature

1 Cole, M. (2004). Food safety objectives – Concept and current status. Mitt. Lebensm. Hyg. 95, 13-20.
2 World Health Organization. 2002. Global surveillance of foodborne disease: developing a strategy and its interaction with risk analysis. Cited in: Fares, A. (2007). Quantitative risk assessment model of human salmonellosis linked to the consumption of Camembert cheese made from raw milk. Thesis, ENVA, UEAR, Unité d'épidémiologie et d'analyse des risques, F-94700 Maisons-Alfort (available at: http://pastel.archives-ouvertes.fr/docs/00/50/07/56/PDF/Fares_Thesis_PDF.pdf).
3 Centers for Disease Control and Prevention 1600 Clifton Rd. Atlanta, GA 30333, USA (available at: http://www.cdc.gov/foodnet).
4 European Center for disease prevention and control (ECDC) (available at: http://www.ecdc.europa.eu/en/activities/surveillance/Pages/index.aspx).
5 European Food Safety Authority (available at: http://www.efsa.europa.eu/en/networks.htm).
6 US-CDC (pulseNet) (available at: http://www.cdc.gov/pulsenet).
7 WHO Global Foodborne Infections network (available at: http://www.who.int/gfn/en/).
8 Public Health Agency of Canada, 130 Colonnade Road A.L. 6501H, Ottawa, Ontario K1A 0K9 (available at: http://www.phac-aspc.gc.ca/surveillance-eng.php).
9 Institut de veille sanitaire, 12, rue du Val d'Osne, 94415 Saint-Maurice cedex, France (available at: http://www.invs.sante.fr).
10 USDA-ARC Eastern Regional Research Center`s Pathogen Modeling Program (available at: http://pmp.arserrc.gov/PMPOnline.aspx).
11 ComBase Consortium`s ComBase (available at: http://www.combase.cc/index.php/en/).
12 Cellule Opérationnelle SymPrevius Creach Gwen - 29196 QUIMPER Cedex - FRANCE (available at: http://www.symprevius.net/).
13 Food and Drug Administration Center for Food Safety and Applied Nutrition (FDA/CFSAN), Joint Institute for Food Safety and Applied Nutrition (JIFSAN) and Risk Sciences International (RSI). 2012. FDA-iRISK version 1.0. FDA CFSAN. College Park, Maryland (available at: http://irisk.foodrisk.org/. http://irisk.foodrisk.org/ .
14 Mühlemann, M. Practitioner framework for the evaluation and prioritization of food and feed safety hazards and related research needs. ALP science. (545), 2013, 1-12 (available at: http://www.agroscope.admin.ch/publikationen/suche/index.html?sb_pubsearch=1&pubkeywords=&pubautor=m%C3%BChlemann&pubjahrvon=&pubjahrbis=&pubtyp=&pubsprache=&lang=de&pubsuche=Suchen).
15 Larsen, J. Chr. (2006). Risk assessment of chemicals in European traditional Foods. Trends in Food Science & Technology 17, 471 – 481 (available at: http://www.agronavigator.cz/userfiles/File/Agronavigator/Kvasnickova/tradice_1.pdf).
16 Lampen, A. (2010). Lebensmittel. In: Vohr, H. W. (ed.) Toxikologie Band 1, pp 377 - 406. Wiley-VCH: Weinheim.
17 U.S. Environmental Protection Agency, Office of the Science Advisor (8105R), 1200 Pennsylvania Avenue, NW Washington, DC 20460 . Benchmark Dose Software (available at: http://www.epa.gov/raf/publications/benchmarkdose.htm).
18 European Food Safety Authority; 2011. Use of BMDS and PROAST software packages by EFSA Scientific Panels and Units for applying the Benchmark Dose (BMD) approach in risk assessment. EN-113. [190 pp.].
19 The National Institute for Public Health and the Environment of the Netherlands (RIVM) software for dose-response modelling and benchmark dose analysis (PROAST) (available at: http://www.rivm.nl/en/foodnutritionandwater/foodsafety/proast.jsp).
20 EFSA, MRL-team, EFSA calculation model Pesticide Residue Intake Model "PRIMo" revision 2, (available at: http://www.efsa.europa.eu/de/mrls/docs/calculationacutechronic_2.xls).
21 The National Institute for Public Health and the Environment (RIVM) (The Consumer exposure to chemical substances (i.e. an overview of Exposure models (available at: http://www.rivm.nl/en/Topics/C/Consumer_exposure_to_chemical_substances/Exposure_models; the ConsExpo Model itself at: http://www.rivm.nl/en/Documents_and_publications/Scientific/Models/Download_page_for_ConsExpo_software : http://www.rivm.nl/en/Topics/Topics/C/ConsExpo).
22 EFSA AGREEMENT NUMBER EFSA/PPR/2007/01 FINAL REPORT, Prepared by the Pesticides Safety Directorate, UK (Paul Hamey, Neil Byron, Louise Hanley, Wendy Leslie & Neil Morgan) in collaboration with Ghent University, BE (Walter Steurbaut, Eline de Backer & Sofie Vergucht) (available at: http://www.efsa.europa.eu/en/scdocs/doc/26e.pdf).
23 European Food Safety Authority, 2014. Modern methodologies and tools for human hazard assessment of chemicals. EFSA Journal 2014;12(4):3638, 87 pp. doi:10.2903/j.efsa.2014.3638 (available at: www.efsa.europa.eu/efsajournal).