



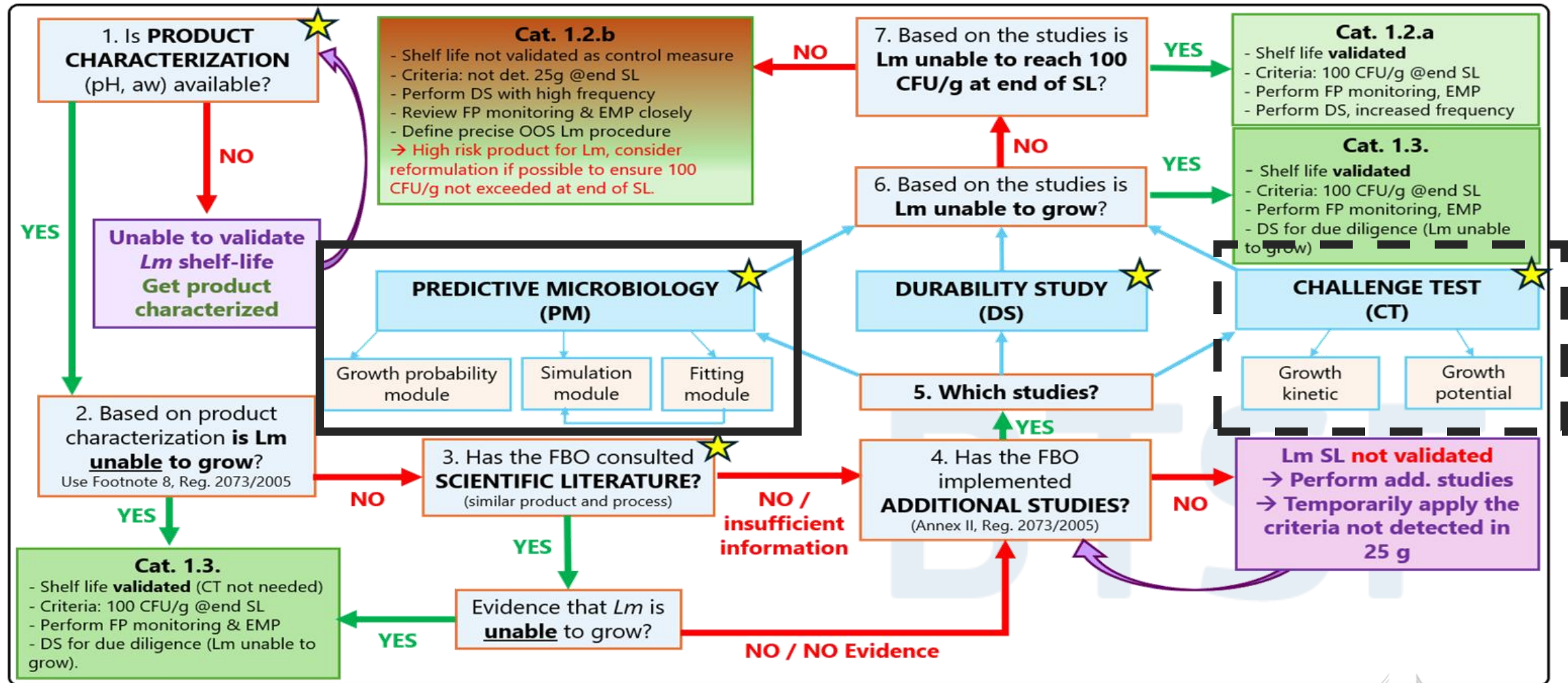
BTSF ACADEMY

Predictive Microbiology

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STM - Microbiological shelf-life studies of
ready-to-eat foods related
to *L.monocytogenes*

Mariem Ellouze
Lithuania, Session 1: 14- 17/04/2026;
Session 2: 05-08/05/26



- 1 What is Predictive Microbiology
- 2 Digital transformation applied to microbiology
- 3 Modeling approaches
- 4 Predictive Microbiology tools
- 5 Conclusions

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$$\begin{aligned}
 & p_k = \binom{n}{k} p^k (1-p)^{n-k} \\
 & \left| \int_{|x|>A} f(x) \log_2 \frac{1}{f(x)} dx \right| < \varepsilon \quad g^{-1} \cdot g = e \\
 & \sum_{k=0}^{\infty} e^{-\frac{k^2}{2}} = H(k) \quad \prod_{k \leq b} \bigcup_{i=1}^{n-1} H_i / \bigcap_{n=0} \\
 & G_k(x) \geq \frac{1}{2} \sum_{n=0}^{\infty} \frac{1}{n!} e^{-\frac{k^2}{2}} = H(k) \\
 & f_{n-1}(t) = \int_0^t f_n(u) f_1(t-u) du = \frac{2^{n+1} t^n e^{-2t}}{n!} \quad \lim_{t \rightarrow \infty} (G_k) = 0 \\
 & \log \psi(t) = i \gamma t - c |t|^k \left[1 + i \beta \frac{t}{|t|} \omega(t) \right] \quad \beta(u) = \sum_{k=1}^r \psi^*(k) u^k \\
 & \int_{-\infty}^{\infty} e^{-\frac{u^2}{2}} du = F(x) \left(\frac{1}{\sqrt{2\pi}} \right)^{-1} \quad |\psi_g(t)| = \left| \int_{-\infty}^{\infty} e^{itx} dF(x) \right| \leq \int_{-\infty}^{\infty} e^{-ux} dF(x) = \\
 & \prod_m = \prod_r / \prod_{m-r} \\
 & |X \cup \Psi| = |X| + |\Psi| - |X \cap \Psi| \quad \lim_{n \rightarrow \infty} \frac{1}{n} k_n \left(\frac{x}{\sqrt{n}} \right) = \frac{1}{\sqrt{2\pi}} e^{-\frac{x^2}{2}} \\
 & f: X \rightarrow X \cap W \\
 & \int_1^{\infty} \chi(\omega) dP \quad f'(x) = -\log_2 \left(\frac{\sum_{k=1}^r P_k^* \log_2 \frac{1}{P_k}}{\sum_{k=1}^r P_k^*} \right) - \left(\frac{\sum_{k=1}^r P_k^* \log_2 \frac{1}{P_k}}{\sum_{k=1}^r P_k^*} \right) \\
 & \frac{1-g}{q} - 1 = x \left[\frac{g(1-g)}{u} + q \left(\frac{1}{u} \right) \right] \quad \prod_{k=1}^r \left[g_k \left(\frac{t}{T_{k0}} \right) \right]^{1/g_k} \\
 & \int_1^{\infty} dx \geq \int_1^{\infty} f(x) dx
 \end{aligned}$$

Using mathematical models to describe the effect of **time** and **environmental factors** on microbial behavior (growth / inactivation / survival).

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BTSF Predictive Microbiology is one of the tools to validate shelf life studies (EC) 2073/2005

ANNEX II

The studies referred to in Article 3(2) shall include:

- specifications for physico-chemical characteristics of the product, such as pH, a_w , salt content, concentration of preservatives and the type of packaging system, taking into account the storage and processing conditions, the possibilities for contamination and the foreseen shelf-life, and
- consultation of available scientific literature and research data regarding the growth and survival characteristics of the micro-organisms of concern.

When necessary on the basis of the abovementioned studies, the food business operator shall conduct additional studies, which may include:

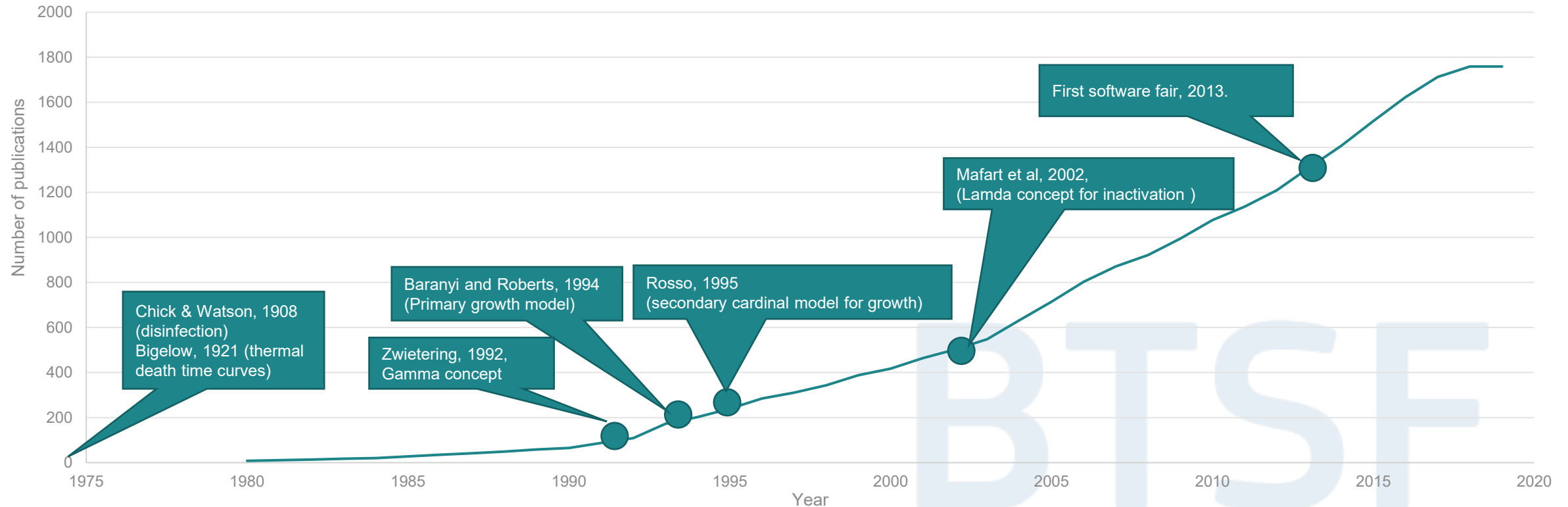
- predictive mathematical modelling established for the food in question, using critical growth or survival factors for the micro-organisms of concern in the product,
- tests to investigate the ability of the appropriately inoculated micro-organism of concern to grow or survive in the product under different reasonably foreseeable storage conditions,
- studies to evaluate the growth or survival of the micro-organisms of concern that may be present in the product during the shelf-life under reasonably foreseeable conditions of distribution, storage and use.

The above mentioned studies shall take into account the inherent variability linked to the product, the micro-organisms in question and the processing and storage conditions.

Well established science

BTSF History of PM : From research to Application

Number of publications with key word "predictive microbiology"



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How does it work?

- There is no magic !
- Predictability is the key
- The behaviour of micro-organisms is predictable: they behave the same under the same conditions.



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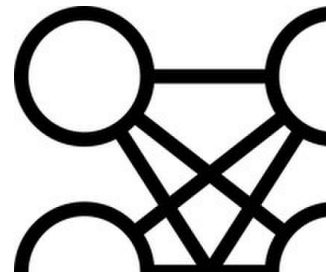
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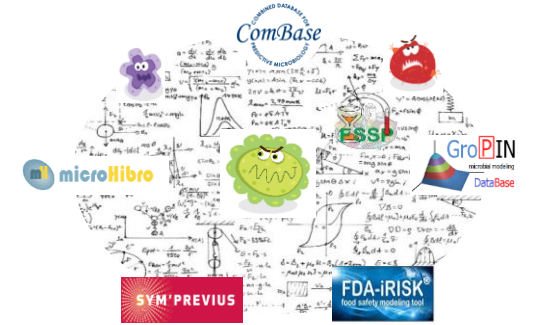
Data

→ Microbiological counts and meta data (pH, aw...) from
Challenge tests



Models

→ Calibrate the equations to the food and validate that it works well



Tools

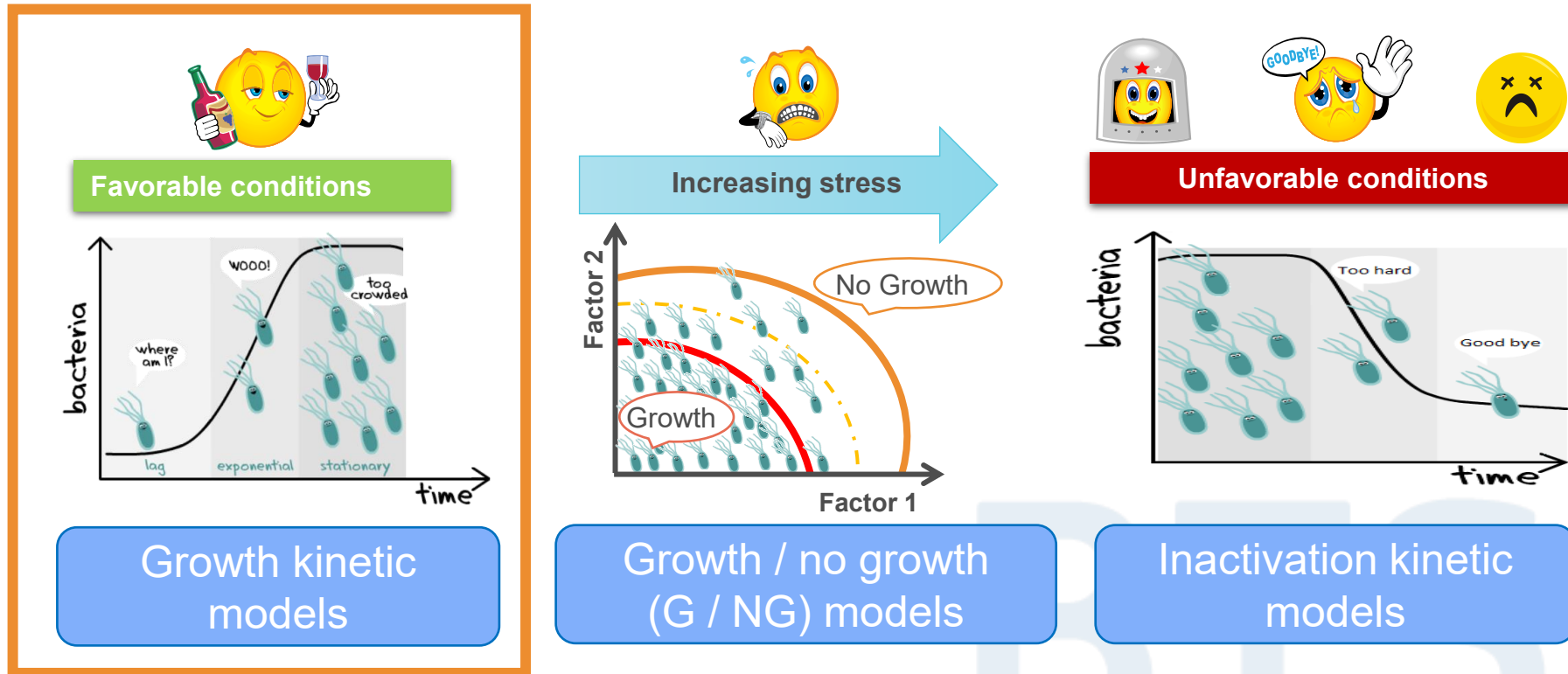
→ Use the validated models in a user friendly IT platform to predict Microbial behavior

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Modeling approaches



Kintaic models use mathematical models considering the *time* and the *environmental factors*

Growth / No Growth (G / NG) models use other mathematical models not considering the *time*

→ We will focus on kinetic models for growth

BTSF Predictive models for growth

Objective: Predict **microbial growth** using mathematical models taking into account the **time** and the **environmental factors**



Primary models

describe the changes in the number of organisms over **time** during growth using specific parameters (e.g. speed of growth called Maximum specific growth rate).



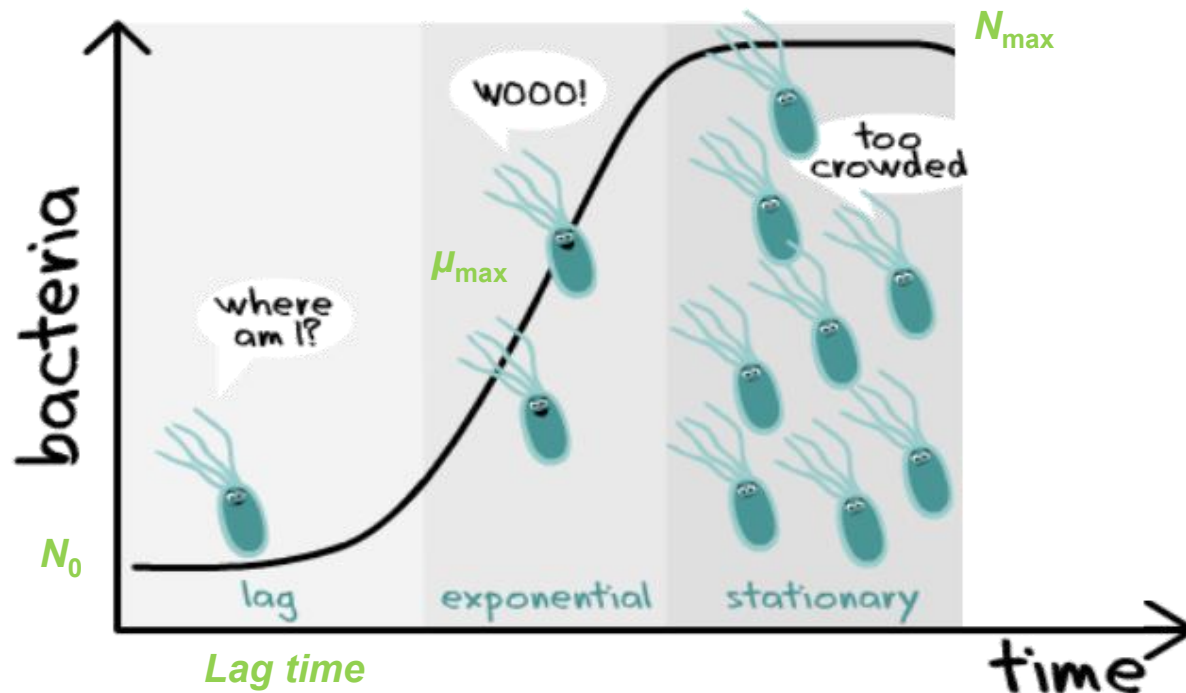
Secondary models

describe the effects of **environmental factors** on the parameters of the primary models like the speed of growth.

BTSF Primary models



→ Describe the changes in the number of organisms over **time** during growth using specific parameters.



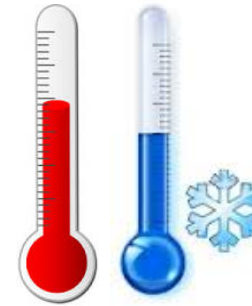
- *Lag* (*h*) time before growth starts
- μ_{max} (h^{-1}) maximum specific growth rate
- Rate / speed at which *L. monocytogenes* will multiply
- From challenge tests data, literature, specific databases using plate-counts or OD- binary dilution methods
- N_0 (CFU/g or /ml) initial population density
- N_{max} (CFU/g or /ml) maximum population density

BTSF Secondary models



→ Describe the effects of environmental factors

- Temperature, pH, aw
- Organic acids and preservatives
- Other environmental factors : CO₂



on the primary parameters (μ_{\max} , lag, N_{\max})

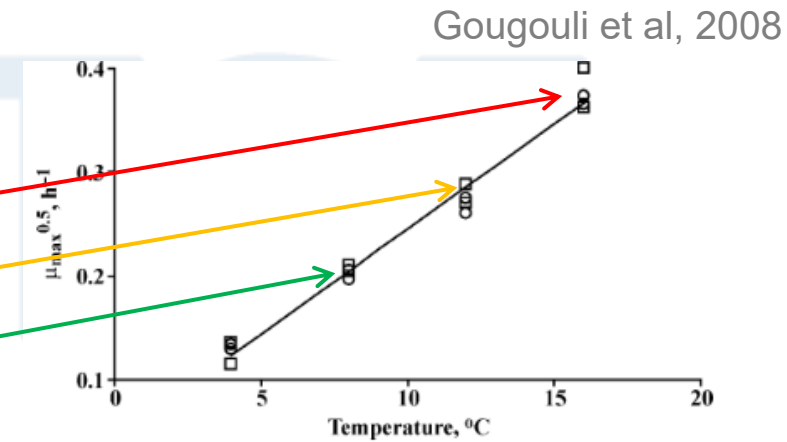
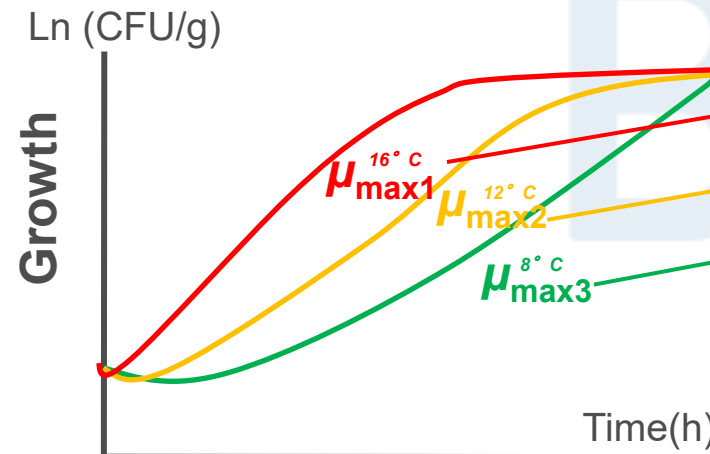
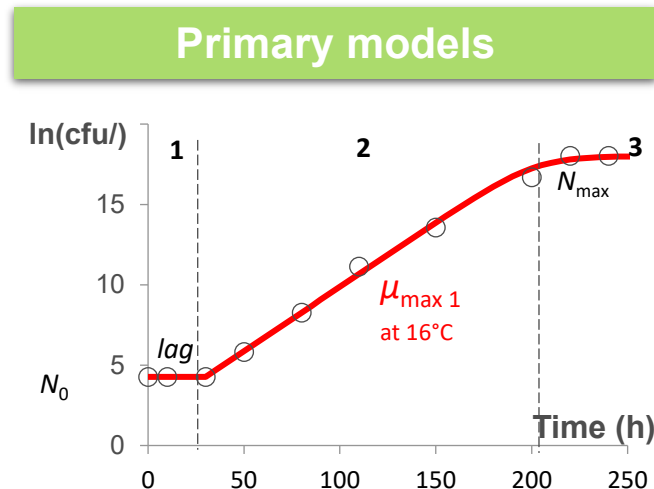


Figure 2. Square root model for the effect of chilling temperature on the maximum specific growth rate (μ_{\max}) of *Listeria monocytogenes* in 2 ice cream products (\square : product A; \circ : product B). Each point represents the mean of 2 replications from one experiment.

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BTSF Examples of Predictive Microbiology tools



- Based on broth models
- Polynomial modeling approach for the rate
- No variability: One input at a time, possible to run different simulations



- Based on models in pre-defined food
- Modeling approach is based on Square root and gamma for the rate
- No variability, one input at a time, possible to run 2 products in parallel for static scenarios



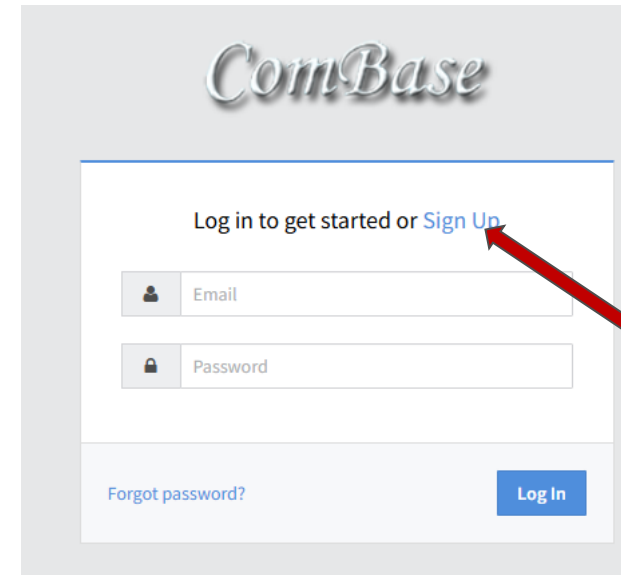
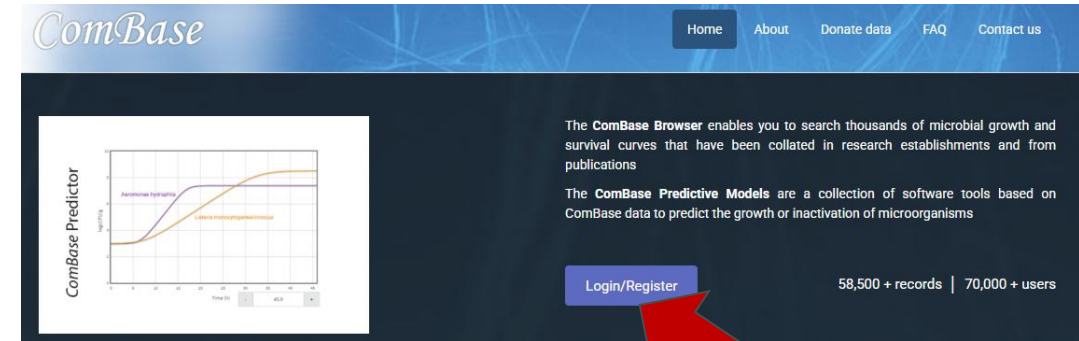
- Based on models in pre-defined foods and also specific CT
- Modeling approach is based on cardinal and gamma for the rate
- Possible to include variability

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<https://combase.errc.ars.usda.gov/>

Or just type Combase in your search engine



Only first time you
use the website

BTSF How to use ComBase www.combase.cc

The screenshot shows the ComBase web interface. On the left is a navigation menu with 'Broth Models' and 'Food Models'. Under 'Broth Models', 'Growth (revised)' is selected, showing sub-options: 'Thermal Inactivation', 'Non-thermal Survival', and 'Growth (legacy)'. The main content area shows a 'Static | Dynamic' toggle with 'Dynamic' selected. A dropdown menu shows 'List: Monocytogenes/innocua'. Below this, there are input fields for 'Init. level' (3), 'Phys. state' (1), 'pH' (7), and 'Aw | NaCl (%)' (0.99734). To the right of these fields are sliders for 'Max. rate (log CFU/g)' (8.52) and 'Lag time (Hours)' (0). A 'Chart' tab is active, showing a graph of log CFU/g vs Time (h). The graph shows a curve starting at 0 and rising to approximately 8.5 log CFU/g at 33 hours. A 'Data points' tab is also visible. Red arrows point from callout boxes to specific elements in the interface.

Chose the Broth models / Growth revised

Choose the target moo as *L. monocytogenes* / *innocua*

Indicate temperature only for static here

Indicate initial contamination in log: e.g. 10 CFU/g = 1 log CFU/g

Indicate physiological state if known, or keep worst case scenario to 1

Indicate product pH

Indicate product aw

Choose Dynamic for time temperature scenario or static for fixed temperature

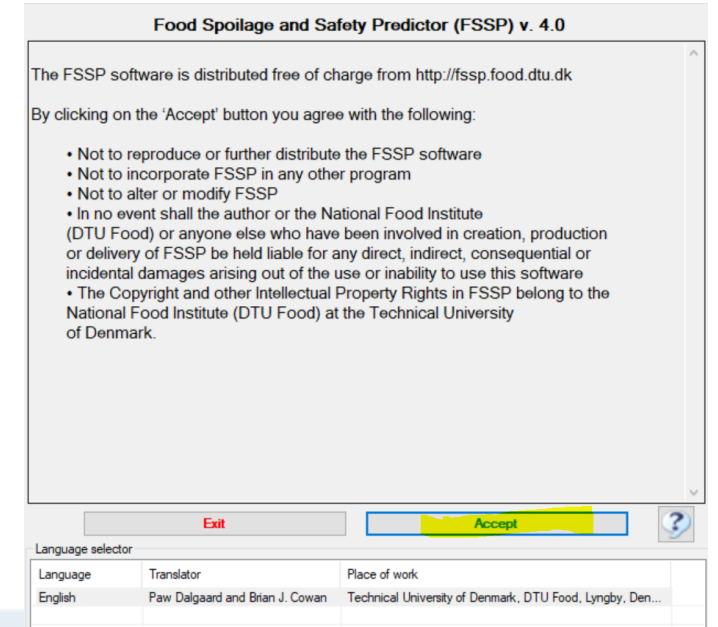
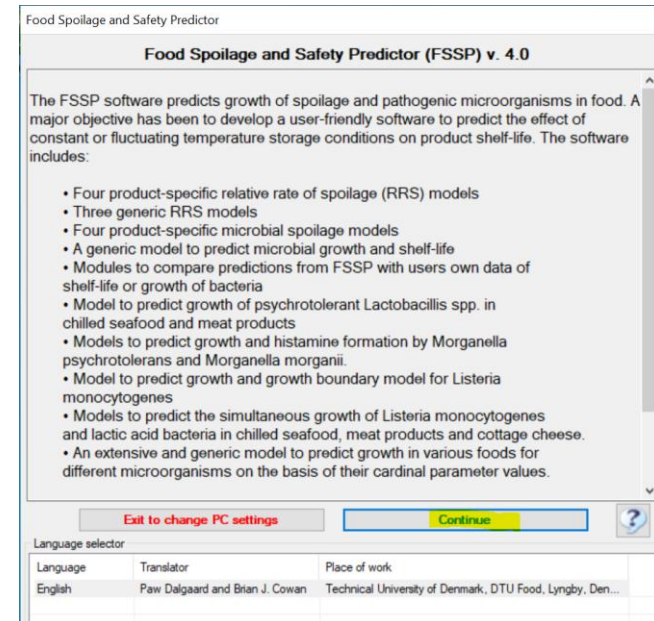
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FSSP



Installed on your computers



Food Spoilage and Safety Predictor

File Options Help

Time-Temperature Integration Software

- [-] Food Spoilage and Safety Predictor (FSSP)
 - [+] Relative rate of spoilage (RRS) models
 - [+] Microbial spoilage models (MSM)
 - [+] Psychrotolerant Lactobacillus spp. (LAB)
 - [+] Histamine formation models
 - [-] Listeria monocytogenes in chilled seafood and meat products
 - [+] Growth of L. monocytogenes
 - Effect of temp., atmosphere, salt, smoke, pH, nitrite and organic acids (acetic/diacetate, benzoic, citric, lactic and sorb...
 - [+] Growth boundary of L. monocytogenes
 - [+] Listeria monocytogenes and lactic acid bacteria (LAB)
 - [+] Listeria monocytogenes and lactic acid bacteria (LAB) in cottage cheese
 - [+] Generic growth models

BTSF FSSP Inputs and ranges

Product characteristics		Product 1	Product 2	Organic acids in water phase of product			
L. monocytogenes initial cell level (cfu/g)		<input type="text" value="1"/>	<input type="text" value="1"/>				
Temperature (°C)		<input type="text" value="5.0"/>	<input type="text" value="5.0"/>				
NaCl in water phase %		<input type="text" value="4.0"/>	<input type="text" value="4.0"/>	Acetic acid (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
pH		<input type="text" value="6.2"/>	<input type="text" value="6.2"/>	Benzoic acid (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
Smoke components - phenol(ppm)		<input type="text" value="10"/>	<input type="text" value="10"/>	Citric acid (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
% CO2 in headspace gas at equilibrium		<input type="text" value="0"/>	<input type="text" value="0"/>	Diacetate (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
Nitrite, mg/kg		<input type="text" value="0"/>	<input type="text" value="0"/>	Lactic acid (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
Storage period (d)		<input type="text" value="40"/>		Sorbic acid (ppm)	<input type="text" value="0"/>	<input type="text" value="0"/>	<input type="text" value="0"/>
Include lag time for L. monocytogenes		<input type="checkbox"/>	<input type="checkbox"/>	<input type="button" value="Apply"/> <input type="button" value="Clear"/>			

- Temperature (2-25°C),
- NaCl in water phase (0.7-9.0 %),
- pH (5.6-7.7),
- Smoke components/phenol (0-20 ppm),
- CO₂ (0-100 %),
- Nitrite (0-150 ppm in product),

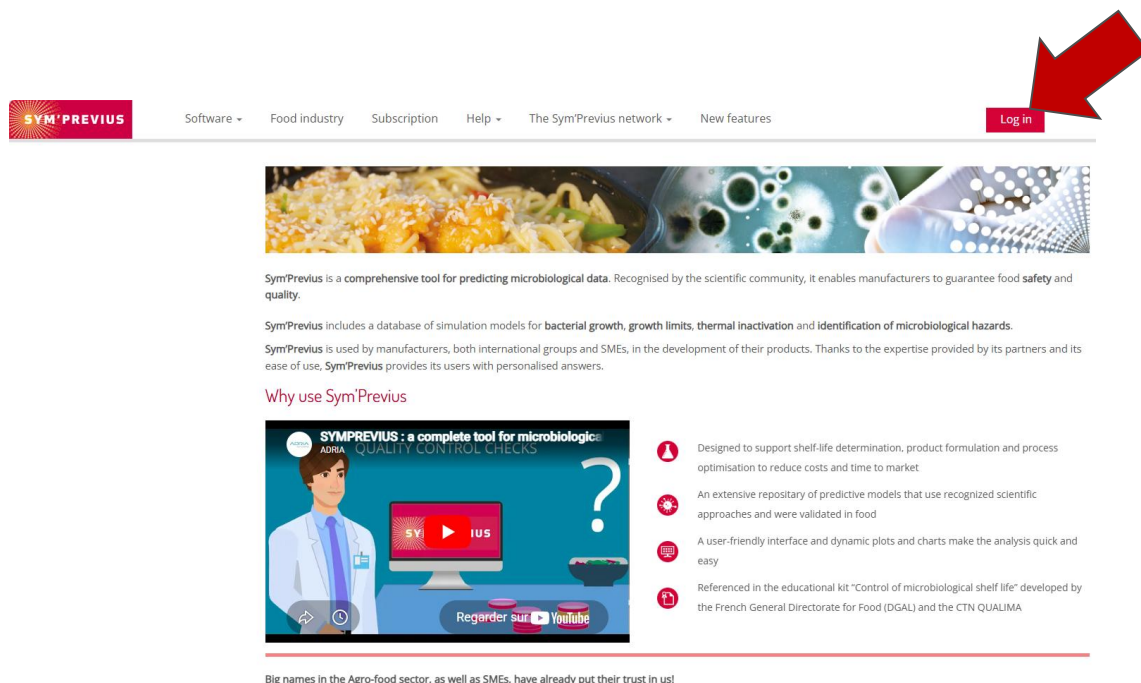
- Acetic acid (0-11000 ppm in water phase),
- Benzoic acid (0-1800 ppm in water phase),
- Citric acid (0-6500 ppm in water phase),
- Diacetate (0-3800 ppm in water phase),
- Lactic acid (0-60000 ppm in water phase)
- Sorbic acid (0-1300 in water phase).

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Sym'Previus



www.symprevius.eu



The screenshot shows the SYM'PREVIUS website interface. At the top, there is a navigation bar with the SYM'PREVIUS logo on the left and links for Software, Food industry, Subscription, Help, The Sym'Previus network, and New features on the right. A red arrow points to a "Log in" button in the top right corner. Below the navigation bar is a large banner image showing a bowl of food and a petri dish with bacterial colonies. The main content area contains the following text:

Sym'Previus is a comprehensive tool for predicting microbiological data. Recognised by the scientific community, it enables manufacturers to guarantee food safety and quality.

Sym'Previus includes a database of simulation models for bacterial growth, growth limits, thermal inactivation and identification of microbiological hazards.

Sym'Previus is used by manufacturers, both international groups and SMEs, in the development of their products. Thanks to the expertise provided by its partners and its ease of use, Sym'Previus provides its users with personalised answers.

Why use Sym'Previus

SYM'PREVIUS : a complete tool for microbiological QUALITY CONTROL CHECKS

The main content area features an illustration of a scientist in a lab coat standing next to a computer monitor displaying the SYM'PREVIUS logo. Below the illustration is a "Regarder sur YouTube" button. To the right of the illustration is a list of four bullet points:

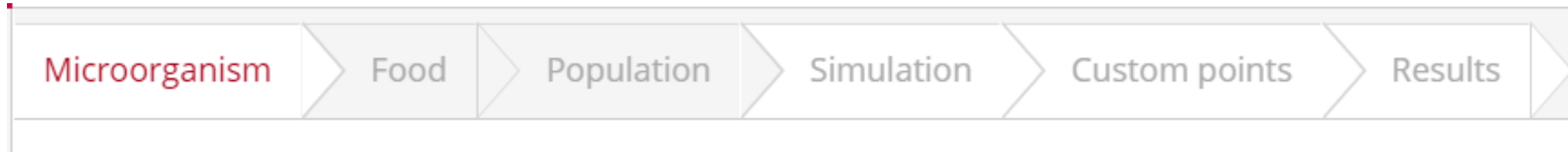
- Designed to support shelf-life determination, product formulation and process optimisation to reduce costs and time to market
- An extensive repository of predictive models that use recognized scientific approaches and were validated in food
- A user-friendly interface and dynamic plots and charts make the analysis quick and easy
- Referenced in the educational kit "Control of microbiological shelf life" developed by the French General Directorate for Food (DGAL) and the CTN QUALIMA

At the bottom of the page, there is a line of text: "Big names in the Agro-food sector, as well as SMEs, have already put their trust in us!"

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Steps for using Sym'Previous



1. Micro-organism

<input type="checkbox"/>	Sym'Previous	<i>Lactobacillus casei</i>	Spoilage	1	
<input type="checkbox"/>	Sym'Previous	<i>Leuconostoc mesenteroides</i>	Spoilage	2	
<input checked="" type="checkbox"/>	Sym'Previous	<i>Listeria monocytogenes</i>	Pathogen	14	
<input type="checkbox"/>	Sym'Previous	<i>Listeria monocytogenes</i> MEAN (no variability)	Pathogen	0	
<input type="checkbox"/>	Sym'Previous	<i>Pseudomonas fluorescens</i>	Spoilage	5	
<input type="checkbox"/>	Sym'Previous	<i>Pseudomonas putida</i>	Spoilage	1	

3. Population

☒ Initial contamination level (log CFU/g)

Mean Standard deviation

☐ Monitoring data (positive/negative)

☐ Monitoring data (bacterial counts)

2. Food

Laboratory medium or specific information in food product

☐ Culture medium

☒ Growth curve (challenge-test)

4. Simulation

Critical threshold level (log CFU/g)

Value

Storage time (d)

Value

☒ Switch on this widget to perform the simulation without lag (worst-case scenario)

Temperature (°C)

☐ Static

☒ Dynamic

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Steps for using Sym'Previous

Microorganism

Food

Population

Simulation

Custom points

Results

4. Simulation

Temperature (°C)

Static

Dynamic

	Time (h)	Temperature (°C)	
t0	0	4	
t1	25	4	🗑️
t2	128	4	🗑️
t3	128.1	8	🗑️
t4	256	8	🗑️
t5	384	8	🗑️

Number of lines : 6

Add a line

Reset

pH

Static

Mean

5.66

Standard deviation

0.07

Monitoring data

Dynamic

Optimum level for growth

Aw

Static

Mean

0.954

Standard deviation

0.01

Monitoring data

Dynamic

Optimum level for growth

Lactic acid

Static

Mean

0

Standard deviation

0

Unit

mMolaire



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Conclusions

- Predictive Microbiology is a mature science and can be reliably used
- Good quality data is required to build the models
- Available Predictive Microbiology software can be used to evaluate if Lm will grow to levels >100 CFU/g at end of shelf life



No need to be a modeler to use these software but it is important to get familiar with their specificities.

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Thank you!

European Commission
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Established by the European Commission

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