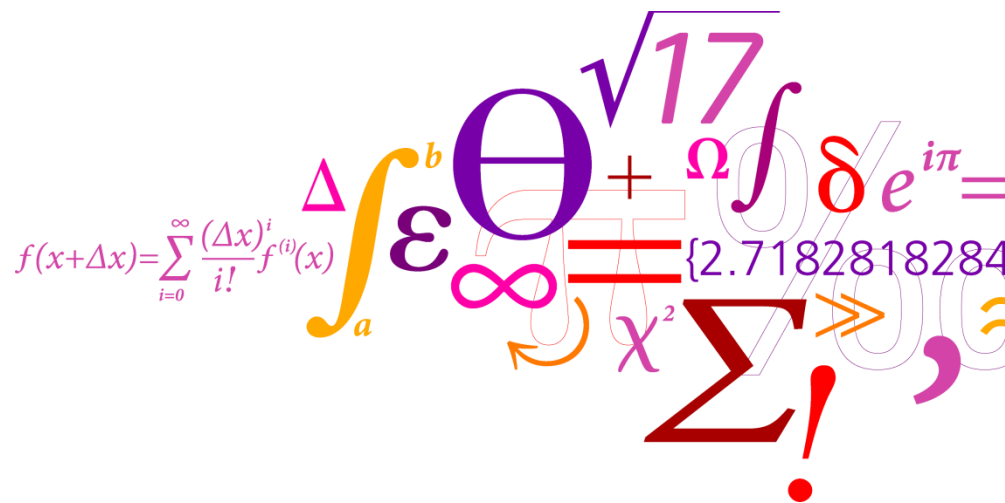# Predictive food microbiology

Tina Beck Hansen (tibha@food.dtu.dk)

- What is it?
- How is it done?
- What is it used for?

Predictive food microbiology

What is it?

Prediction of microbial behaviour in food environments

The idea is not new!

- 1920's – heat inactivation: D-, z- and F-values

- 1930's – Scott from Australia: … if we know the growth rates of the meat spoilage organisms, we can predict when meat is spoiled at different storage temperatures…

But it accelerated in 1980's....
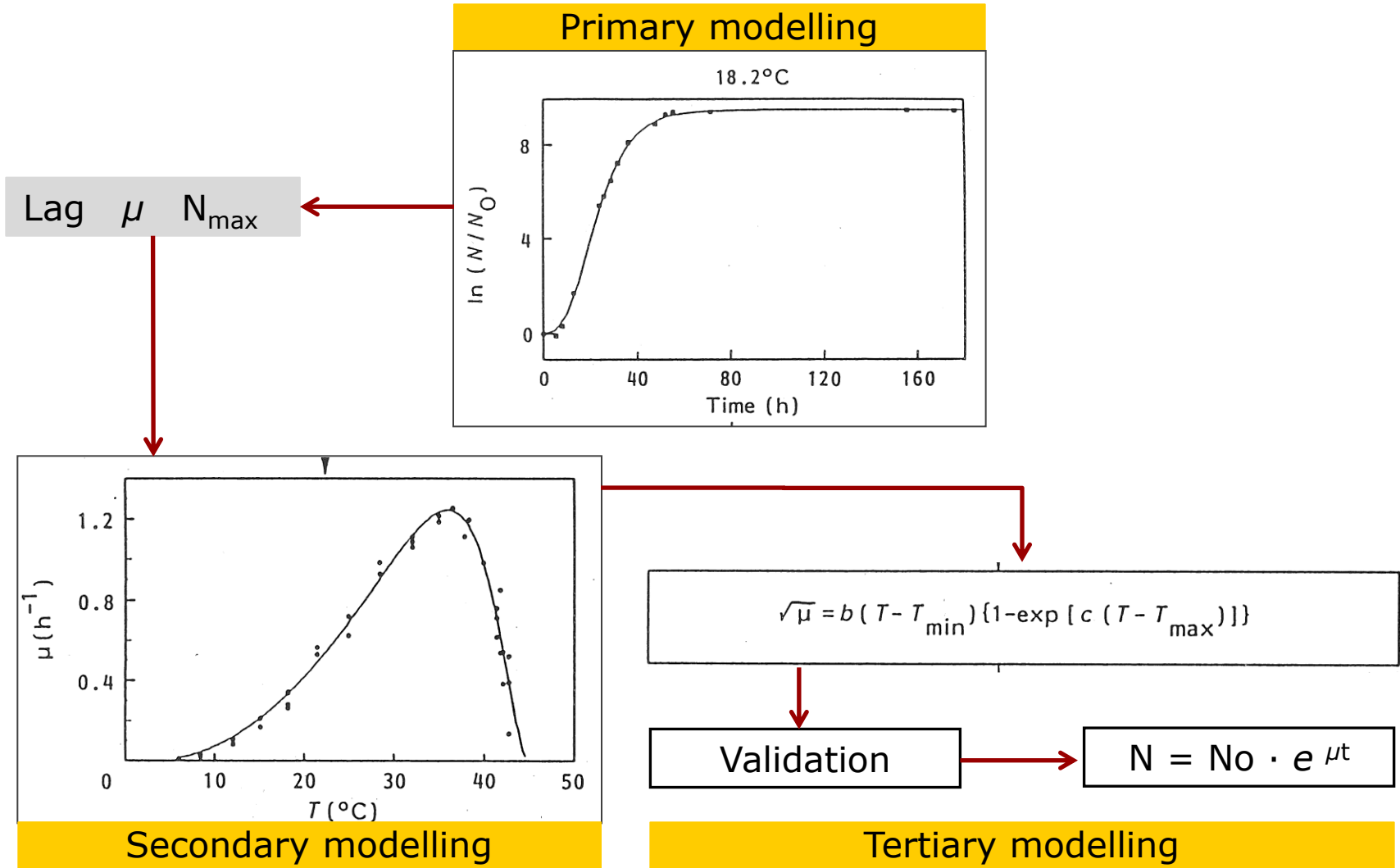
Roberts & Jarvis (1983):

- Growth, survival and inactivation of microorganisms in foods are reproducible responses

- A limited number of environmental parameters in foods determine the kinetic responses of microorganisms
    - Temperature
    - Water activity / salt-in-water
    - pH
    - Preservatives

- Mathematical models that quantitatively describe the combined effect of the environmental parameters can be used to **predict** growth, survival or inactivation of microorganisms

Predictive food microbiology

How is it done?

Using mathematical equations for description of kinetic responses of microorganisms in food

Procedure for development of models

Primary modelling

18.2°C

Lag μ N_{max}

Secondary modelling

Tertiary modelling

$$\sqrt{\mu} = b\,(\,T - T_{min}\,)\,\{1 - \exp[\,c\,(\,T - T_{max}\,)\,]\}$$

Validation

$N = No \cdot e^{\,\mu t}$

Model types

Model type	Response	Predictor	Examples
Primary model: *A function describing microbial response over time*	CFU Toxin formation Metabolic compounds Absorbance Impedance	Time	Exponential Logistic Gompertz Baranyi & Roberts Weibull
Secondary models: *An equation using parameter estimates from a primary model as response*	Growth rate Generation time Lag phase Max. population D-value Growth/no-growth	Time Environmental parameters	Polynomial Arrhenius Square root CPM-models ANN-models z-value Probability models

Validation

- Does the developed model work for real life situations?
- Comparison of observed and predicted values
- Graphical and/or mathematical

Secondary models
(growth rates, lag times)

Bias factor (B_f) =

$$10^{(\Sigma \log(pred/obs) / n)}$$

Accuracy factor (A_f) =

$$10^{(\Sigma |\log(pred/obs)| / n)}$$

Interpretation of B_f

$B_f > 1$, faster growth

$B_f < 1$, slower growth

Good: 0.95-1.11

Acceptable: 0.87-0.95 or 1.11-1.43

Unacceptable: <0.87 or >1.43

(Ross, 1999)

Applications

What is it used for?

For assessment of microbial food safety and stability:

- Predicting the effect of product characteristics and storage

- Supporting HACCP systems

- Easing education and training activities

- Qualifying QMRA models

Tools – curve fitting

- DMFit
  - UK: www.combase.cc
  - Estimation of growth kinetic parameters from growth curve data
  - Estimation of kinetic parameters from inactivation curves of various shapes

Relation between max. growth rate, μ_{max} & generation time:

Growth rate depends on the unit of your data

For ln(CFU/g):   max. specific growth rate = μ_{max}

For $\log_{10}$(CFU/g):   max. growth rate = μ_{max} / ln(10)

And   Generation time = ln(2) / μ_{max}

Tools – databases

- Pathogen Modeling Program (PMP)
  - USA
  - http://ars.usda.gov/services/docs.htm?docid=6786
  - http://pmp.errc.ars.usda.gov/PMPOnline.aspx
  - >40 models (growth, survival and inactivation)
  - Available as freeware
- ComBase
  - UK & USA: www.combase.cc
  - ComBase Predictor: online models for growth and inactivation for mainly pathogens
  - ComBase Perfringens Predictor: online model for evaluation of safe cooling of meat
  - ComBase Browser: data for growth or inactivation of food associated microorganisms

Tools – prediction

- Food Spoilage and Safety Predictor (FSSP)
  - DK: http://fssp.food.dtu.dk
  - Time-temperature integration
  - Shelf-life, specific spoilage organisms
  - *Listeria monocytogenes,* histamine formation
- DMRIpredict
  - DK: http://dmripredict.dk
  - safety models, *L. monocytogenes*, *Clostridium botulinum*, ConFerm, *Yersinia enterocolitica*, Staphtox predictor, F value calculator
  - shelf-life models, pork, beef and chicken cuts, minced pork, minced beef and bacon

Other available tools

Contents lists available at ScienceDirect

Food Microbiology

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Software for predictive microbiology and risk assessment: A description and comparison of tools presented at the ICPMF8 Software Fair

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ARTICLE INFO

ABSTRACT

The 8th International Conference on Predictive Modelling in Food was held in Paris, France in September 2013. One of the major topics of this conference was the transfer of knowledge and tools between academics and stakeholders of the food sector. During the conference, a "Software Fair" was held to provide information and demonstrations of predictive microbiology and risk assessment software. This article presents an overall description of the 16 software tools demonstrated at the session and provides a comparison based on several criteria such as the modeling approach, the different modules available (e.g. databases, predictors, fitting tools, risk assessment tools), the studied environmental factors (temperature, pH, a_w, etc.), the type of media (broth or food) and the number and type of the provided microorganisms (pathogens and spoilers). The present study is a guide to help users select the software tools which are most suitable to their specific needs, before they test and explore the tool(s) in more depth.

Homework

Listeria monocytogenes	Generation times in hours when pH=6, salt-in-water=2% and no additives			
	ComBase	**PMP**	**FSSP**	**DMRI**
At 5ºC				
At 10ºC				
At 20ºC				

Relation between maximum growth rate, μ_{max} & generation time:

Growth rate depends on the unit of your data

For ln(CFU/g):   maximum specific growth rate = μ_{max}

For $\log_{10}$(CFU/g):   maximum growth rate = μ_{max} / ln(10)

And   Generation time = ln(2) / μ_{max}

Questions:

Which model would be most suited for prediction in organic "rullepølse"?

For how long can organic "rullepølse" be served from a buffet?

Problem 1:
Model development and validation

Building and validating models for prediction of growth rate of *L. monocytogenes* in cooked chicken breast for sandwich production.

1. Find growth rates at 5, 10, 20 and 25 °C in ComBase Predictor, PMP and FSSP (pH 6.2, 0.5 % salt-in-water)

2. Collect the growth rates in an excel sheet and convert the growth rates to μ_{max} estimates

3. Plot the square root of μ_{max} as a function of temperature for each model in an X Y scatter plot

4. Fit a linear trendline to the points (now you have three predictive models!)

5. Fit growth curves from chicken stored at 7, 9, 11, 16, 19 and 23 °C using DMFit, collect the growth rates and convert to μ_{max} – these are your **observed** values

6. Predict μ_{max} at 7, 9, 11, 16, 19 and 23 °C with each of your models – these are your **predicted** values

7. Compare observed and predicted values by calculating the Bias factor for each of your three models

Problem 2:
Which one is the fastest?

- Use Combase Predictor
- Predict the time needed to obtain 1 log increase with and without lag at
  - 10 °C
  - Salt = 1 %
  - pH 6.0
  - No preservatives

Pathogen	Time for 1 log increase	
	lag	no lag
B. cereus		
Brochothrix		
C. botulinum (np)		
E. coli (VTEC)		
L. moncytogenes		
Pseudomonas		
Salmonella		
S. aureus		

Problem 3:
Microbiological spoilage and shelf-life

You have prepared pork roast, which you slice and pack in smaller portions in containers with lid to be kept in the fridge and served later.

From experience you know that meat products smell bad when the number of *Brochothrix thermosphacta* reaches 10^6 – 10^7 per gram.

If you have 100 *B. thermosphacta* per gram from the beginning, for how long can you keep the sliced meat at 4°C before it smells bad?

What is the generation time of *Listeria monocytogenes* under these conditions?

Does *L. monocytogenes* increase more than 1 log-unit if you have 10 from the beginning?

> **Hint:**
>
> Find the max. rate for *B. thermosphacta* in the ComBase browser.
>
> Be aware of the unit!!

Problem 4:
Has the mashed potatoes been cooled fast enough?

- Use Perfringens Predictor

- Predict the growth of *C. perfringens* at
  - pH = 5.9
  - Salt = 1.5 %
  - No nitrite

- Discuss whether the mashed potatoes should be discarded?

Time (hours)	Temperature (°C)
0	72.0
0.5	56.6
1	44.5
1.5	35.0
2	27.5
2.5	21.6
3	17.0
3.5	13.4
4	10.5
4.5	8.3
5	6.5