

Contribution of Predictive Microbiology to Control Dry-Fermented Sausage Safety

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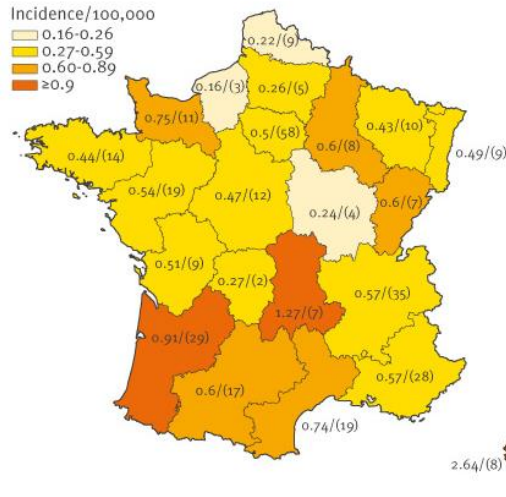
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Contribution of Predictive Microbiology to Control Dry-Fermented Sausage Safety

Incidence rate, per 100,000 inhabitants and per region, of *Salmonella enterica* 4,[5],12:i:- cases isolated by the National Reference Centre, 31 October to 18 December 2011, France (n=337)



EFSA Journal 2013;11(4):3129. [250 pp.].

Gossner, C.M., et al.. (2012). Eurosurveillance, 17 (5), 19-22

- **Fermented sausages are reported contaminated with *Salmonella***
 - Two main outbreaks in France associated with consumption of dried pork sausages
 - 2010: 132 cases
 - 2011: 337 cases
- **Technological strategies ensure the food safety of fermented sausages**
 - Formulation, bioprotective cultures, processing conditions, ...
- ➔ **Quantitative aspects of the microbial behaviour in dry-fermented sausages**
 - Association with challenge testing
 - Relevance of the factors and design control measures.



Process of Dry-Fermented Sausages



- 1) **80% pork lean meat**
20% derinded backfat
Meat ingredients **minced**
@ low temperature (-2°C to 0°C)



- 2) **Blending minced meats**
with other ingredients
 - ± Salt
 - ± Lactose
 - ± Dextrose
 - ± Starter

Process of Dry-Fermented Sausages



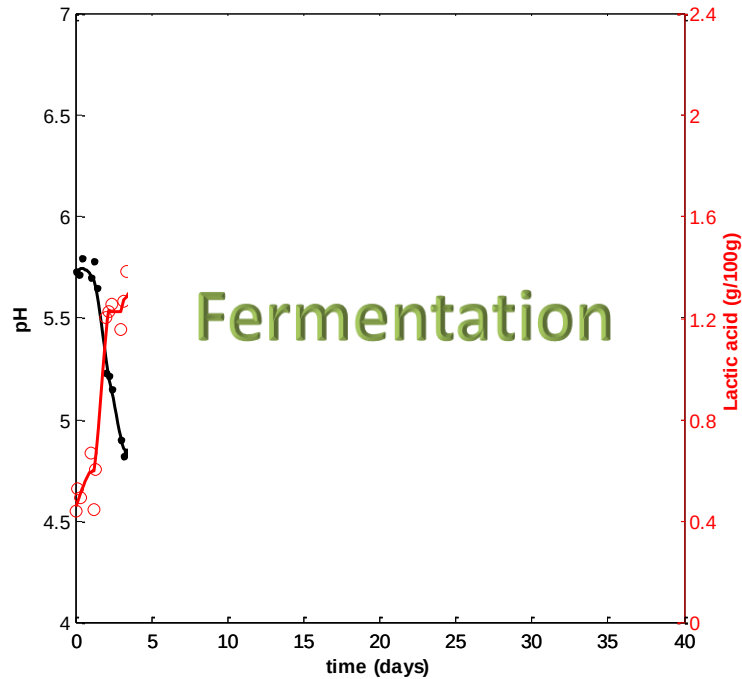
**3) Stuffing batter in casing
with a vacuum hopper**



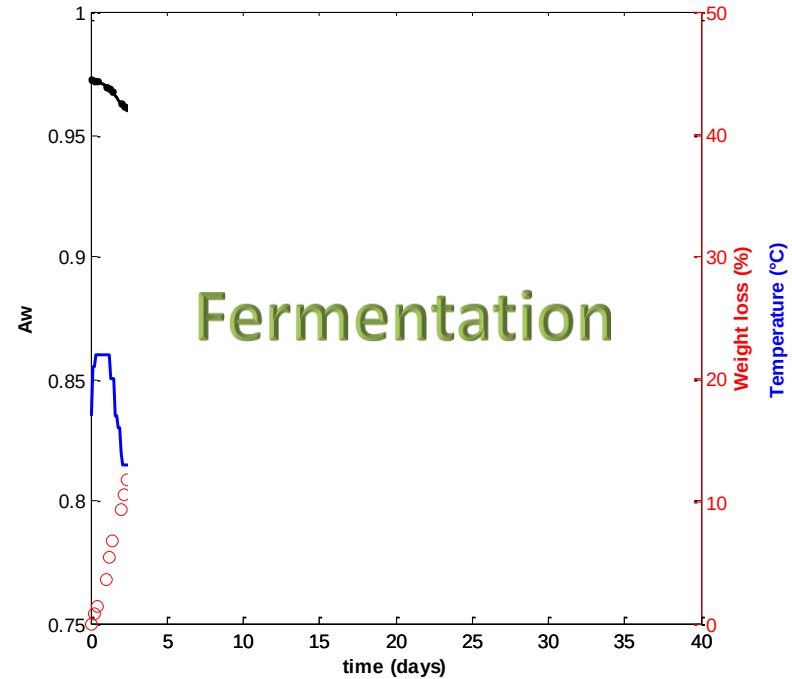
4) Ripening

- **Climatic chamber**
 - @ controlled Temperature
 - @ controlled Relative humidity
- **Fermentation (48h)**
- **Drying (4 weeks)**

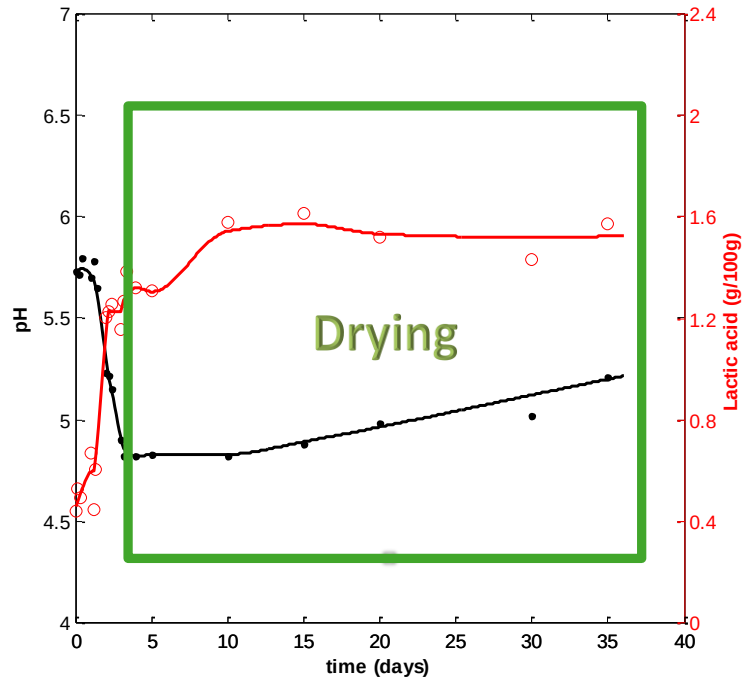
pH
lactic acid



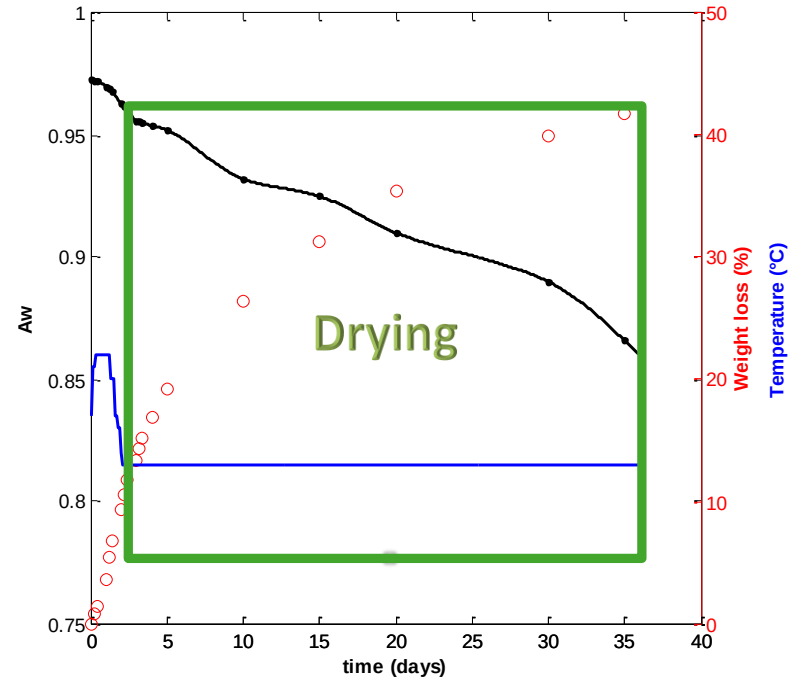
a_w
weight loss
temperature



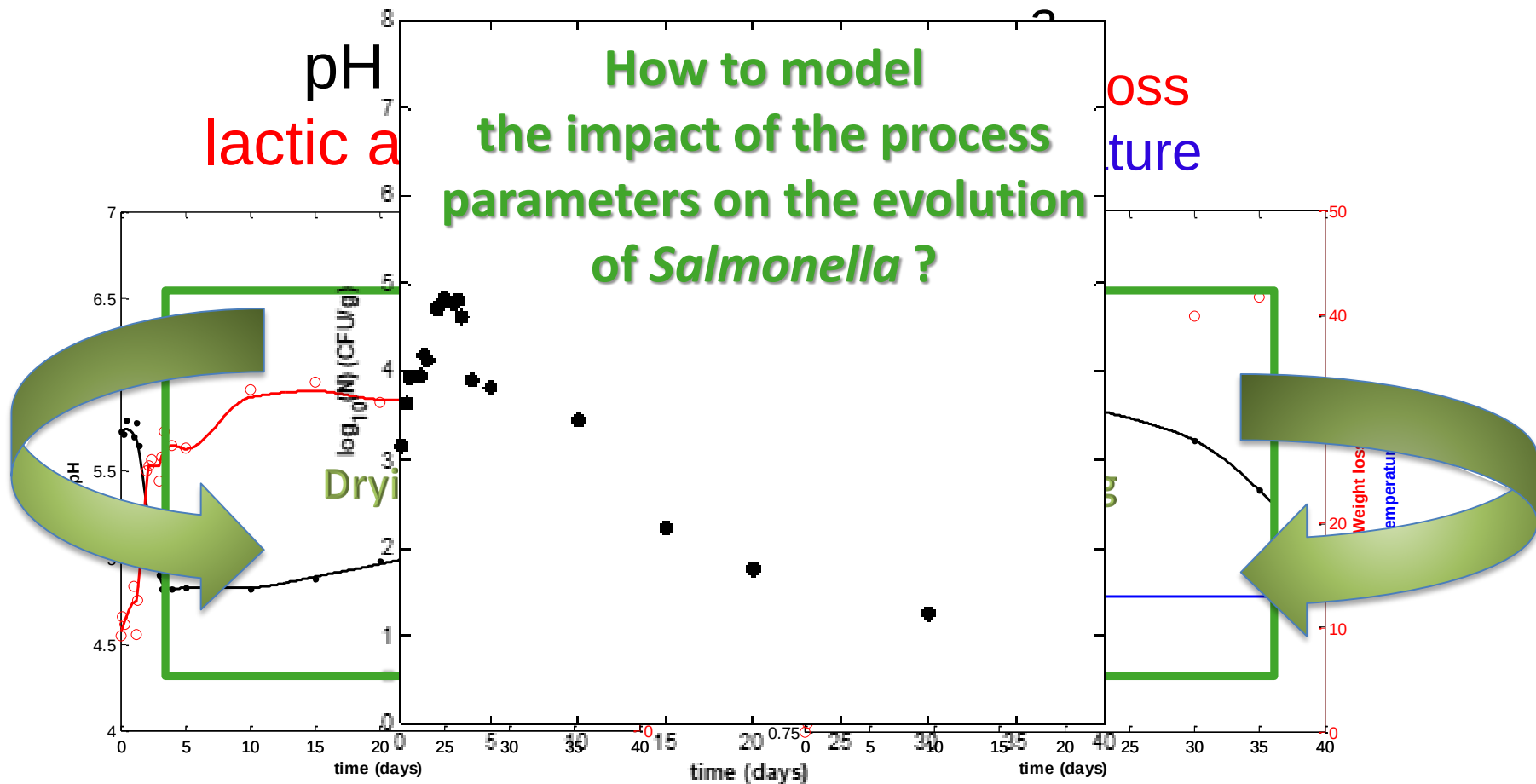
pH
lactic acid



a_w
weight loss
temperature



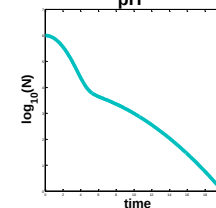
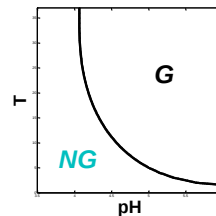
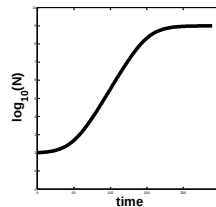
How to model the impact of the process parameters on the evolution of *Salmonella* ?



Background - Evolution of pathogen or spoilage microorganisms

Many models exist for:

- ✓ **Bacterial growth**
(meat, milk, vegetable,...)
growth rate (μ), population size (N)
- ✓ **Growth/no-growth interface**
Growth Probability (p)
- ✓ **Bacterial inactivation**
(salami, salad dressing, ...)
Inactivation rate (μ, k),
Resistance (D, δ, t_{4D}), population size (N)



One combined model for growth and inactivation

- ✓ **Modelling the evolution of *Salmonella* during the process of dried sausages:**

One parameter

$$\Delta \log N / \Delta t$$

- *Variation of bacterial population size for given time of storage*
increase, decrease, stability

- ✓ **Benefits**

- *Quantification of microorganism behaviour as a function of process factors*
- *Mapping of product formulation options*

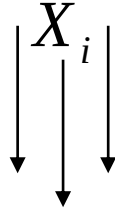
- ✓ **“Easy to use”**

- *Biological parameters found in literature, expert opinion, ...*
- *Bacterial behaviour predicted from food properties & vice-versa*
- *Easy to extend to new environmental factors*

One combined model for growth and inactivation

Environment

pH, lactic acid, aw , T



$$\prod \gamma_i(X_i) \cdot \xi$$

$\gamma(X_i)$

Effect of each factor (X_i)

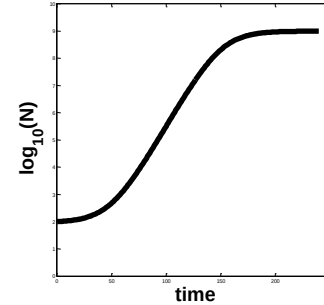
ξ

Effect of the interactions

These values are ranged from 0 (no-growth) to 1 (optimal growth)

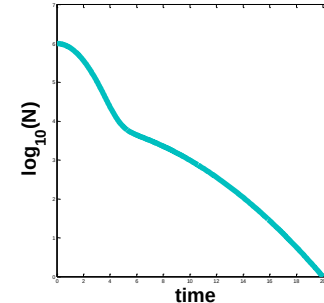
> 0

Growth



$= 0$

Inactivation



One combined model for growth and inactivation

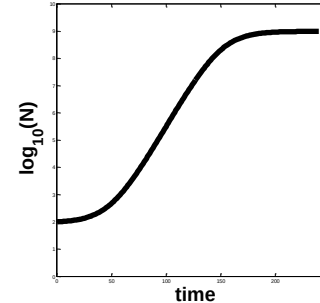
Growth

Primary modelling

- Rosso model (1995)

Secondary modelling

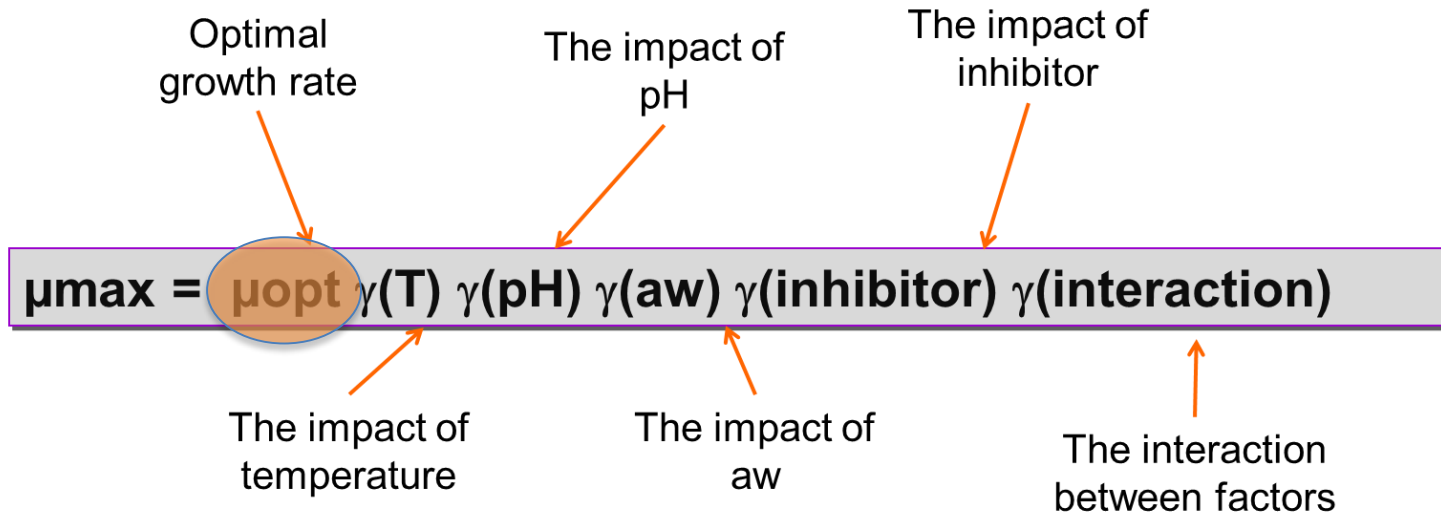
- With interactions



$$\mu = \mu_{opt} \cdot \prod \gamma_i(X_i) \cdot \xi$$

Zuliani et al. (2007). Journal of Applied Microbiology

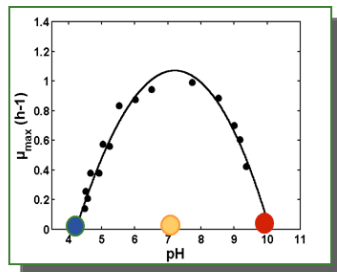
The modular (gamma) concept



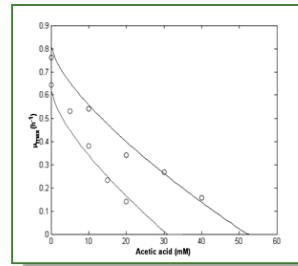
Factors which are not modeled by a gamma are summarized in the μ_{opt} in this specific product.

The Modular concept

Optimal growth rate

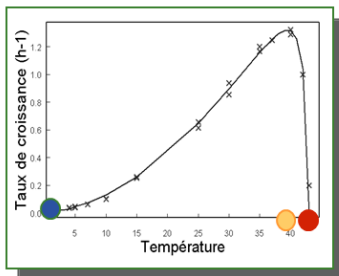


pH_{min} pH_{opt} pH_{max}

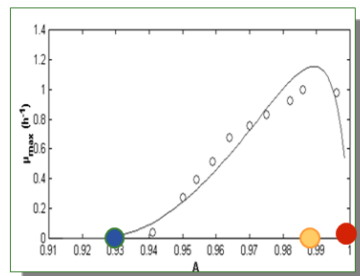


alpha MIC

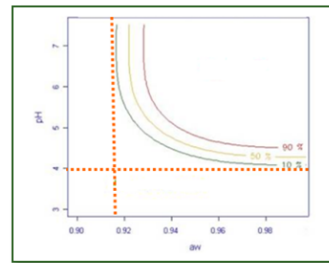
$$\mu_{\max} = \mu_{\text{opt}} \gamma(T) \gamma(\text{pH}) \gamma(\text{aw}) \gamma(\text{inhibitor}) \gamma(\text{interaction})$$



T_{min} T_{opt} T_{max}



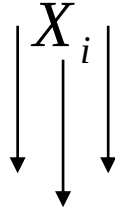
aw_{min} aw_{opt} aw_{max}



One combined model for growth and inactivation

Environment

pH, lactic acid, aw , T



$$\prod \gamma_i(X_i) \cdot \xi$$

$\gamma(X_i)$

Effect of each factor (X_i)

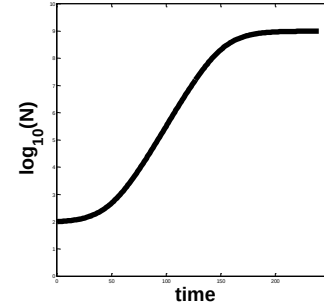
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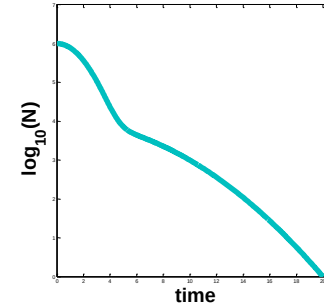
> 0

Growth



$= 0$

Inactivation



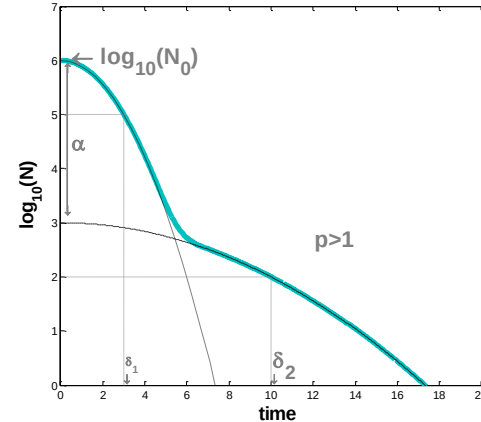
One combined model for growth and inactivation

Inactivation

Primary modelling

Double Weibull model

- 2 subpopulations
- δ_1 and δ_2 : first decimal reduction time
- Compatible with D value



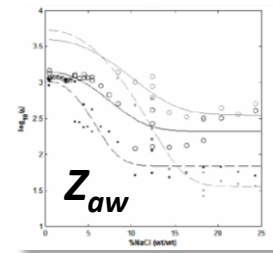
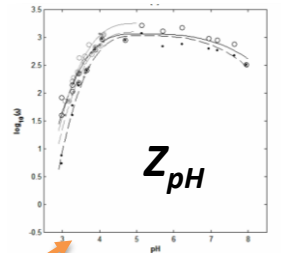
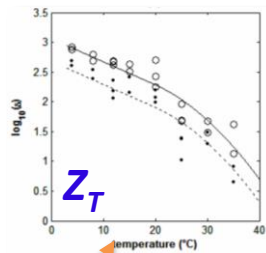
Coroller et al.(2006) AEM
Coroller et al. (2012) IJFM

Secondary modelling

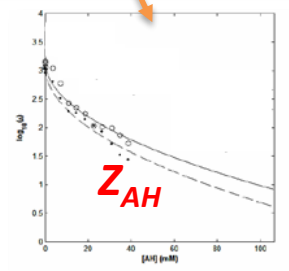
Gamma concept like model

$$\frac{1}{\delta_{T_{HT}, pH, aw}} = \frac{1}{\delta_{max}} \lambda_T (T_i) \cdot \lambda_{pH} (pH_i) \cdot \lambda_{AH} (AH_i) \cdot \lambda_{aw} (aw_i)$$

Modular inactivation model



$$\frac{1}{\delta_{T_{HT}, pH, aw}} = \frac{1}{\delta_{max}} \cdot \lambda_T(T_i) \cdot \lambda_{pH}(pH_i) \cdot \lambda_{AH}(AH_i) \cdot \lambda_{aw}(aw_i)$$

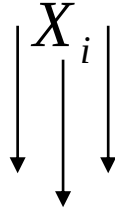


δ_{max} : first decimal reduction
@reference conditions (T, pH, aw...)
Media/strain dependant

One combined model for growth and inactivation

Environment

pH, lactic acid, aw , T



$$\prod \gamma_i(X_i) \cdot \xi$$

$\gamma(X_i)$

Effect of each factor (X_i)

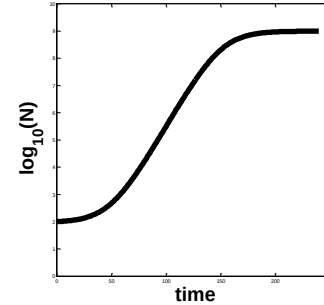
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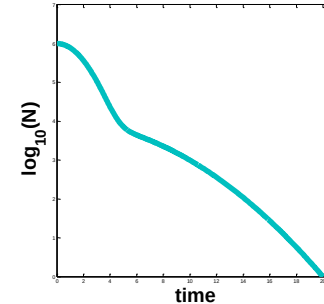
> 0

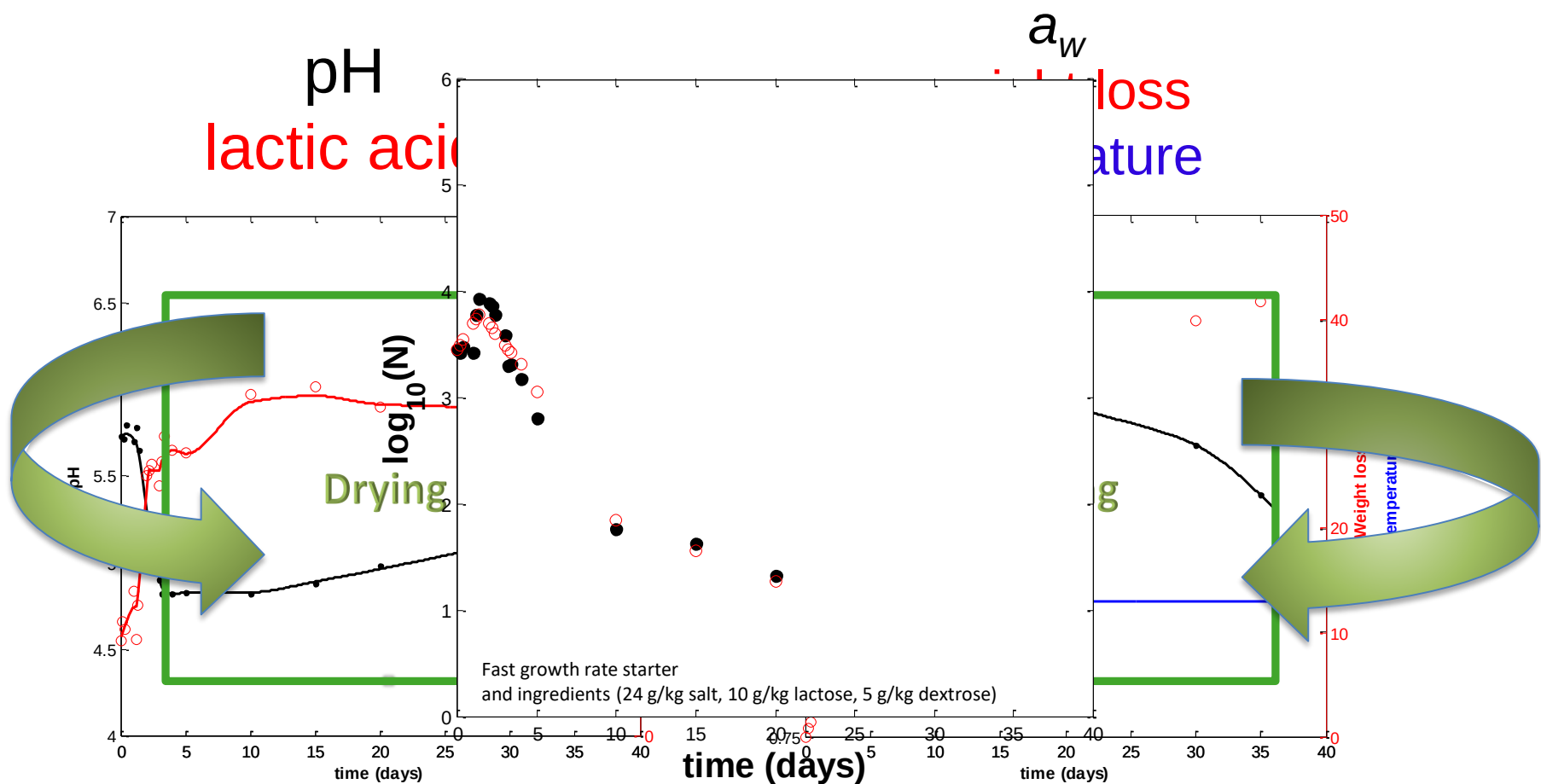
Growth



$= 0$

Inactivation





Growth/**Inactivation** transition

~ second day of ripening

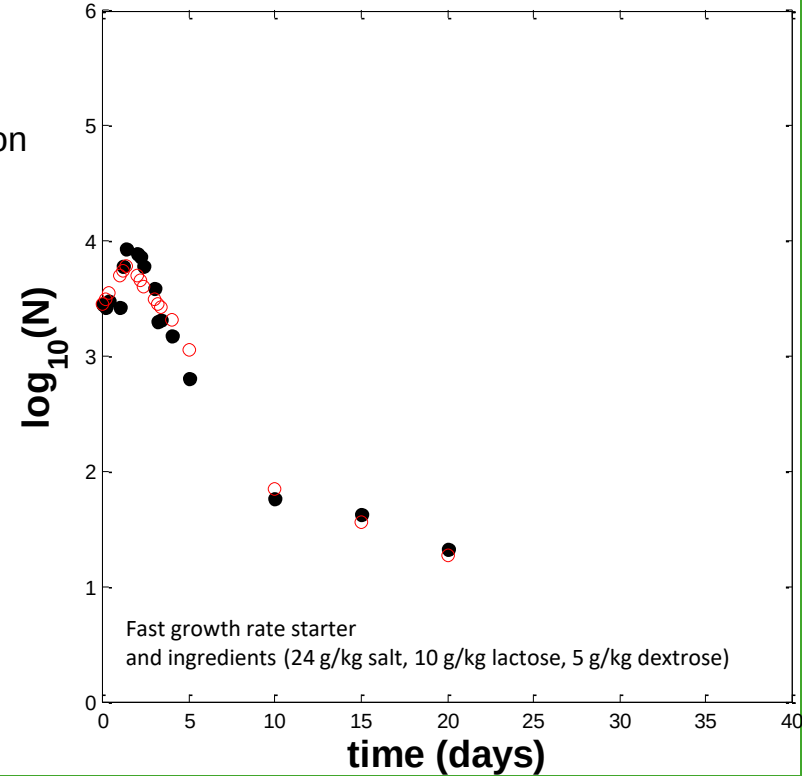
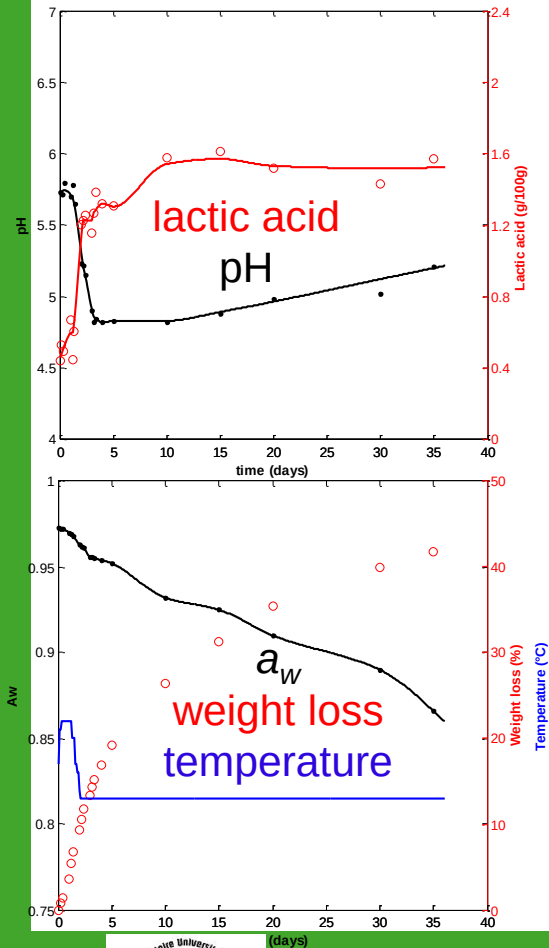
Temperature shift 20°C / 13°C

Decrease of water activity

Decrease of pH

Increase lactic acid concentration

➔ Synergetical effect



Growth/**Inactivation** transition

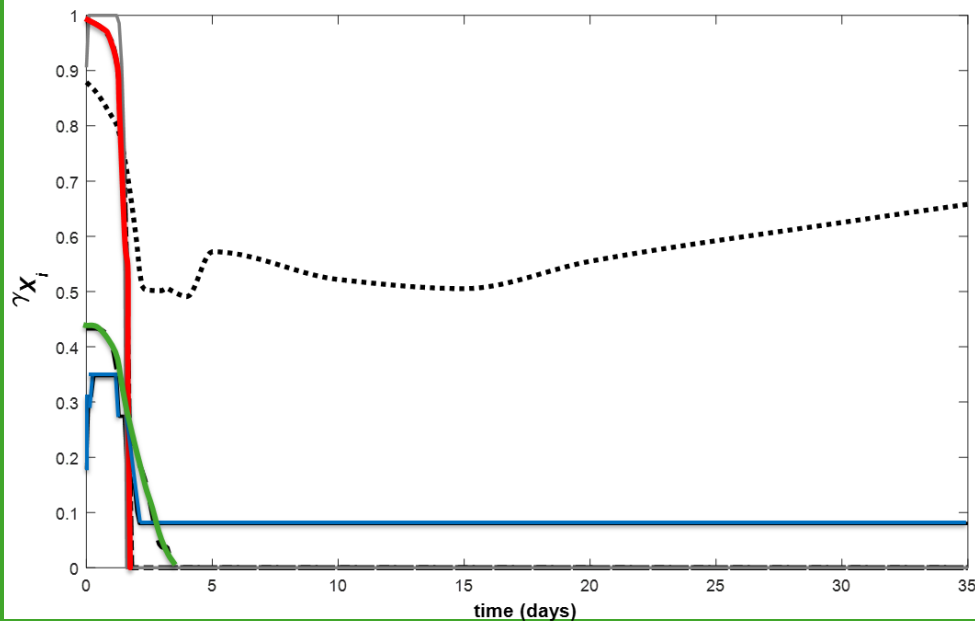
~ second day of ripening

Temperature shift from 20°C to 13°C

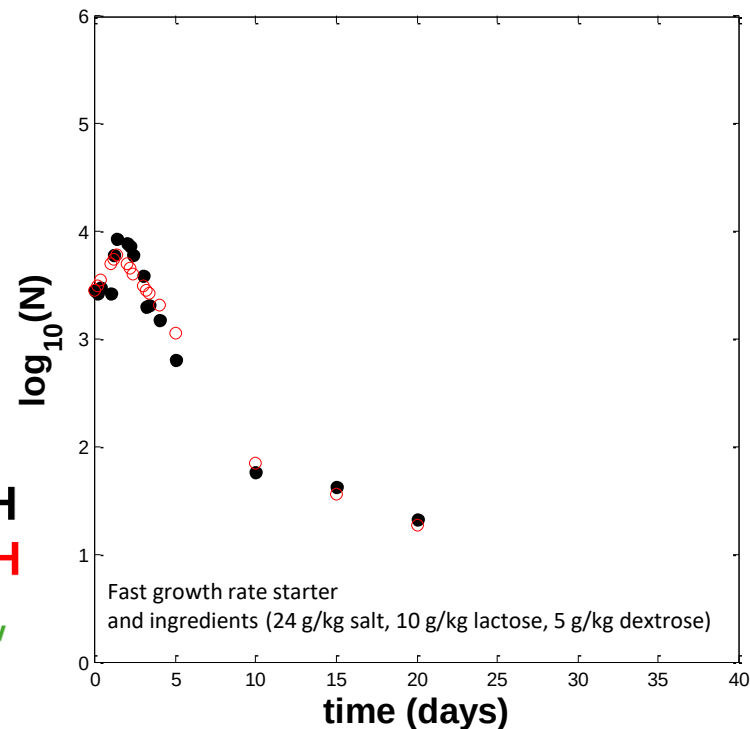
Decrease of **water activity**

Decrease of pH

Increase **lactic acid** concentration

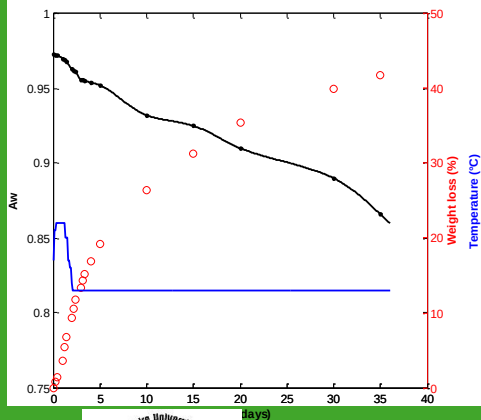


T
pH
AH
a_w
 ξ



What are the relevant process parameters to control the *Salmonella* population ?

- **pH** associated with increasing **lactic acid** concentration
- **Low temperature**
- Decrease of **water activity** $\Leftrightarrow$ **Weight loss**



$$W = C1 + C2 \ln(1 - aw)$$

Moisture sorption isotherm (Smith 1947, Andrade et al. 2011)

W the water content (g of water/g of dried weight),
C1 the quantity of water of the first sorbed fraction
C2 the quantity of water in the multilayer humidity fraction.

What are the relevant process parameters to control the *Salmonella* population ?

« Fast acidifying » starter

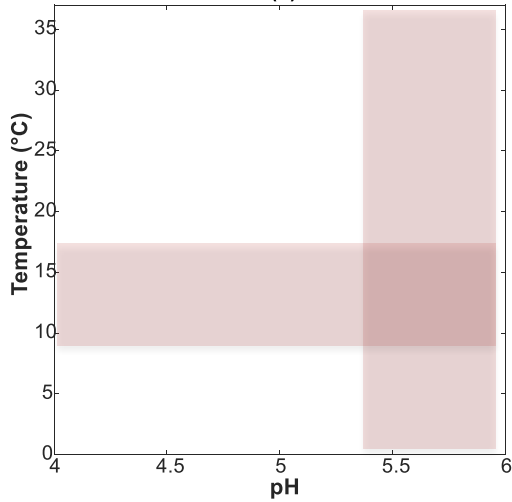
10g/kg lactose & 8g/kg dextrose



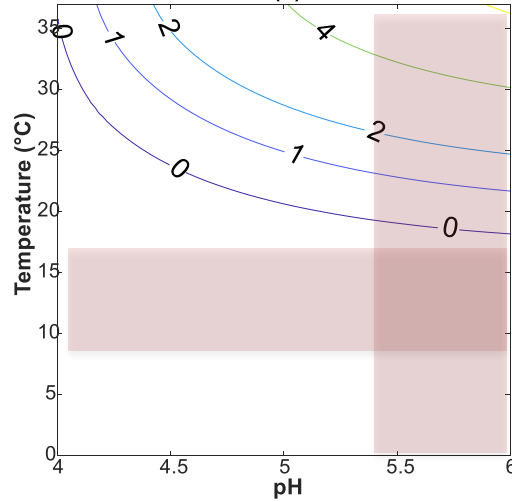
⇒ Slower growth

⇒ Lower resistance

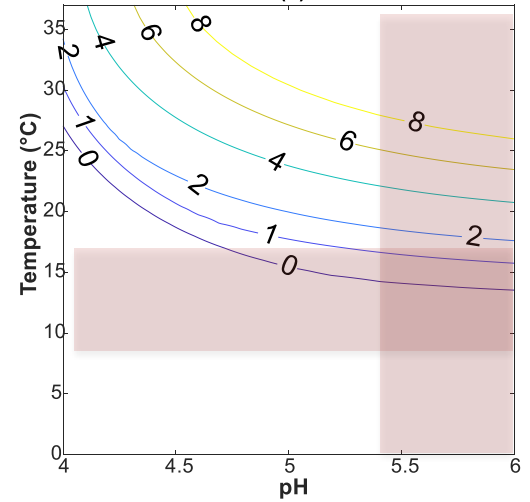
2 days



15 days

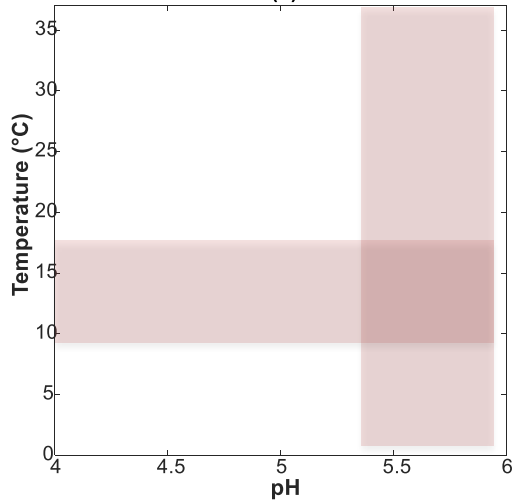


35 days

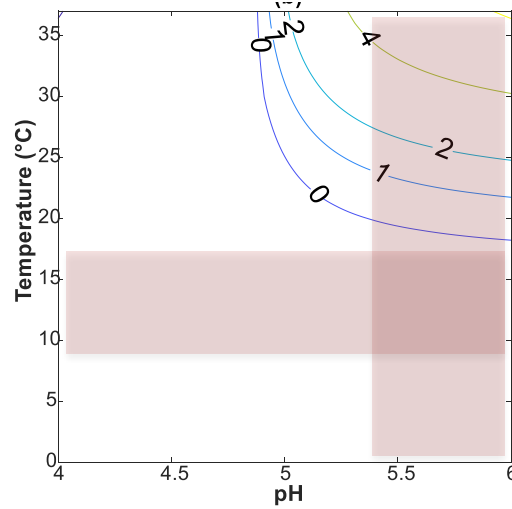


AH=0.0 % lactic acid
aw=0.97 $\Leftrightarrow$ 0 % Weight loss

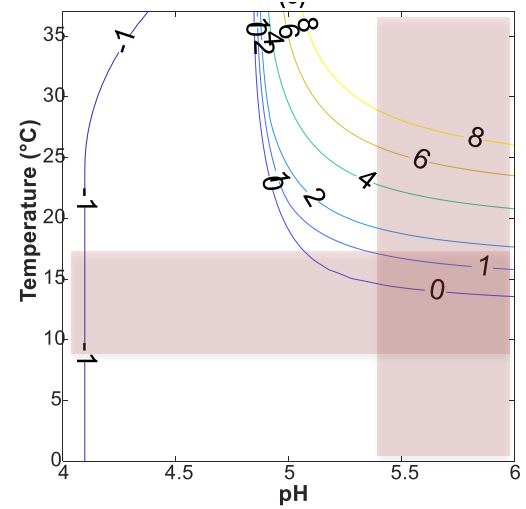
2 days



15 days

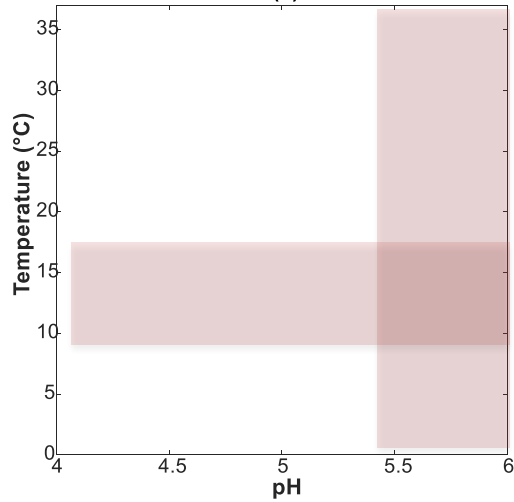


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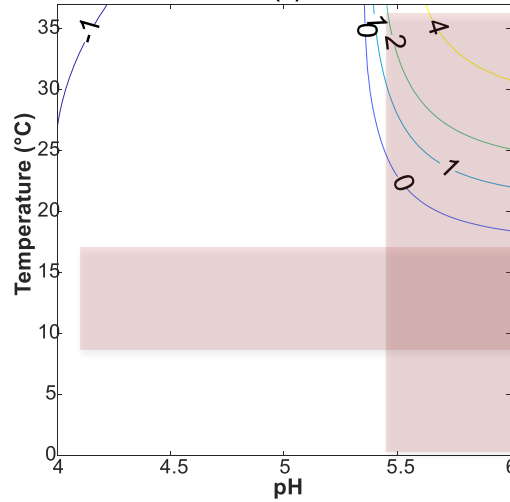


AH=0.5 % lactic acid
aw=0.97 $\Leftrightarrow$ 0 % Weight loss

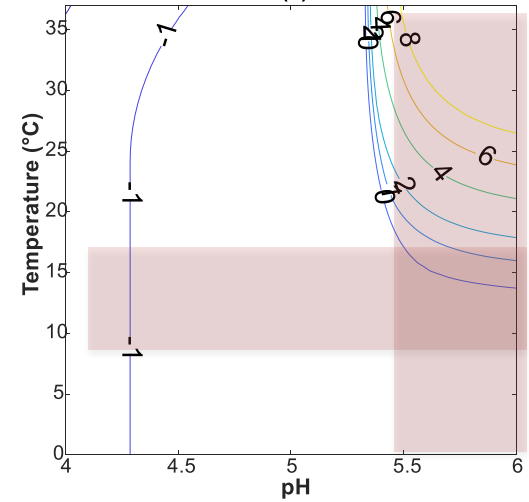
2 days



15 days

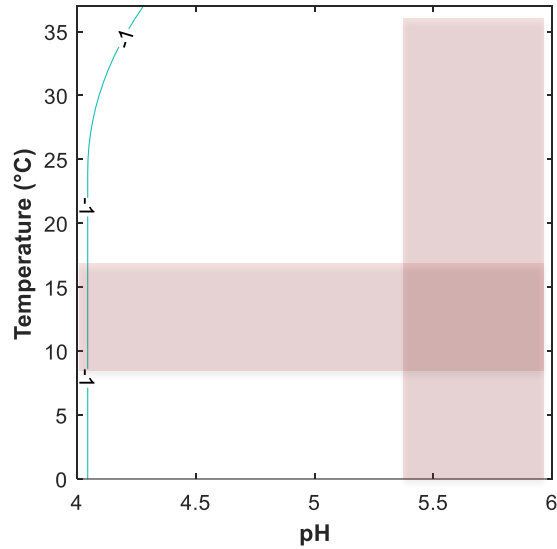


35 days

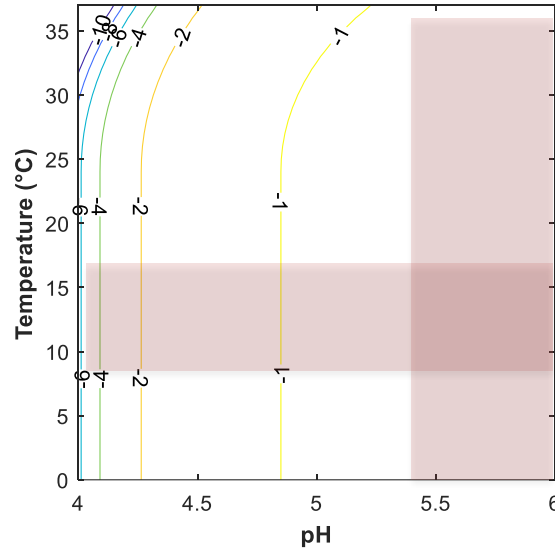


AH=1.5 % lactic acid
aw=0.97 $\Leftrightarrow$ 0 % Weight loss

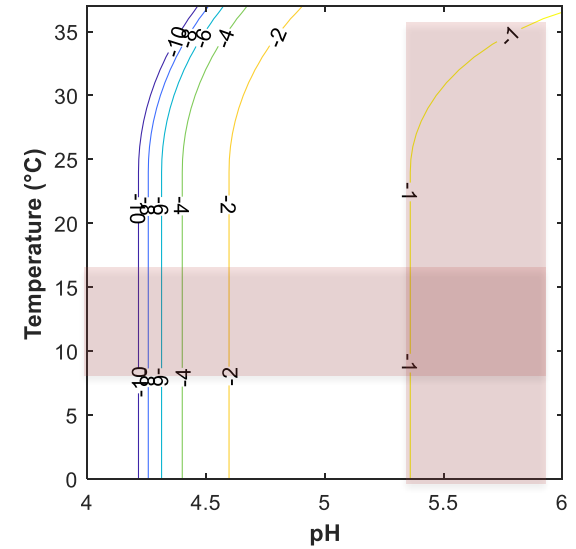
2 days



15 days



35 days



AH=1.5 % lactic acid
 $aw=0.95 \Leftrightarrow 10\%$ Weight loss

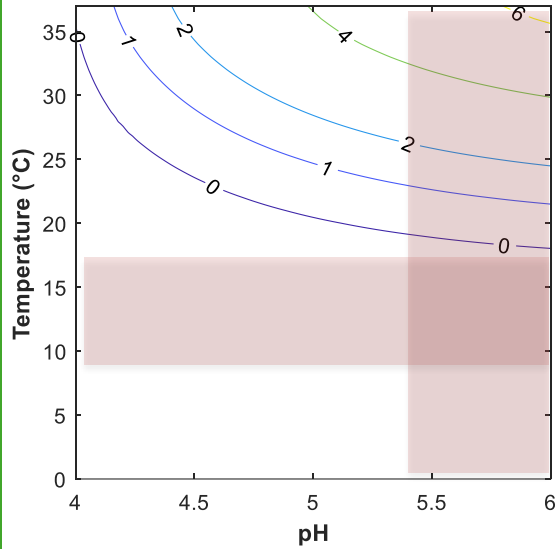
What are the relevant process parameters to control the *Salmonella* population ?

« moderate acidifying » starter
10g/kg lactose & 8g/kg dextrose

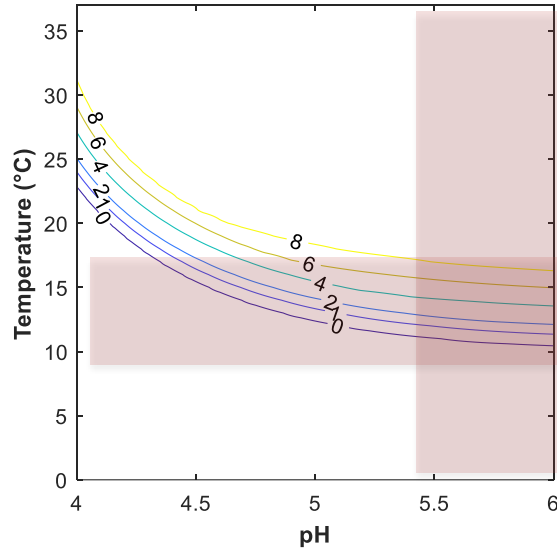


- ⇒ higher growth potential
- ⇒ higher resistance

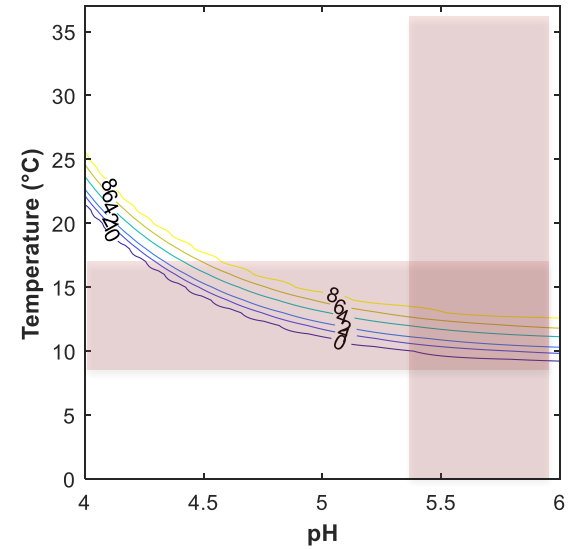
2 days



15 days

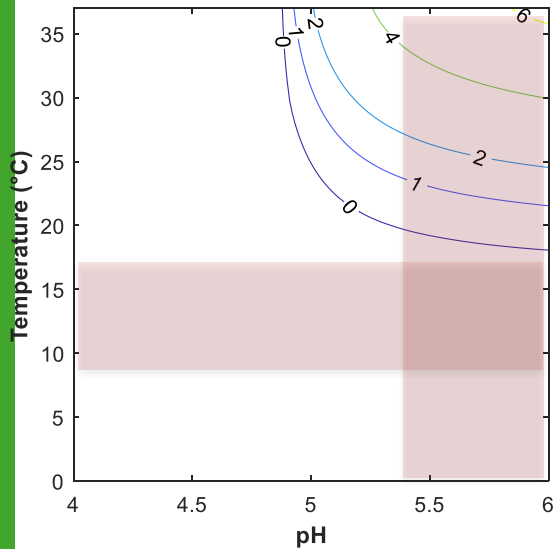


35 days

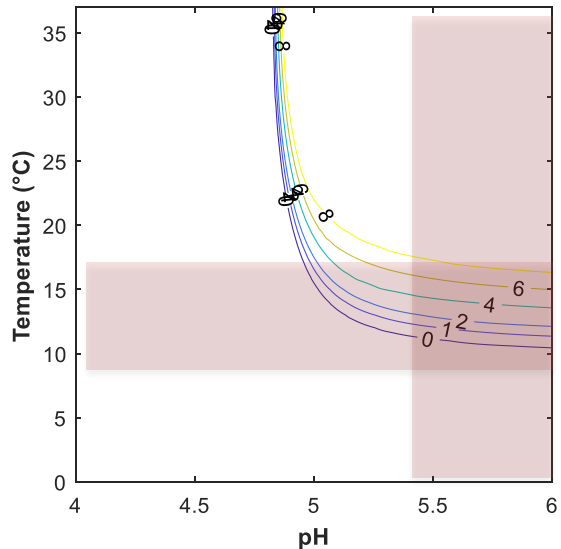


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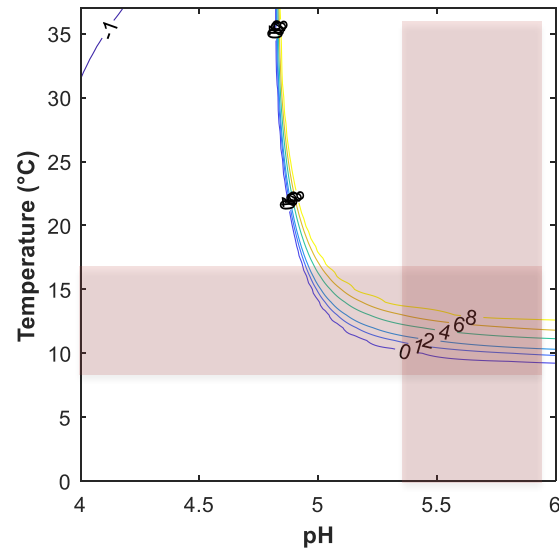
2 days



15 days

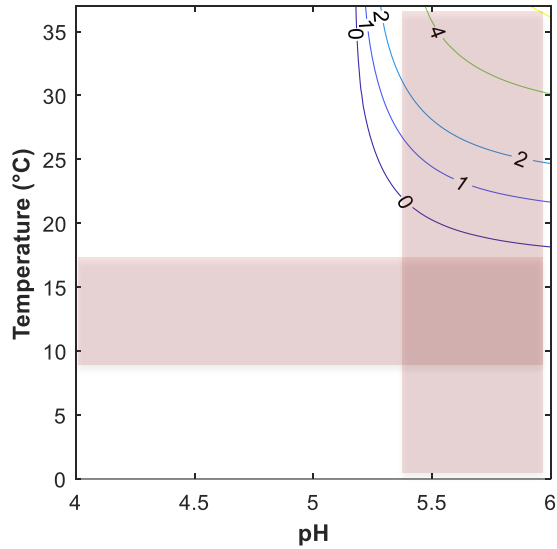


35 days

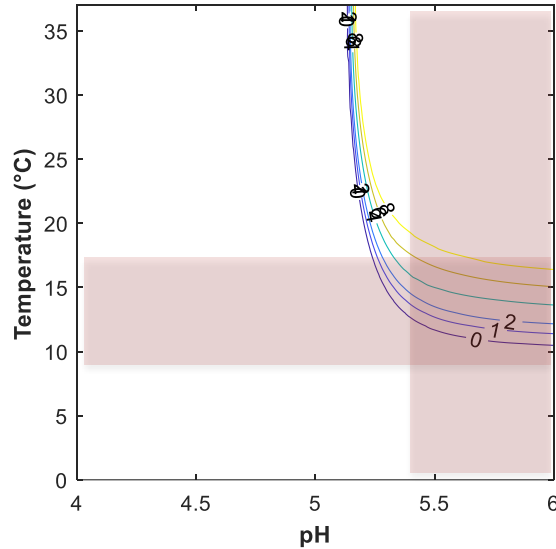


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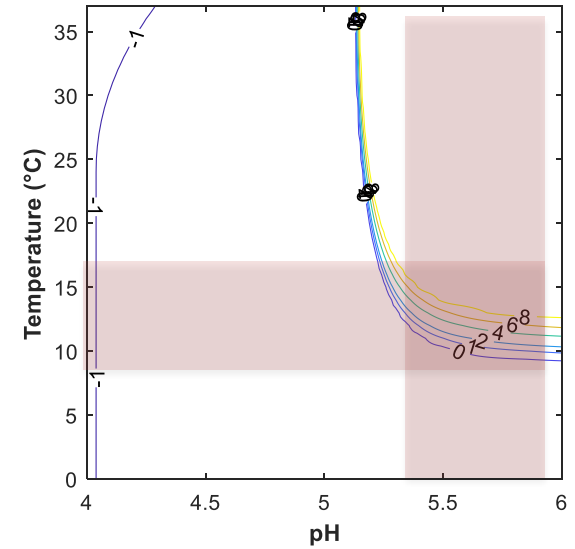
2 days



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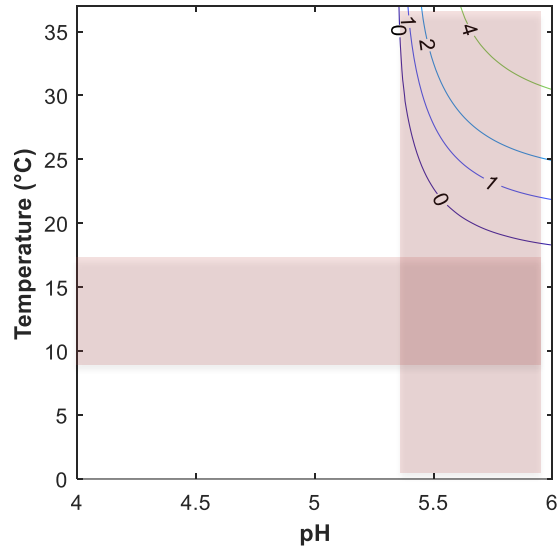


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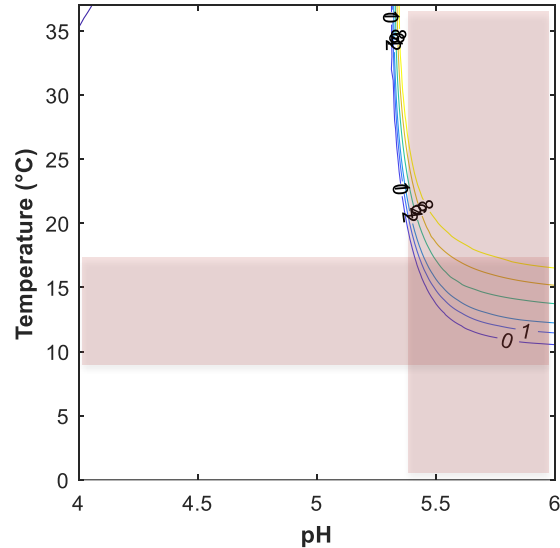


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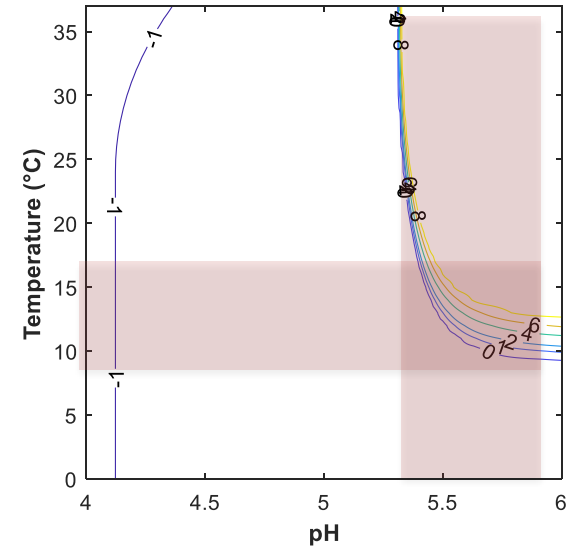
2 days



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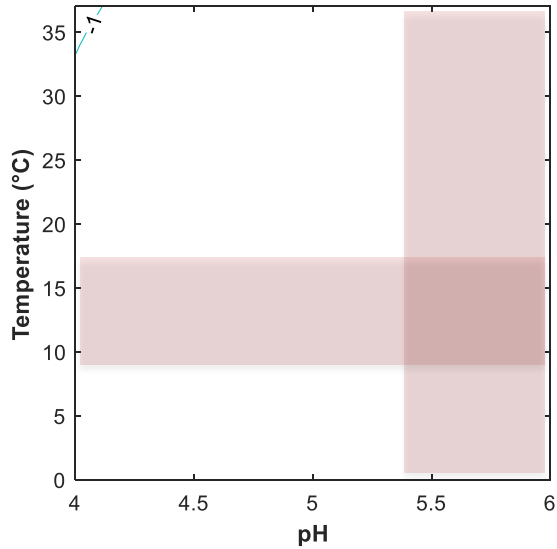


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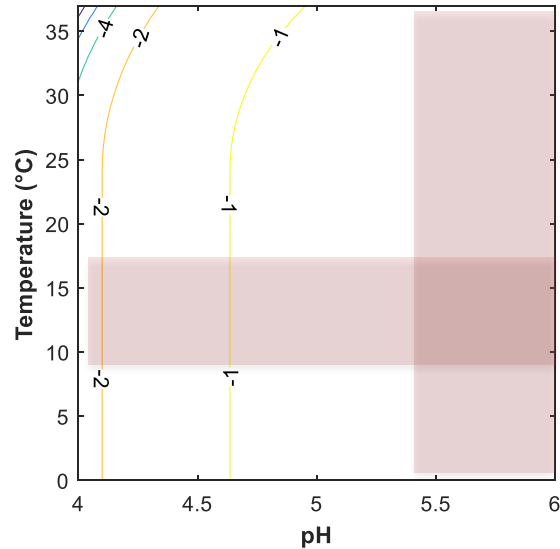


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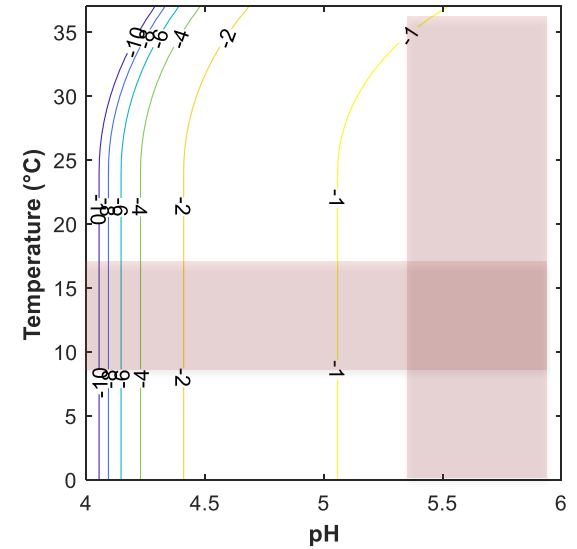
2 days



15 days



35 days



AH=1.5 % lactic acid
aw=**0.95** ⇔ 10 % Weigh loss

Conclusions

Whatever dynamic of the process parameters,
the growth rate or the resistance of *Salmonella* is:

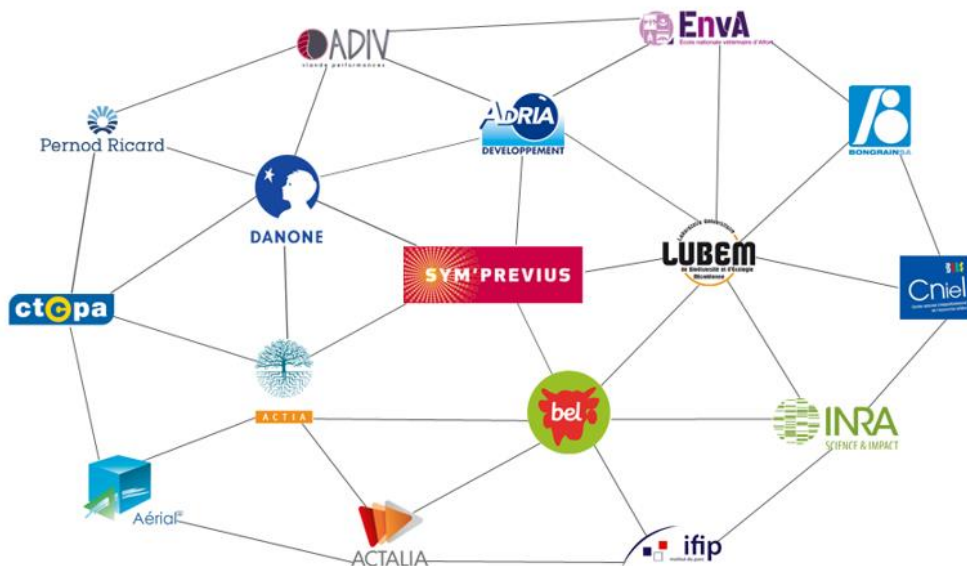
- **not dependent on the salt content**
 - the salt content impacts the pH, acid, and a_w kinetics but not the growth potential or the resistance.
- **dependent on the sugar contents**
 - High sugar contents could be linked to a higher *Salmonella* growth
- **dependent on the starter type**
 - Moderate growth rate starter could be linked to higher *Salmonella* growth rate and resistance

Conclusions

One combined model for growth and inactivation

- « Easy to use » 😊
 - Biological parameters, expert opinions...
- Specificity of the food effect 😊
- Parameters consuming 😞
 - Parameters must be known for growth and inactivation
- Improvements: physiological states of cells, link between resistance and growth potential, specie, strain and cellular variability...

A network of Food Business Operators, Food Technology Institutes and Academics



HACCP Assistant

Growth/ No Growth Interface

Growth Simulation

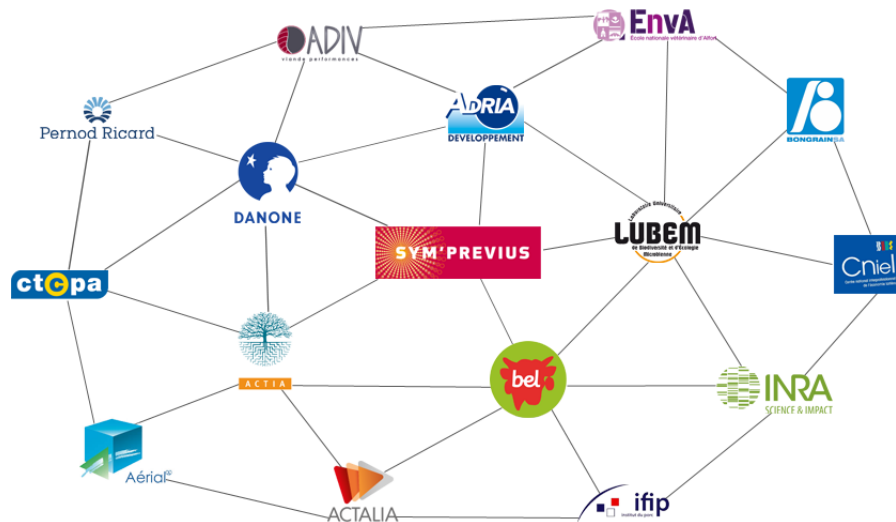
Thermal inactivation

Available in English, Spanish and French

symprevius@adria.tm.fr



Université de Bretagne Occidentale



Thank you for your attention