meat technology
Founder and publisher: Institute of Meat Hygiene and Technology, Belgrade

UDK: 579.67
ID: 225206284

Review paper

Predicting microbial growth: Theory and Application

Vesna V. Jankovic[1], Ivan Nastasijevic[1], Brankica Lakicevic[1]

A b s t r a c t: Predictive microbiology is a recent area within food microbiology, which studies the responses of microorganisms in foods to environmental factors (e.g. temperature, pH, NaCl) through mathematical functions. These functions enable scientists to predict the behavior of pathogens and spoilage microorganisms under different combinations of factors. Predictive microbiology models have immediate practical applications to improve microbial food safety and quality, and are leading to the development of a quantitative understanding of the microbial ecology of foods. Predictive models in foods have developed significantly in the last 20 years due to the emergence of powerful computational resources and sophisticated statistical packages.

Modeling microbial responses in food requires the interdisciplinary collaboration of food microbiologists and mathematicians; food technologists and computing scientists; molecular microbiologists and statisticians.

Key words: *predictive microbiology, microbial ecology of food, food safety and quality.*

Introduction

Predictive microbiology – or the quantitative microbial ecology of foods – represents a proactive approach to food quality and safety by accumulating information on bacterial responses related to intrinsic and extrinsic factors characterizing the food and its environment and summarizing the responses in databases and mathematical models (*Bjerre*, 2014; *McMeekin et al*., 1997; *McMeekin and Ross*, 2002; *Toldra*, 2009).

Predictive microbiology in foods is a research area within food microbiology intended to provide mathematical models to predict microbial behavior in food environments (*Fakruddin et al.*, 2011). Although the first predictive models date to the beginning of the 20th century, rapid development has occurred in recent decades as a result of computer software advances. In addition to exhaustive knowledge of food microbiology, the predictive microbiology field is based on important mathematical and modeling concepts that should be previously introduced for predictive microbiology beginners (*McMeekin et al.*, 1997).

The different typology of predictive models allows the prediction of growth, inactivation, or the probability of bacterial growth in foods under different environmental conditions and considering additional factors such as the physiological state of cells or interaction with other microorganisms. Nowadays, predictive models have become a necessary tool, allowing rapid responses to specific questions. Predictive models allow the estimation of the shelf-life of foods, define critical points in the production and distribution process and can give insight on how environmental variables affect the behavior of pathogenic or spoilage bacteria. Furthermore, predictive models have been incorporated as helpful elements into the self-control systems such as Hazard Analysis for Critical Control Point (HACCP) programs and food safety risk-based metrics. National and international food safety policies are now based on the development of Quantitative Microbial Risk Assessments studies, which is greatly supported but at the same time is turning into an important tool for improving food safety and quality (*Fakruddin et al*., 2011; *Perez-Rodriguez and Valero*, 2013). Microorganisms of interest are foodborne pathogens such as *E. coli* O157:H7, *Listeria monocytogenes*, *Salmonella* spp., *Clostridium botulinum* and spoilage microorganisms such as *Enterococcus* spp., *Pseudomonas* spp. and *Enterobacter* spp. *(Jankovic et al.,* 2013; 2014; 2015; *Lakicevic et al.*, 2014; 2015; *Nastasijevic*, 2011; *Nastasijevic*, 2014; *Nastasijevic et al.*, 2014).

History

The origin of predictive microbiology, as pointed out by Perez-Rodriguez and Valero (2013) is often linked to the works by *Bigelow et al.* (1920),

[1]*Institute of Meat Hygiene and Technology, Kacanskog 13, 11000 Belgrade, Republic of Serbia.*

Corresponding author: Vesna Jankovic, vessna@inmesbgd.com

Bigelow (1921) and *Esty and Meyer* (1922), in which a log linear model was proposed to describe bacterial death kinetics by heat. Their model found wide application in the food industry, and especially in the canning industry. Indeed, nowadays, these results are still applied by the food industry to reduce *Clostridium botulinum* in "low acid" canned foods. This model simply says that at a given temperature, the relative (or specific) death rate of the bacteria is constant with time. In other words, the percentage of the cell population inactivated in a unit time is constant. This is a simple, logical and understandable model, similar to those commonly used in physical and chemical sciences for processes such as dissipation, diffusion, etc., when the force that causes the decrease of a certain quantity is constant with time (*Baranyi et al.*, 1994; 2004). A step forward was taken by *Scott* (1936), who investigated how the specific death rate depended on the available water, quantified today by the so-called water activity, a dimensionless number between 0 (dry) and 1 (wet). He subsequently studied the effect of the temperature on the specific microbial death rate. Modeling microbial growth was also being done in the field of industrial microbiology (*Monod*, 1949). During the 1960s and 1970s, several efforts were devoted to apply mathematical models to inactivation of pathogens (*Clostridium botulinum* and *Staphylococcus aureus*) and growth of spoilage bacteria (*Nixon*, 1971; *Spencer and Baines*, 1964). Nonetheless, the great development of predictive microbiology started during the 1980s when computers and specific software facilitated the development of more complex and precise models. The term "predictive" microbiology, which is relatively recent, was coined by *Roberts and Jarvis* (1983), establishing the conceptual basis of modern predictive microbiology (*Brul et al.*, 2008). In the first book on the subject, *McMeekin et al.* (1993) defined it as a quantitative science that enables users to evaluate objectively the effect of processing, distribution and storage operations on the microbiological safety and quality of foods. *McMeekin et al.* (1993) suggested, as another possible explanation for the development of predictive microbiology, the marked increase in foodborne diseases during those years together with a major awareness of the limitations of the microbiological methods applied at that time. The scientific discipline of predictive microbiology aims to condense microbiological knowledge and mathematical techniques into mathematical models, capable of describing and predicting microbial growth in various environments, mostly related to food products (*Baranyi and Roberts*, 1994; *Ross and McMeekin*, 1994).

Development and limitations of predictive models

It is a general goal of food microbiologists to know in advance the behavior of microorganisms in foods under foreseeable conditions. To do so, exhaustive control of physicochemical factors that could influence microbial growth is needed (such as t, pH, a_w, salt, etc.), as well as in-depth knowledge about the biological characteristics of the target microorganisms (*Fakruddin et al.*, 2011; *Hajmeer and Cliver*, 2002).

The premise behind the scientific basis of predictive microbiology is that microbial responses in foods are reproducible when considered in the context of several extrinsic and intrinsic environmental factors (*Ross et al*, 2000). This behavior can be translated into diverse mathematical models that estimate microbial growth/inactivation/toxin production/probability of growth etc. This emerging area was redefined recently as modeling microbial responses in foods (*McMeekin et al.*, 2002).

Several authors suggested different classifications of predictive models based on their final purpose, the type of microorganisms to be studied, and their impact on food spoilage or food safety (*Roberts,* 1989; *Ross and McMeekin*, 2003; *van Boeckel*, 2008).

Basically, predictive models are split up into three groups: survival/inactivation models, boundary (growth/no growth) models and growth models. Basing on their development, models can be classified as follows (Figure 1):

1. Primary models: aim to describe the kinetics of a process with as few parameters as possible while being able to accurately define the growth and inactivation phases. They are represented as the increase/decrease in population density against time. Primary models developed in the 90s are still widely used, but are mainly empirical (*Baranyi and Roberts*,1994; *Buchanan et al.*, 1997; *Geeraerd et al*., 2000).
2. Secondary models: describe the effect of environmental conditions (physicochemical and biological factors) on the values of the parameters of a primary model. Most currently-used secondary models can be subdivided into four classes (*Adopted from Van Impe et al.* (2013): (i) square root models (*Ratkowsky et al.,* 1982; *Ratkowsky et al.,* 2003), (ii) cardinal parameter models (*Rosso et al.*, 1995; *Sautour et al.*, 2001), (iii) neural networks (*Geeraerd et al*.,1998; *Panagou et al.*, 2007), and (iv) response surface models (*Baranyi et al.*, 1996; *Geeraerd et al.*, 2004). Secondary

models used for describing the effect of environmental conditions on microbial growth include (*McKellar and Lu*, 2004): Arrhenius-type models, Belehradek type models, models based on the gamma concept, cardinal parameter models, and polynomial models.

3. Tertiary models: based on computer software programs that provide an interface between the underlying mathematics and the user, allowing model inputs to be entered and estimates to be observed through simplified graphical outputs. *Whiting and Buchanan* (1997) called the foregoing integrated software-based model "tertiary models". Tertiary models are the application of the aforementioned models, included in user-friendly software in order that they can be used without additional modeling; moreover, nonmodelers can use these tertiary models (*Fakruddin et al*., 2011). One or more of the primary and secondary models are compiled to provide a prediction, generally coupled with databases gathering the input parameters such as cardinal values, optimal growth rate, and so on required for running the simulation (*McDonald and Sun*, 1999). Usually, various factors can be specified, such as temperature, a_w, pH, NaCl concentration, and so on. All of these input parameters used in tertiary models were previously validated in primary and/or secondary models. One of the main uses of such software is in product development, since they let the user examine the effect of formulation changes on the safety of the product without costly pilot plant trials. The software packages that are available include: ComBase (www. combase.cc), Sym'Previus (www.symprevius.net), USDA pathogen (http://www.ars.usda.gov), Food Spoilage and Safety Predictor, (www.fssp.dtu.dk).

Predictive microbiological models are normally developed assuming that microbial responses are consistent (*McMeekin et al*., 2002, 2010b; *McMeekin*, 2007; *Mejlholm et al*., 2010). While predictive models can provide a cost-effective means to minimize microbiological testing in determining shelf-life, there may be occasions when the model's predictions may not be accurate, due to inconsistent microbial responses and variations in the growth media (*DVFA,* 2014; *EC*, 2005). Finally, models cannot be applied if a validation process is not previously accomplished, which typically consists of confirming the predictions experimentally, using a quantitative method. The validation process is conducted considering biological knowledge of the system and statistical tools. Once models are validated and users are aware of the limitations of the models, they are useful tools to obtain information and make

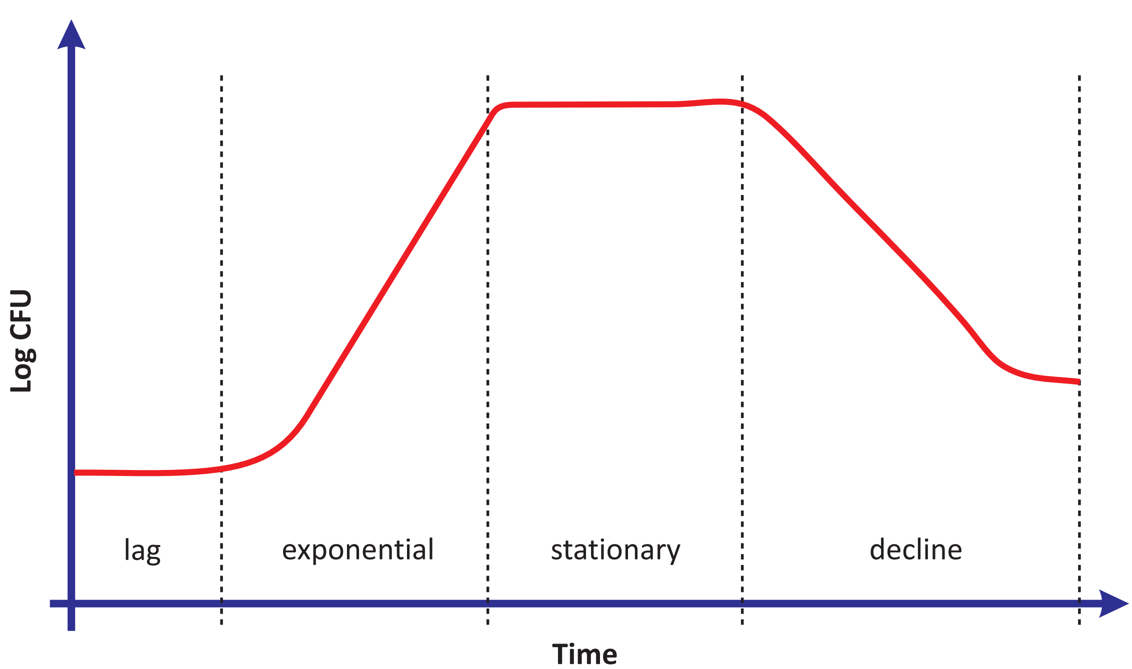

Figure 1. Theoretical presentation of the bacterial growth curve with four phases: (i) lag phase, (ii) exponential growth phase, (iii) stationary phase and (iv) decline (*Perez-Rodriguez and Valero*, 2013)

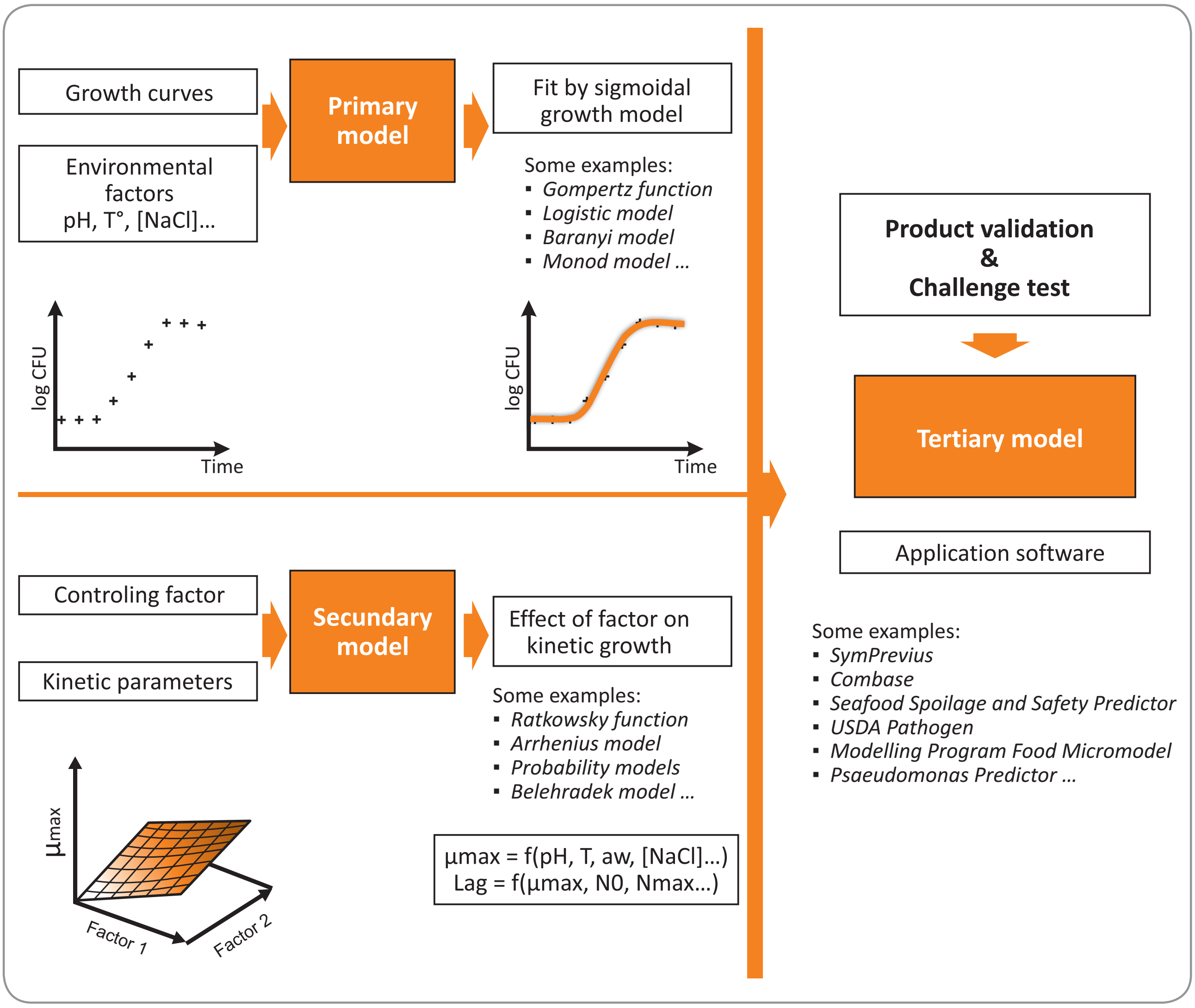

Figure 2. Schematic development, classification, and some examples of predictive microbiology models in food products (Adopted from *Fakruddin et al.*, 2011; *McDonald and Sun,* 1999).

decisions for the following situations (*Alzamora et al*., 2000a; *Buchanan and Whiting*, 1997):

1. **Prediction of safety**: Estimate the risk of growth or survival of pathogens during food processing.
2. **Quality control**: Improve systems like HACCP (Hazard Analysis of Critical Control Points) to ensure food safety.
3. **Product development**: Redesign processes and recipes, set priorities in product design and evaluation.
4. **Data analysis and laboratory planning**: The model could save resources, time, and money.
5. **Risk assessment models**: Evaluate the probability that a food could cause foodborne illness

Microbial modeling and applications of predictive microbiology

Microbial modeling

In all predictive microbiology, a prediction must only be used as a guide to the response of microorganism(s) to a particular set of environmental conditions (pH, a_w, t). However, food businesses should never rely solely on any predictive microbiological model to determine the safety of foods and/or processing systems (*Toldra*, 2009). Determining the growth, survival or inactivation of pathogens in food requires (*FDA*, 2015):

1. The determination of the intrinsic and extrinsic properties of the product, taking into account the storage and processing conditions,

the possibilities for contamination and the foreseen shelf-life.

2. Consultation of available scientific literature and research data regarding the survival, growth and inactivation of microorganisms of concern.
3. Where necessary on the basis of these studies food businesses should also conduct additional studies which may include:
4. Laboratory-based microbiological sampling and analysis.
5. Predictive microbiological modeling.
6. Challenge tests to investigate the ability of microorganisms of concern to grow or survive in the food product under reasonably foreseeable conditions of distribution and storage (no challenge testing for Campylobacter spp., Shigella spp. and Yersinia enterocolitica are recommended because other organisms, such as *Salmonella*, have similar routes of contamination and are easier to culture and have less fastidious growth and survival requirements (*FDA*, 2015).
7. Predictive microbiological models are also useful when the shelf-life has been determined, but the product is then subject to a minor process or formulation change (either planned or unplanned through loss of process control). A predictive microbiological model can then be used to initially establish if the change might have any effect on the safety and shelf-life of the product. Table 1 shows predicted pH limits for growth (p=0.5, 0.1, 0.01) of selected pathogens at various a_w and temperature conditions (EFSA, 2012).

Table 1. Predicted pH limits for growth (p = 0.5, 0.1, 0.01) of selected pathogens at various a_w and temperature conditions (*EFSA*, 2012)

Pathogen	a_w	Predicted pH limits at various aw, temperature and p-values											
		5°C P			10°C P			15°C P			25°C P		
		0.5	0.1	0.01	0.5	0.1	0.01	0.5	0.1	0.01	0.5	0.1	0.01
Listeria monocytogenes (Koutsoumanis et al., 2004)	0.99	4.76	4.69	4.61	4.45	4.39	4.34	4.29	4.24	4.19	4.23	4.19	4.14
	0.98	4.84	4.77	4.69	4.53	4.47	4.41	4.37	4.32	4.26	4.32	4.28	4.23
	0.97	4.96	4.87	4.79	4.62	4.56	4.49	4.46	4.41	4.35	4.43	4.38	4.33
	0.96	5.10	5.00	4.91	4.73	4.66	4.60	4.56	4.51	4.44	4.54	4.48	4.43
	0.95	5.28	5.16	5.05	4.86	4.78	4.71	4.68	4.61	4.55	4.66	4.60	4.54
Salmonella (Koutsoumanis et al., 2004)	0.99				4.66	4.41	4.37	4.40	4.18	4.14	3.95	3.91	3.87
	0.98				4.85	4.61	4.56	4.56	4.35	4.31	4.23	4.06	4.01
	0.97		–		5.04	4.80	4.74	4.71	4.51	4.45	4.45	4.18	4.13
	0.96				5.24	4.98	4.92	4.86	4.65	4.59	4.66	4.29	4.23
	0.95				5.47	5.17	5.10	5.02	4.80	4.73	4.88	4.39	4.33
Escherichia coli O157:H7 (Skandamis et al., 2007)	0.99				5.31	5.10	4.89	4.46	4.33	4.20	3.94	3.83	3.72
	0.98				5.16	5.02	4.88	4.63	4.51	4.38	4.03	3.92	3.80
	0.97		–		6.20	5.95	5.69	5.28	5.09	4.89	4.38	4.22	4.05
	0.96				–	–	–	–	6.69	6.06	5.01	4.74	4.48
	0.95				–	–	–	–	–	–	–	–	5.80
Bacillus cereus (Lanciotti et al., 2001)	0.99										4.85		
	0.98										4.92		
	0.97		–			–		–	–	–	5.10	–	–
	0.96										6.02		
	0.95										–		
Staphylococcus aureus (Lanciotti et al., 2001)	0.99							5.44	5.22	5.03	4.79	4.68	4.59
	0.98							5.64	5.39	5.16	4.89	4.77	4.65
	0.97		–			–		5.97	5.66	5.38	5.06	4.90	4.77
	0.96							6.66	6.24	5.85	5.40	5.19	5.00
	0.95							–	–	–	7.39	6.85	6.36

Table 2. Applications of predictive microbiology (according to *Fakruddin et al.*, 2011)

Area of Application	Example
Hazard Analysis Critical Control Point (HACCP)	Preliminary hazard analysis identification and establishment of critical control point(s) Corrective actions Assessment of importance of interaction between variables Risk assessment
Risk assessment	Estimation of changes in microbial numbers in a production chain Assessment of exposure to a particular pathogen
Microbial shelf life studies	Prediction of the growth of specific food spoilage microorganisms Prediction of growth of specific foodborne pathogens
Product research and development	Effect of altering product composition on food safety and spoilage Effect of processing on food safety and spoilage Evaluation of effect of out-of-specification circumstances
Temperature function integration and hygiene regulatory activity	Consequence of temperature in the cold chain for safety and spoilage
Education	Education on safety, especially non-technical people
Design of experiments	Number of samples to be prepared Defining suitable intervals between sampling

Applications of predictive microbiology

Some of the applications of predictive microbiology are listed in Table 2.

Limitations of predictive microbiology

Even though predictive microbiology models are widely used when correctly validated, they have several limitations because of the complexity of microbial behavior and food systems.

Mathematical models are simplifications of complex biochemical processes and in some cases, not every important variable or factor that affects the system is included in the model (*Alzamora et al*., 2005; *Buchanan and Whiting*, 1997). Usually, models are not designed for the same conditions in which microorganisms exist in food systems (biofilms, starved and unknown nutrients, among many others), since the majority of the data to generate the predictive models are derived from broth-based experiments. It is known that bacterial pathogens are more resistant in real food products than in broth cultures (*Alzamora et al*., 2005). Most of the models describe changes of microbial behavior for homogeneous populations; nevertheless, competition among microorganisms affects the food environment, and models do not account for this (*Lebert and Lebert*, 2006).

Some models make a good description of linear relationships; but when more than one factor is involved, reparameterization of the model becomes necessary.

It is important the model developer clearly specify directly or through the model what the limits of the model are, i.e., what microorganisms, what factors, what ranges of each factor and what combinations of factors will give valid answers. The presence of additional inhibitory factors in a food, and which have not been included in the model, invalidates the model or requires caution to be used to interpret the predictions. Currently, growth models do not usually include factors such as anion effects from the acidulent used, phosphates, sorbates, and bacteriocins, and humectants other than sodium chloride. No broth models include competition from other microorganisms. Some models developed with foods include the “normal” spoilage microbiota, but how this microbiota changes in species and number with plant or season and the effect upon the modeled microorganism is largely unknown (*Van Impe et al.*, 2013).

Because pathogens grow in most foods, the important question, then, is whether the pathogens will grow to a significant population before the spoilage microbiota causes the food to be rejected by the consumer. There is a need for systematic modeling of representative classes of spoilage microorganisms so tertiary models can then plot comparative growth curves for both pathogenic and spoilage organisms. For some pathogens with very low infective or toxic dose, such as *Listeria*, *Yersinia* and *C. botulinum*, the criteria may be growth-no growth and the spoilage flora has little significance unless they alter the environment by lowering the pH or produce a bacteriocin (*Fakrudin*, 2011).

Risk analysis and predictive microbiology

Management of foodborne threats is an ongoing challenge due to changes in primary and secondary production, microbial adaption, increases in international trade, changes in consumer demands and behavioral and demographic changes. Risk analysis has been introduced as a means to face these challenges and to evaluate and control microbial risks (*Bjerre*, 2014). Risk analysis includes three components; (i) risk assessment, (ii) risk management and (iii) risk communication (CAC/GL 63- 2007). Risk assessment is the scientific evaluation of known or potential adverse health effects of a food product and comprises: hazard identification, hazard characterization, exposure assessment and risk characterization (*Marvin et al*., 2009). The outcome of the risk characterization is an estimate of the likelihood of adverse health effects in the population due to exposure to the hazard in question (*FAO/WHO*, 1995). In a quantitative microbiological risk assessment, the exposure assessment describes the routes by which the microbiological hazard can be introduced, distributed and altered during the production, distribution and consumption of a given food product (*WHO/FAO*, 2004).

Predictive microbiology is of particular interest in relation to evaluation of alterations in numbers (increase or decrease) of the hazard over time. For quantitative risk analyses, it is often stated that data is lacking and available data often originate from modeling experiments with, for example, unrealistically high initial bacterial numbers. In general, high quality, relevant and timely data is lacking (*Gardner*, 2004; *Ross and Sumner*, 2002; *WHO/FAO*, 2004). In spite of that, as a means to provide information and to fill data gaps, predictive models for growth and inactivation can be helpful and efficient tools. Predictive models, successfully validated in growth environments comparable to the products of concern, can be used to predict the effect of intrinsic and extrinsic factors on the response of the pathogen in question (*Toldra*, 2009). This quantification is important since the effects of both spoilage and pathogenic microorganisms are highly correlated to the number of microbes present in the food product at the point of consumption (*Bjerre*, 2014).

Future perspectives

Foods are complex feedback systems. Generally, substantial quantities of data have been derived from modeling studies conducted under experimental conditions, but this data is often not immediately relevant to real-life conditions for the pathogen, the food vehicle or the consumer. Mathematical models are able to bridge some gaps but are also an approximation of reality. Evidently, modelers need to be diligent when relating and extrapolating data, and using or interpreting mathematical growth models and their outcomes, particularly when conducting exposure assessment and hazard characterization, as these impact on the validity of a risk characterization. Additionally, estimation of pathogen prevalence and level (number) in food products is key for exposure assessment and indispensable for the generation of reliable risk estimates (*ILSI*, 2010). Thus, risk assessors require an understanding of the biology and ecology of the pathogen(s), and of the properties of food materials they investigate. Often, dose-response models are the element where least information is available. Risk assessments should include an integral evaluation of the quality of data and models that are included and this is often accomplished by including an explicit evaluation concerning uncertainty and variability in the risk characterization outcome.

Besides a move towards stochastic modeling approaches, other subjects are also forecasted to be a part of the future of predictive microbiology. In 2004, Bernaerts and co-workers strongly advocated for the development of more mechanistically-inspired predictive models in order to obtain a better understanding of the underlying mechanisms, but also to develop more robust models (*Bernaerts et al*., 2004). *McMeekin et al*. (2010) suggested focusing on the ecophysiology of foodborne pathogens and to model growth responses from, for example, thermodynamics. The introduction of systems biology into predictive microbiology has been suggested by *Brul et al*. (2008) and *Van Impe et al*. (2013) in order to apply “bottom-up” approaches and to work at the microscopic level e.g. by developing metabolic network-based modeling approaches. Belief in systems-biology as an integrated part of predictive microbiology has also been expressed by *McMeekin et al*. (2013) in order to induce a shift from empirical predictive microbiology towards mechanistic predictive systems biology models. These new, emerging approaches within predictive microbiology should be considered and, if obtainable, tested when developing new models.

References

Alzamora, S. M., López-Malo, A. & Tapia de Daza, M. S. (2000a). Overview, in: *Minimally processed fruits and vegetables-fundamental aspects and applications*. Alzamora S. M., Tapia M. S., López-Malo A., Eds. Aspen Publishers Inc. Gaithersburg, MD, USA, 1–9.

Alzamora, S. M., Guerrero, S., Viollaz P. & Welti-Chanes, J. (2005). Experimental protocols for modeling the response of microbial populations exposed to emerging technologies: some points of concern, in: *Novel food processing technologies*. Barbosa-Cánovas G., Tapia M. S., Cano P. M., Eds. CRC Press. Boca Ratón, FL, USA.

Baranyi, J. & Roberts, T. A. (1994). A dynamic approach to predicting bacterial growth in food. *International Journal of Food Microbiology, 23, 3–4*, 277–294.

Baranyi, J. & Pin, C. (2004). Modeling the history effect on microbial growth and survival, in: *Modeling microbial responses in food. McKellar R., Lu X., Eds. Contemporary Food Science*. CRC Press. Boca Ratón, FL, USA, 285–301.

Baranyi, J., Pin, C. & Ross, T. (1999). Validating and comparing predictive models. *International Journal of Food Microbiology, 48*, 159–166.

Baranyi, J., Ross, T., McMeekin, T. A. & Roberts, T. A. (1996). Effects of parameterization on the performance of empirical models used in 'predictive microbiology'. *Food Microbiology, 13*, 83–91.

Bernaerts, K., Dens, E., Vereecken, K., Geeraerd, A.H., Standaert, A.R., Devlieghere, F., Debevere, J. & Van Impe, J.F. (2004). Concepts and tools for predictive modeling of microbial dynamics. *Journal of Food Protection, 67*, 2041–2052.

Bigelow, W. D. (1921). The logarithmic nature of thermal death time curves. *Journal of Infection Diseases, 29*, 528–536.

Bigelow, W. D., Bohart, G. S., Richardson, A.C. & Ball, C.O. (1920). Heat penetration in processing canned foods. *National Canners Association Bulletin. 16–L.*

Bjerre, N. (2014). *Food microbiology: new tools for risk assessments and diary product development*. PhD thesis, Technical University of Denmark, 40–43.

Brul, S., Mensonides, F. I. C., Hellingwerf, K. J. & Teixeira de Mattos, M. J. (2008). Microbial systems biology: new frontiers open to predictive microbiology. *International Journal of Food Microbiology*, 128, 16–21.

Buchanan, R. L., Whiting, R. C. & Damert, W. C. (1997). When is simple good enough: a comparison of the Gompertz, Baranyi, and three phase linear models for fitting bacterial growth curves. *Food Microbiology, 14*, 313–326.

CAC/GL 63-2007 (2007). Principles and guidelines for the conduct of microbial risk management (MRM). *Codex Alimentarius*, 1–19.

DVFA (2014). Danish Veterinary and Food Administration. *Listeria monocytogenes*. http://www.altomkost.dk/Fakta/Foedevarer/maelk_og_ost/forside.htm (accessed 6.4.14).

EC (2005). Commission Regulation (EC) No 2073/2005 of 15 November 2005 on microbiological criteria for foodstuffs. *Official Journal of the European Union 338*, 1–26.

EFSA (2012). Scientific opinion on public health risks represented by certain composite products containing food of animal origin. *EFSA Journal, 10, 5*, 2662.

Esty, J. R. & Meyer, K. F. (1922). The heat resistance of the spore of *Bacillus botulinus* and allied anaerobes, XI. *Journal of Infection Disease, 3*, 650–663.

Perez-Rodriguez, F. & Valero, A. (2013). Predictive microbiology in Foods, Springer Briefs in Food, *Health and Nutrition, 5*.

Fakruddin, M., Mohammad, R., Khanjada, S. & Bin Mannan (2011). Predictive microbiology: modeling microbial responses in food. *Ceylon Journal of Science (Bio. Sci.), 40, 2*, 121–131.

FAO/WHO (1995). Application of risk analysis to food standards issues. *Report of the Joint FAO/WHO Expert Consulation, WHO/FNU/FOS*. World Health Organization, Geneva.

FDA (2015). Evaluation and definition of potentially hazardous foods – Chapter 6. Microbiological challenge testing. www.fda.gov

Gardner, I. A. (2004). An epidemiologic critique of current microbial risk assessment practices: the importance of prevalence and test accuracy data. *Journal of Food Protection, 67*, 2000–2007.

Geeraerd, A. H., Herremans, C. H. & Van Impe, J. F. (2000). Structural model requirements to describe microbial inactivation during a mild heat treatment. *International Journal of Food Microbiology*, 59, 185–209.

Geeraerd, A. H., Valdramidis, V. P., Devlieghere, F., Bernaert, H., Debevere, J. & Van Impe, J. F. (2004). Development of a novel approach for secondary modelling in predictive microbiology: incorporation of microbiological knowledge in black box polynomial modelling. *International Journal of Food Microbiology, 91*, 229–244.

Geeraerd, A., Herremans, C., Cenens, C. & Van Impe, J. (1998). Application of artificial neural networks as a non-linear modular modeling technique to describe bacterial growth in chilled food products. *International Journal of Food Microbiology, 44*, 49–68.

Hajmeer, M. N. & Cliver, D. O. (2002). Microbiology of food preservation and sanitation, in: *Foodborne diseases*. Cliver D. O., Riemann H., Eds. Academic Press. New York, NY, USA, 330–352.

ILSI (2010). Impact of microbial distributions on food safety. ILSI Europe risk analysis in food microbiology task force, Brussels, Belgium. Report. www.ilsi.org

Van Impe, J. F., Vercammen, D. & Van Derlinden, E. (2013). Toward a next generation of predictive models: a systems biology primer. *Food Control, 29*, 336–342.

Jankovic, V., Lakicevic, B., Dzinic, N., Tasic, T., Ikonic P. & Saric, Lj. (2015). Microbiological profile of Petrovská klobása made from warm meat in the traditional way depending on the method of packaging. *Procedia Food Science, 5*, 117–120.

Jankovic, V., Lakicevic, B., Ikonic, P., Tasic, T., Borovic, B., Veskovic, S. & Saric, Lj. (2014). *Enterobacteriaceae* presence during fermentation of traditional dry fermented sausage – petrovská klobása. Proceedings of the II International Congress of Food Technology, Quality and Safety, Novi Sad, Serbia, 202–208.

Jankovic, V., Petrovic, Lj., Lakicevic, B., Matekalo Sverak, V., Lilic, S. & Mitrovic, R. (2013). Determination of typical house flora during production process of the Petrovská klobása. *African Journal of Microbiology Research, 7*, 32, 4130–4137.

Koutsoumanis, K. P., Kendall, P. A. & Sofos, J. N. (2004). Modeling the boundaries of growth of *Salmonella* Typhimurium in broth as a function of temperature, water activity, and pH. *Journal of Food Protection, 67*, 53–59.

Lakicevic, B., Nastasijevic, I. & Raseta, M. (2015). Sources of *Listeria monocytogenes* contamination in retail establishments. *Procedia Food Science 5*, 160–163.

Lakicevic, B., Velebit, B., Jankovic, V., Spiric, D., Baltic, T., Mitrovic, R. & Babic, J. (2014). Taq Man real time PCR detection of *Listeria monocytogenes*: a study of enrichment incubation time affecting sensitivity in experimental dry fermented sausages. *Tehnologija Mesa, 55* (1), 60–65.

Lanciotti, R., Sinigaglia, M., Gardini, F., Vannini, L. & Guerzoni, M. E. (2001). Growth/no growth interfaces of *Bacillus cereus, Staphylococcus aureus* and *Salmonella Enteritidis* in model systems based on water activity, pH, temperature and ethanol concentration. Food Microbiology, 18, 659–668.

Lebert, A. & Lebert, I. (2006). Quantitative prediction of microbial behaviour during food processing using an integrated modeling approach: a review. *International Journal of Refrigeration, 29*, 968–984.

Lopez-Malo, A., Alzamora, S. M. & Palou, E. (2005). Aspergillus flavus growth in the presence of chemical preservatives and naturally occurring antimicrobial compounds. *International Journal of Food Microbiology*, 99, 119–128.

Marks, B. P. (2008). Status of microbial modeling in food process models. Comprehensive Reviews in Food Science and Food Safety, 7, 1, 137–143.

Marvin, H. J. P., Kleter, G. A., Frewer, L. J., Cope S., Wentholt, M. T. A. & Rowe, G. (2009). A working procedure for identifying emerging food safety issues at an early stage: implications for European and international risk management practices. *Food Control 20,* 345–356.

McDonald, K. & Sun ,D. W. (1999). Predictive food microbiology for the meat industry: a review. *International Journal of Food Microbiology*, 52, 1–27.

McKellar, R. & Lu, X. (2004). *Modeling Microbial Responses on Foods*. CRC Press. Boca Ratón, FL, USA.

McMeekin, T., Brow, J. & Kris, K. (1997). Quantitative microbiology: a basis for food safety. Emerging Infectious Diseases, 3, 541–549.

McMeekin, T. A. (2007). Predictive microbiology: quantitative science delivering quantifiable benefits to the meat industry and other food industries. *Meat Science, 77*, 17–27.

McMeekin, T. A., Olley, J. N., Ross, T. & Ratkowsky, D. A. (1993). Predictive Microbiology. John Wiley & Sons Ltd. Chichester, UK.

McMeekin, T. A. & Ross, T. (2002). Predictive microbiology: providing a knowledge-based framework for change management. *International Journal of Food Microbiology, 78*, 133–153.

McMeekin, T. A., Tamplin, M. L., Ross T. & Koseki, S. (2010b). The way forward with predictive microbiology in the dairy industry. *Australian Journal of Dairy Technology, 65*, 91–97.

McMeekin, T., Olley, J., Ratkowsky, D., Corkrey, R. & Ross, T. (2013). Predictive microbiology theory and application: is it all about rates? *Food Control, 29*, 290–299.

Mejlholm, O., Gunvig A., Borggaard C., Blom-Hanssen, J., Mellefont L., Ross T., Leroi F., Else, T. Visser D. & Dalgaard, P. (2010). Predicting growth rates and growth boundary of *Listeria monocytogenes* – an international validation study with focus on processed and ready-to-eat meat and seafood. *International Journal of Food Microbiology 141*, 137–150.

Monod, J. (1949). The growth of bacterial cultures. *Annual Review of Microbiology, 3*, 371–394.

Nastasijevic, I. (2011). STEC O157 in the beef chain – risk assessment and management. CAB Reviews: Perspectives in Agriculture, Veterinary Science, Nutrition and Natural Resources 6, 061, 1–19 (Animal Science Reviews ISSN 1749-8848, 219–239).

Nastasijevic, I. (2014). Verotoxigenic *Escherichia coli* in the meat chain and control strategies. 12th International Conference on Fundamental and Applied Aspects of Physical Chemistry, 4th Workshop: Specific methods for food safety and quality, September 23rd, Belgrade, Serbia, Proceedings, 1–4.

Nastasijevic, I., Mitrovic, R. & Jankovic, V. (2014). STEC in the beef chain – one health approach. *Tehnologija Mesa, 55, 2*, 93–116.

Nixon, P. A. (1971). Report on quality of fish products. Semin. 3, Fish. Ind. Board, Wellington, New Zealand, 34.

Panagou, E. Z., Tassou, C. C., Saravanos, E. K. A. & Nychas, G. J. E. (2007). Application of neural networks to simulate the growth profile of lactic acid bacteria in green olive fermentation. *Journal of Food Protection, 70*, 1909–1916.

Pérez-Rodríguez, F. & Valero, A. (2013). Predictive Microbiology in Foods. Springer Briefs in Food, Health, and Nutrition. Springer, New York, USA. Volume 5, p. 128. ISBN: 978-1-4614-5519-6.

Ratkowsky, D. A., Olley, J., McMeekin, T. A. & Ball, A. (1982). Relationship between temperature and growth rate of bacterial cultures. *Journal of Bacteriology, 149*, 1–5.

Roberts, T. A. (1989). Combinations of antimicrobials and processing methods. *Food Technology, 43*, 156–163.

Roberts, T. A. & Jarvis, B. (1983). Predictive modeling of food safety with particular reference to *Clostridium botulinum* in model cured meat systems, in: *Food microbiology: advances and prospects*. Roberts T.A., Skinner F. A., Eds. Academic Press. Orlando, FL, USA, 85–95.

Ross, T., Dalgaard P. & Tienungoon, S. (2000). Predictive modelling of the growth and survival of *Listeria* in fishery products. *International Journal of Food Microbiology 62*, 231–245.

Ross, T. & McMeekin, T. A. (1994). Predictive microbiology. *International Journal of Food Microbiology 23*, 241–264.

Ross, T. & McMeekin, T. A. (2003). Modeling microbial growth within food safety risk assessments. Risk Analysis 23, 179–197.

Ross, T., Ratkowsky, D. A., Mellefont, L. A. & McMeekin, T. A. (2003). Modelling the effects of temperature, water activity, pH and lactic acid concentration on the growth rate of *Escherichia coli. International Journal of Food Microbiology*, 82, 33–43.

Ross, T. & Sumner, J. (2002). A simple, spreadsheet-based, food safety risk assessment tool. *International Journal of Food Microbiology 77*, 39–53.

Rosso, L., Lobry, J. R., Bajard, S. & Flandrois, J. P. (1995). Convenient model to describe the combined effects of temperature and pH on microbial growth. *Applied and Environmental Microbiology, 61*, 610–616.

Sautour, M., Dantigny, P., Divies, C. & Bensoussan, M. (2001). A temperature-type model for describing the relationship between fungal growth and water activity. *International Journal of Food Microbiology, 67*, 63–69.

Scott, W. J. (1936). The growth of microorganisms. The influence of water content of substrate on rate of growth at –1°C. *Journal of the Council for Scientific and Industrial Research (Australia), 9*, 177–182.

Skandamis, P. N., Stopforth, J. D., Kendall, P. A., Belk, K. E., Scanga, J. A., Smith, G. C. & Sofos, J. N. (2007). Modeling the effect of inoculum size and acid adaptation on growth/no growth interface of *Escherichia coli* O157:H7. *International Journal of Food Microbiology, 120*, 237–249.

Spencer, R. & Baines, C. R. (1964). The effect of temperature on spoilage of wet white fish. Food Technology, 18, 769–773.

Toldra, F. (2009). *Safety of meat and processed meat*, Marie Laure Delignette-Muller, 535–541.

Van Boeckel, M. (2008). Kinetics of microbial growth, in: Kinetic modelling of reactions in foods. CRC Press. Boca Ratón, FL, USA, 12–1; 12–40.

Van Impe, J. F., Poschet, F., Geeraerd, A. H. & Vereecken, K. M. (2005). Towards a novel class of predictive microbial growth models. *International Journal of Food Microbiology, 100*, 97–105.

Van Impe, J. F., Vercammen, D. & Van Derlinden, E. (2013). Toward a next generation of predictive models: a systems biology primer. *Food Control 29*, 336–342.

Whiting, R. C. & Buchanan, R. L. (1997). Predictive modelling, in: Food microbiology – fundamentals and frontiers. Doyle M. P., Beuchat L. R., Montville T. J., Eds. ASM Press. Washington, USA, 728–739.

WHO/FAO (2004). Risk assessment of *Listeria monocytogenes* in ready-to-eat foods. Technical Report. World Health Organization.

www.combase.cc

www.symprevius.net

www.ars.usda.gov

www.fssp.dtu.dk

Paper received: 20.05.2016.
Paper correction: 6.06.2016.
Paper accepted: 24.05.2016.