



上海理工大學

UNIVERSITY OF SHANGHAI FOR SCIENCE AND TECHNOLOGY

食品微生物安全的风险评估

Microbial Risk Assessment for Food Safety

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目 录

- 食品安全风险
- 预测微生物学概念和发展
- 基于预测微生物学的定量风险评估
- 结论与展望



致病菌导致食物中毒

卫生计生委每年公布的食物中毒事件报告中，2015年微生物性食物中毒事件的报告起数和中毒人数仍为最多，分别占食物中毒事件总起数和中毒总人数的**33.8%**和**53.7%**。

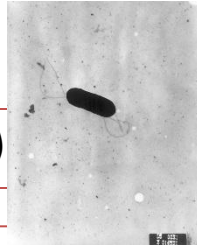
致病菌导致的食源性致病菌是世界公认的主要食品安全问题之一，远比消费者和媒体关注的假冒伪劣和制假掺假问题更为严峻。



食源性致病微生物



蜡状芽孢杆菌(*Bacillus cereus*)



单增李斯特菌(*Listeria monocytogenes*)

空肠弯曲杆菌(*Campylobacter jejuni*)

Norwalk病毒

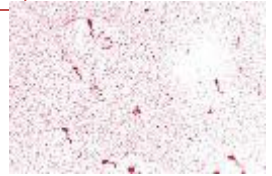
肉毒梭菌(*Clostridium botulinum*)



沙门氏菌(*Salmonella*)

产气梭菌(*Clostridium perfringens*)

耶尔森氏菌(*Yersinia*)



小球隐孢子虫(*Cryptosporidium parvum*)

葡萄球菌(*Staphylococcus*)



志贺氏菌(*Shigella*)



大肠杆菌O157:H7(*Escherichia coli* O157:H7)

弓形虫(*Toxoplasma*)

蓝氏贾第鞭毛虫(*Giardia lamblia*)



弧菌(*Vibrio*)

甲肝(*Hepatitis A*)





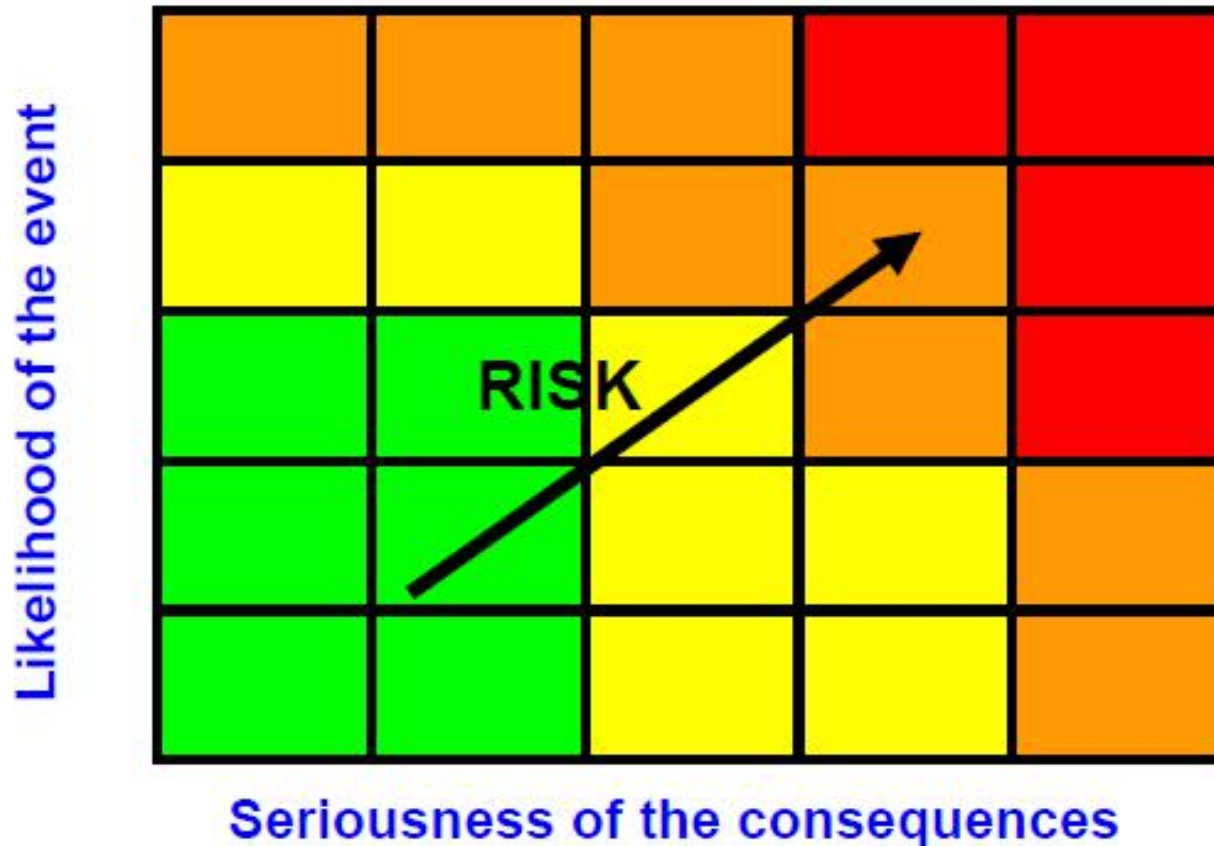
食品安全问题的两个途径

- 大多数人支持——零风险选择，即“黑与白”的方法，要求100%安全，没有风险，不惜一切代价。
- 很多人不理解——没有100%安全的食品，目标是将食品风险降到可接受的水平。





➤ 什么是风险？



Risk = Probability * Severity



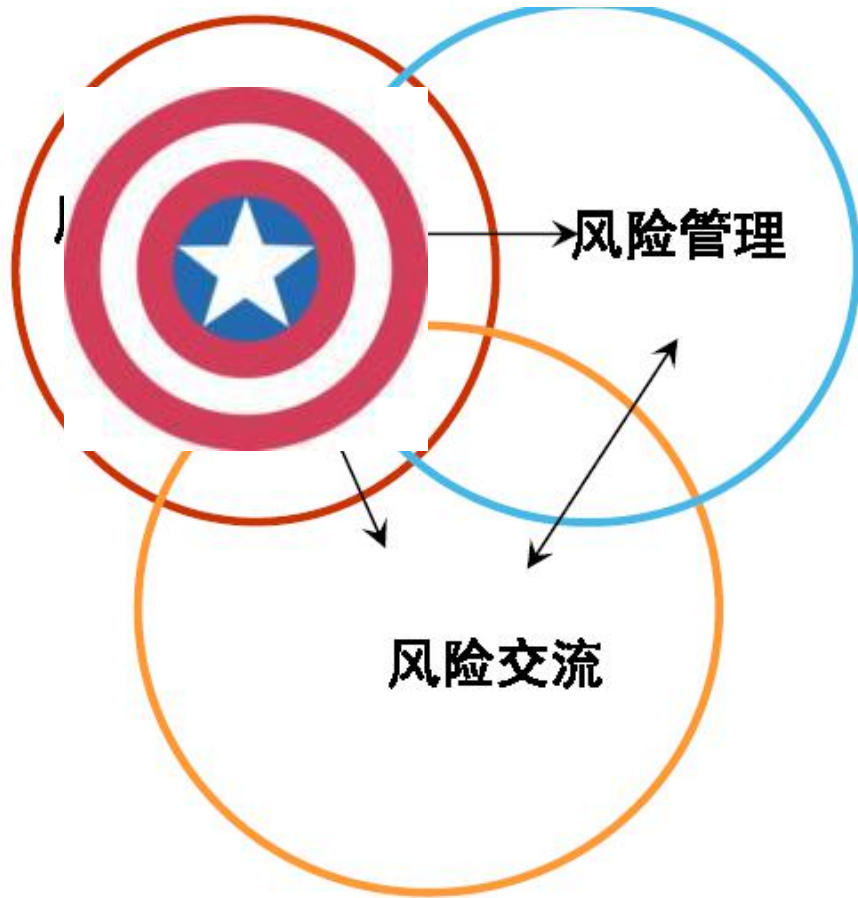
当前食品安全研究领域

Hazards Control
e.g. GMP, HACCP etc

Risk Assessment
e.g. microbial risk
assessment (MRA)

**Food
Safety**

Hazards Detection
e.g. Rapid detection



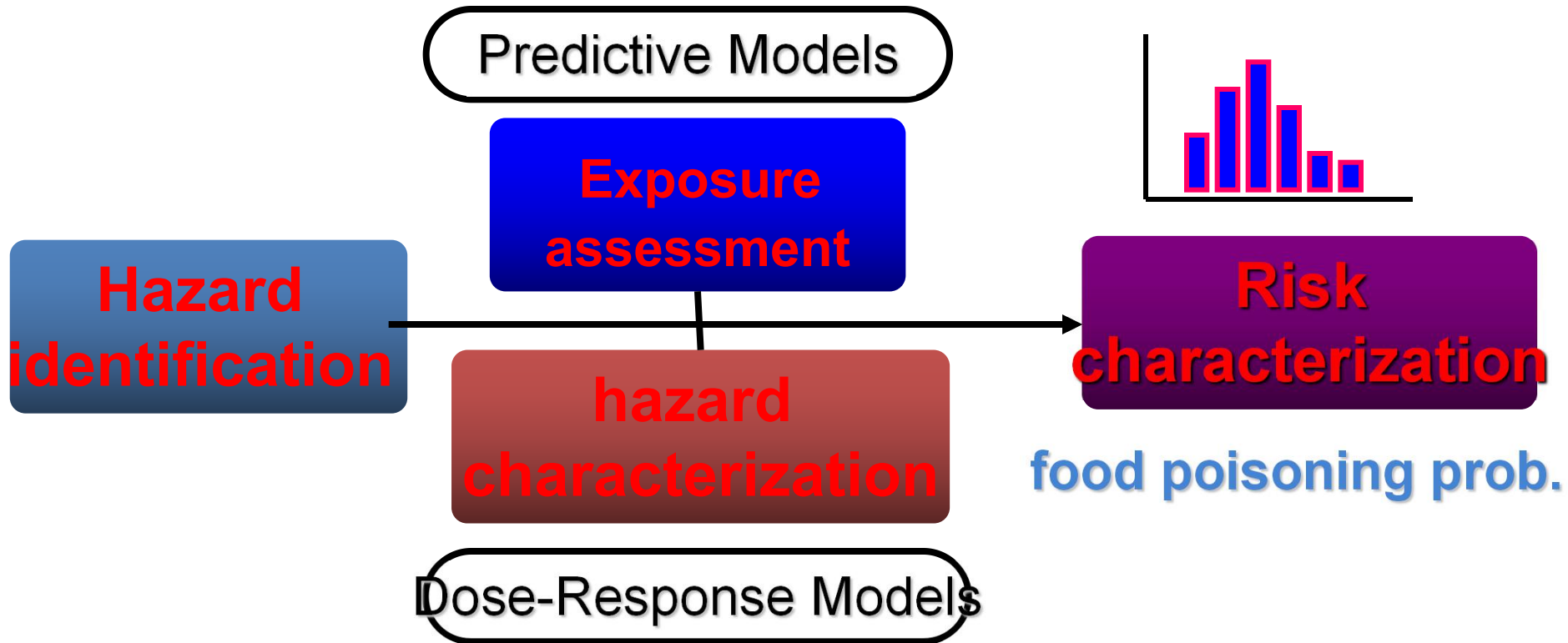
2006年以前

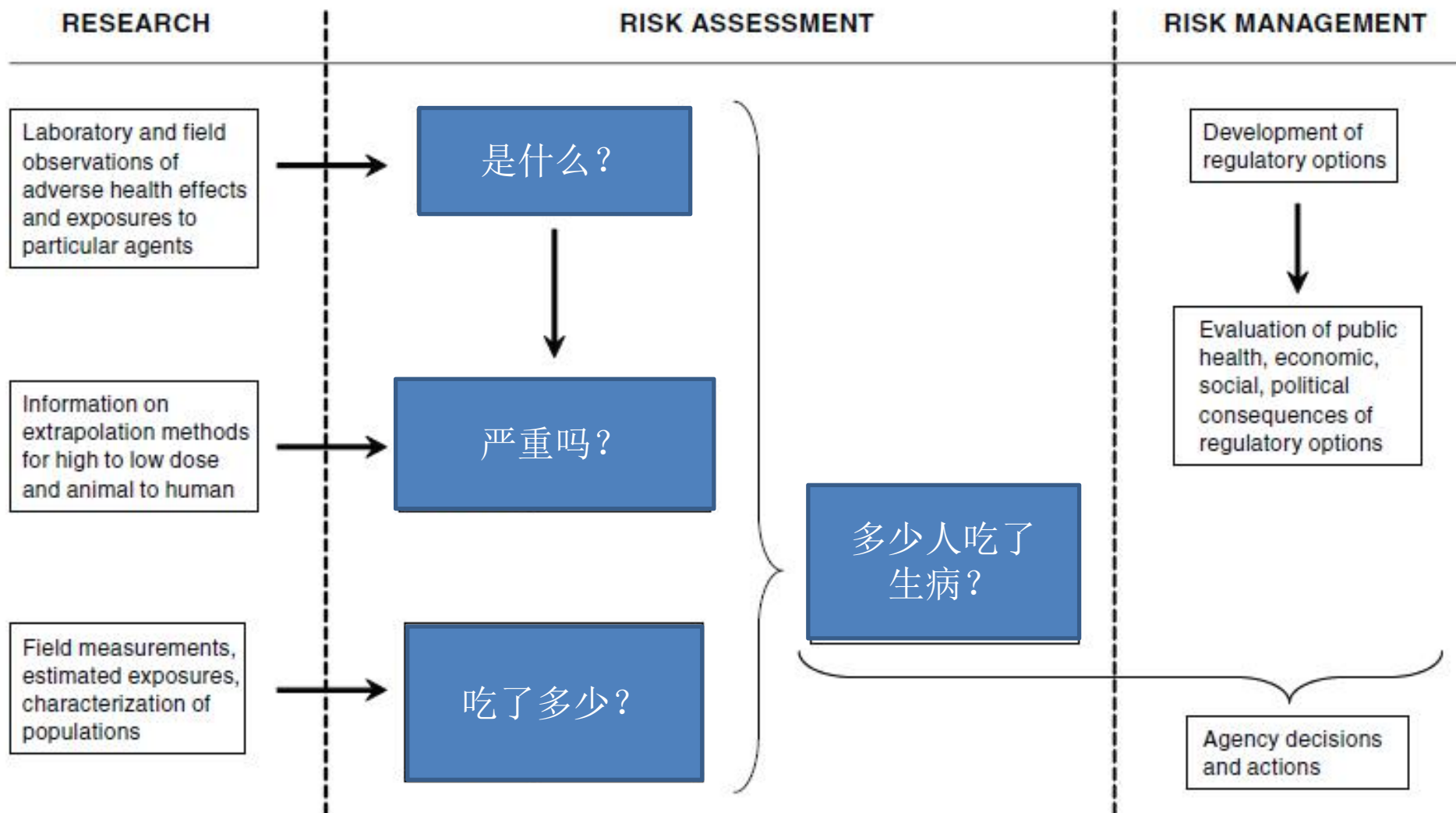


2006年以后



Risk Assessment

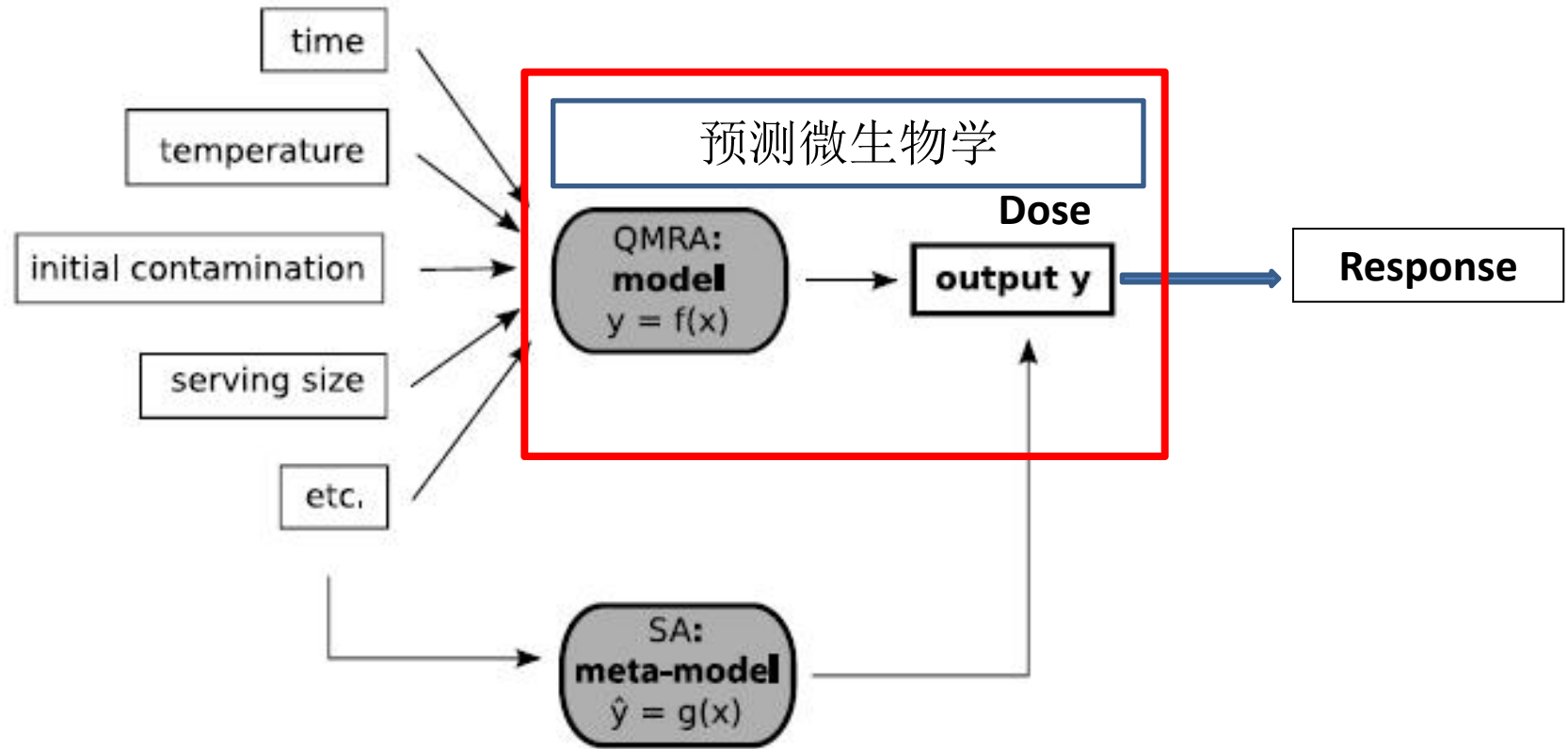




Risk-assessment–risk-management paradigm
(National Research Council, USA, 1983)



➤ Quantitative Microbiological Risk Assessment (QMRA)



SA - Sensitivity Analysis

Adapted from Busschaert et al. 2011





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历史发展

- Heat treatments in closed jars can largely increase the shelf life and safety of foods was described by **Appert** (1810);
- The mechanism through inactivation of microorganisms was described by **Pasteur** around 1860;
- Until the 1920s the sterilization and pasteurization became apparent in (national) legislations.



A French postage stamp in honour of the great inventor Nicolas Appert.



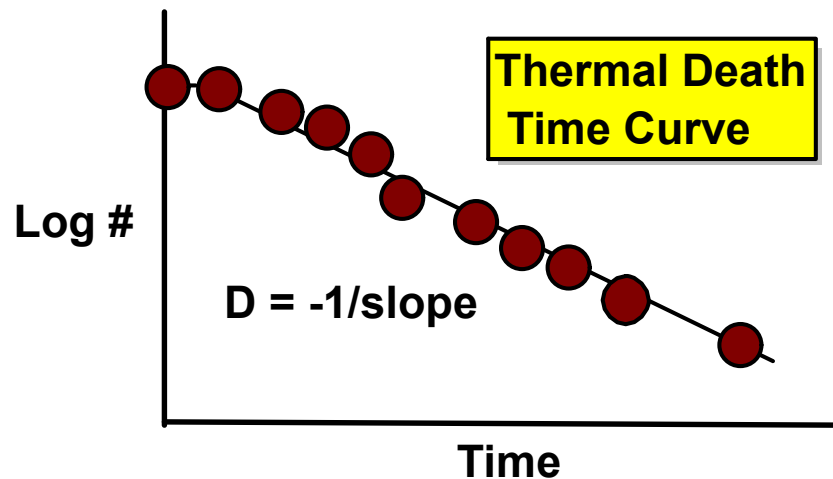
19th century can of soup c.1856.



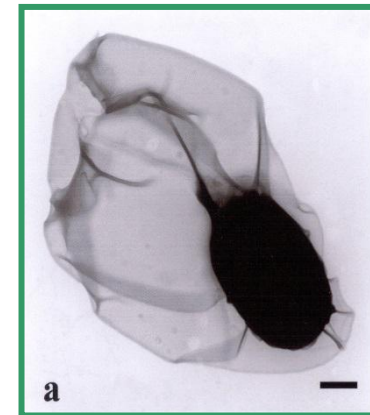


历史发展

- Emergence during the 1930's as a means of describing the inactivation of microorganisms during thermal processing



(Buchanan RL, 2003)



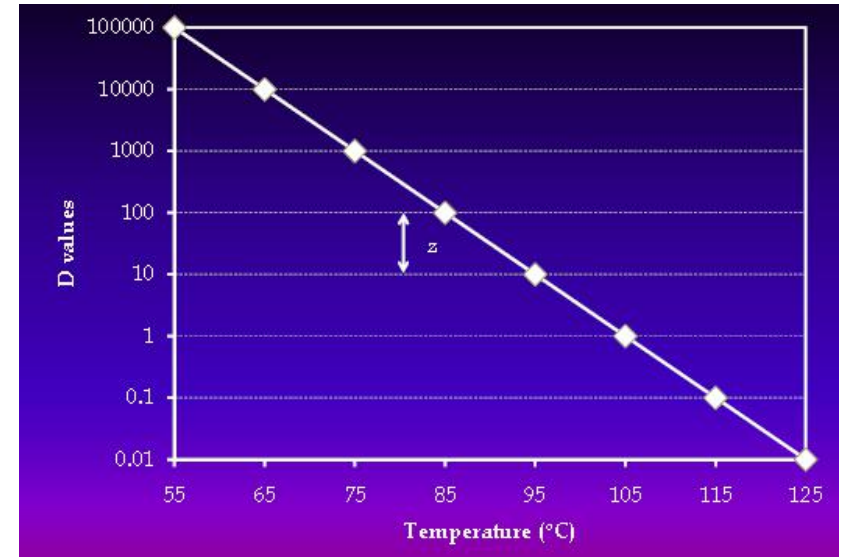
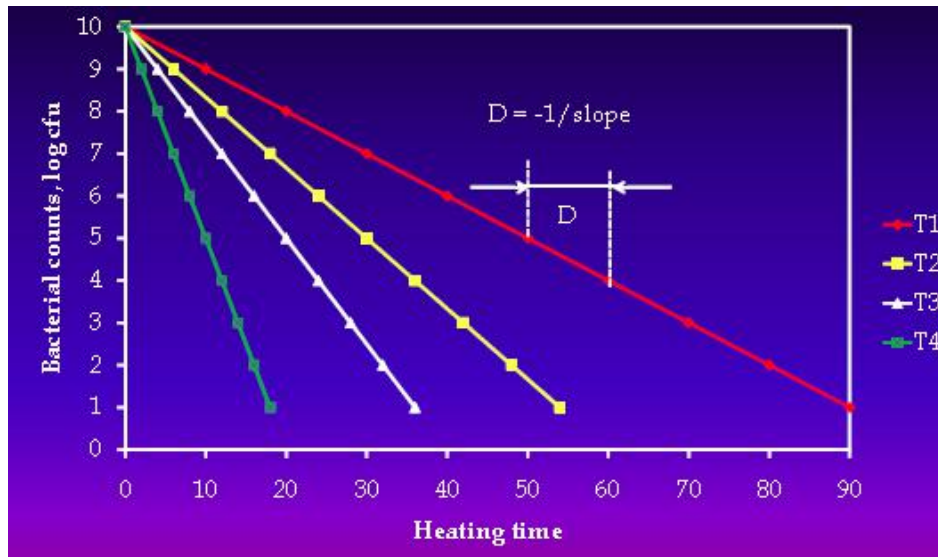
Clostridium botulinum

(Peck M, 2008)



历史发展

- Emergence during the 1930's as a means of describing the inactivation of microorganisms during thermal processing



$$\log(C) = \log(C_0) - \frac{t}{D}$$

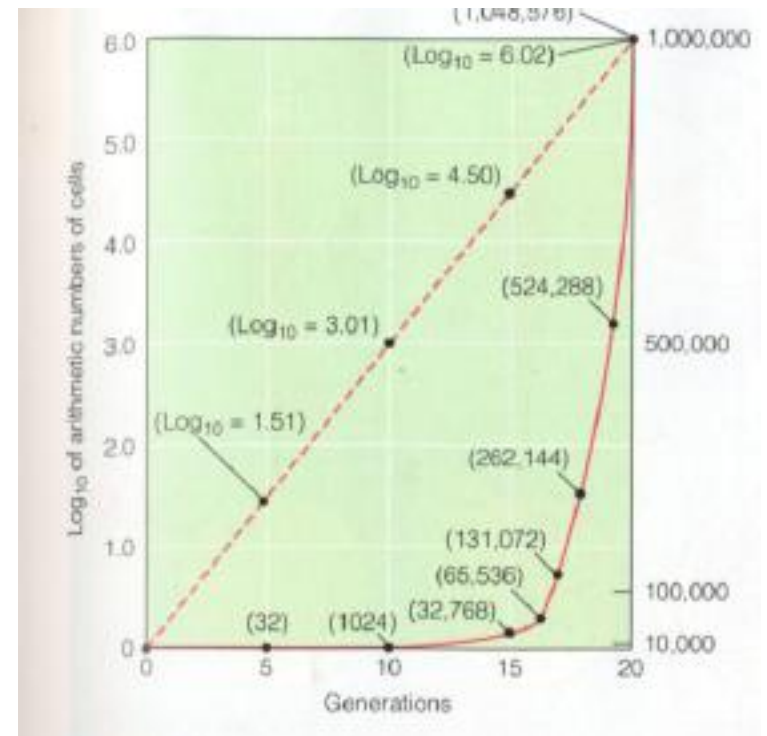
$$\log(D) = \log(D_0) - \frac{T}{z}$$

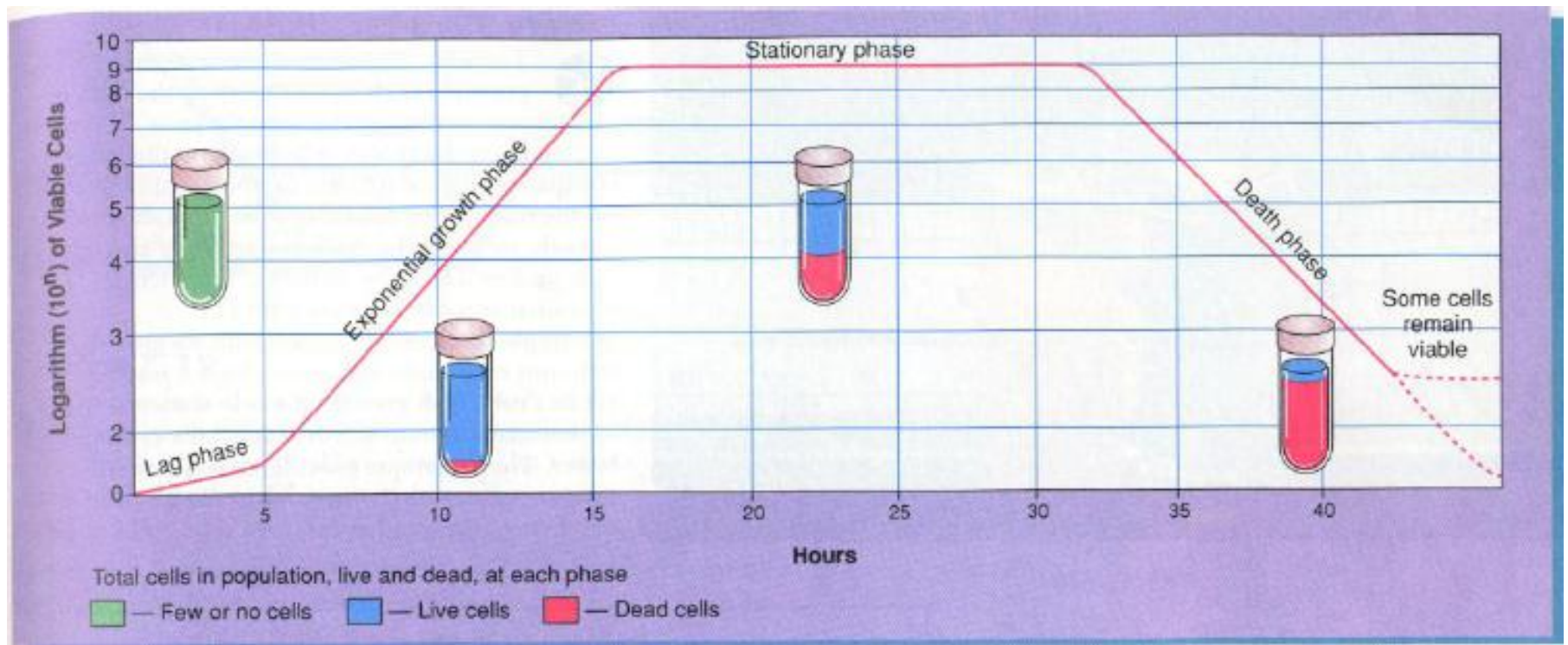


预测微生物学

- 单细胞微生物按照二分裂式指数型生长 (2^n)

<u>Generation Number</u>	<u>Arithmetic Number of Cells</u>	<u>Log₁₀ of Arithmetic Number of Cells</u>
0	1	0
5 (2^5) =	32	1.51
10 (2^{10}) =	1,024	3.01
15 (2^{15}) =	32,768	4.52
16 (2^{16}) =	65,536	4.82
17 (2^{17}) =	131,072	5.12
18 (2^{18}) =	262,144	5.42
19 (2^{19}) =	524,288	5.72
20 (2^{20}) =	1,048,576	6.02

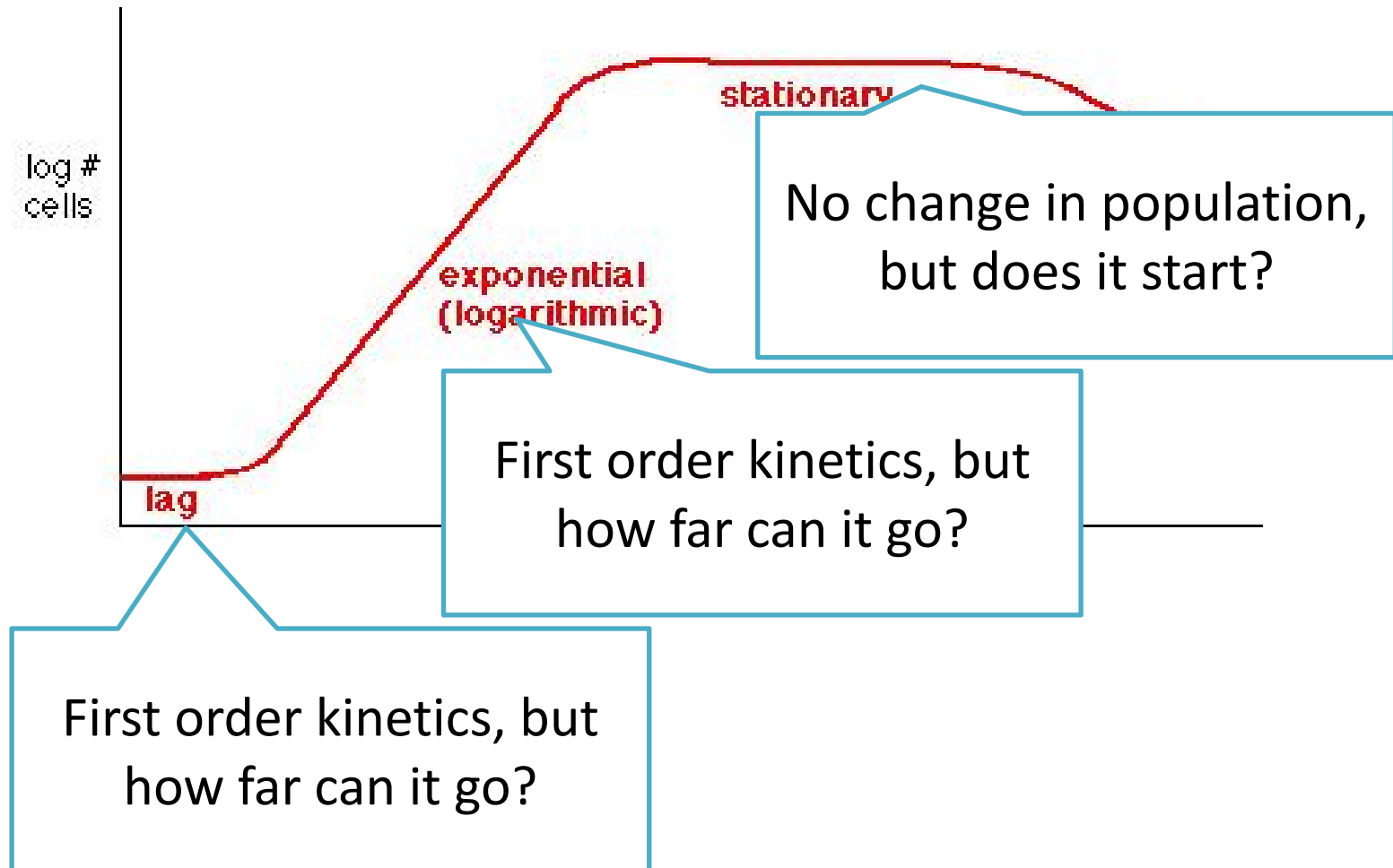




如果微生物生长规律是固定的，可以建立模型来预测其生长，从而更好的监测和控制微生物。



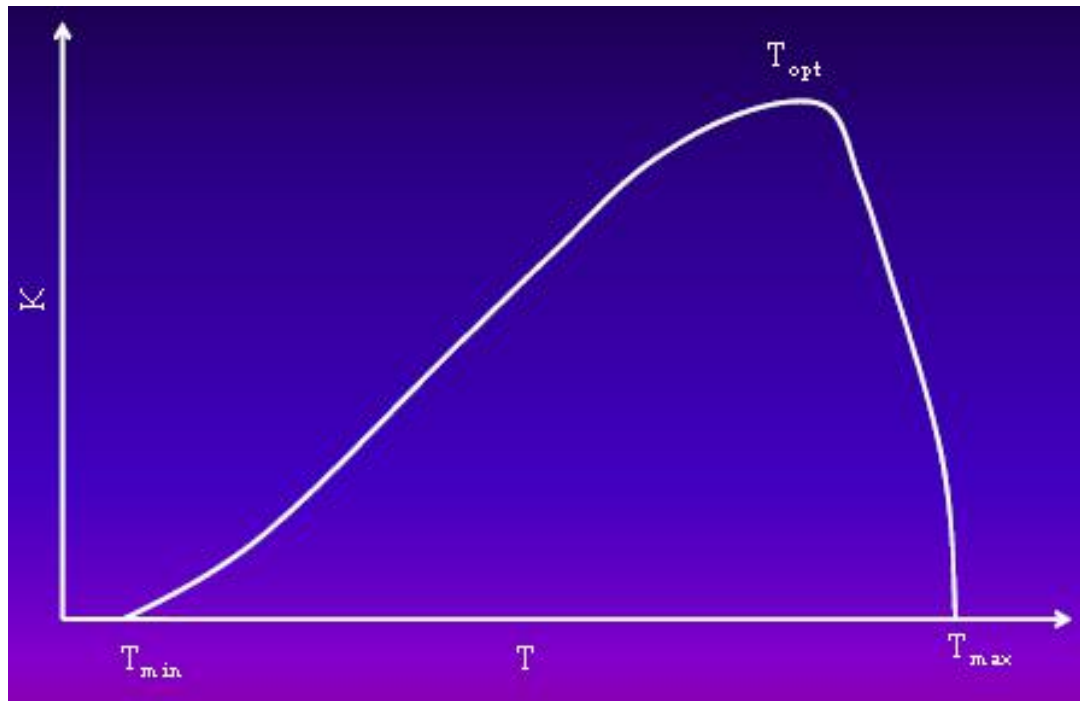
微生物生长的四个阶段





历史发展

The uniqueness of microbial growth is **temp.**-dependent.
Limited progress of applying temp.-based **Arrhenius** model partly results in little progress prior to 1980s.



$$K = K_0 e^{-\frac{E_a}{RT_K}}$$

$$\log(D) = \log(D_0) - \frac{T}{z}$$



历史发展

- Expanded greatly during the 1980's and 1990's due to computer science including modeling of
 - Growth
 - Survival
 - Inactivation
 - Competition
 - Food Unit Operations





预测微生物学

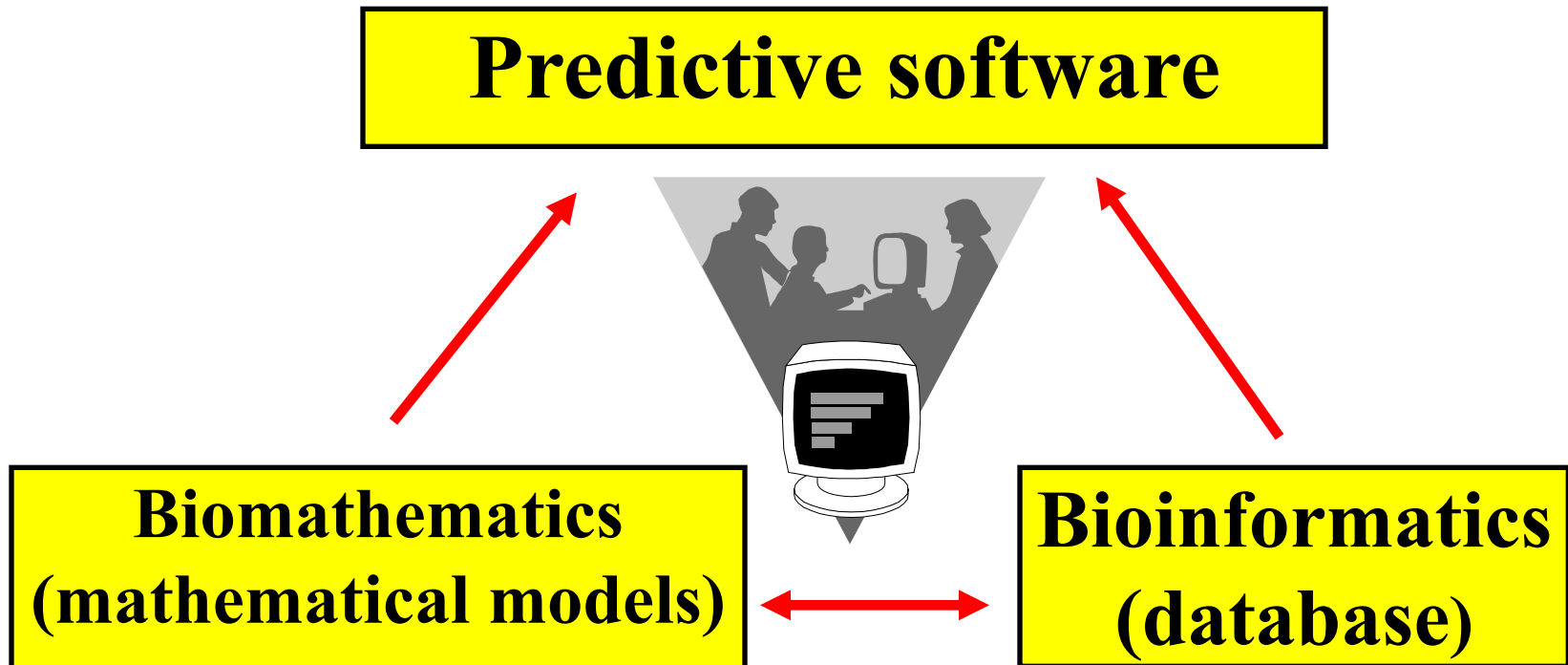
“quantitative microbial ecology of foods”

- ***provides a solution to being “data rich but knowledge poor”***
- ***synthesises*** microbial population responses to environmental factors - temperature, pH, water availability
- finds application in ***replacing challenge tests*** and in supporting the paradigms of food safety management, HACCP, Risk Assessment, FSO's
- constructed from systematic literature reviews or de novo experiments which reveal patterns of microbial behaviour
- indicates regions of physiological interest



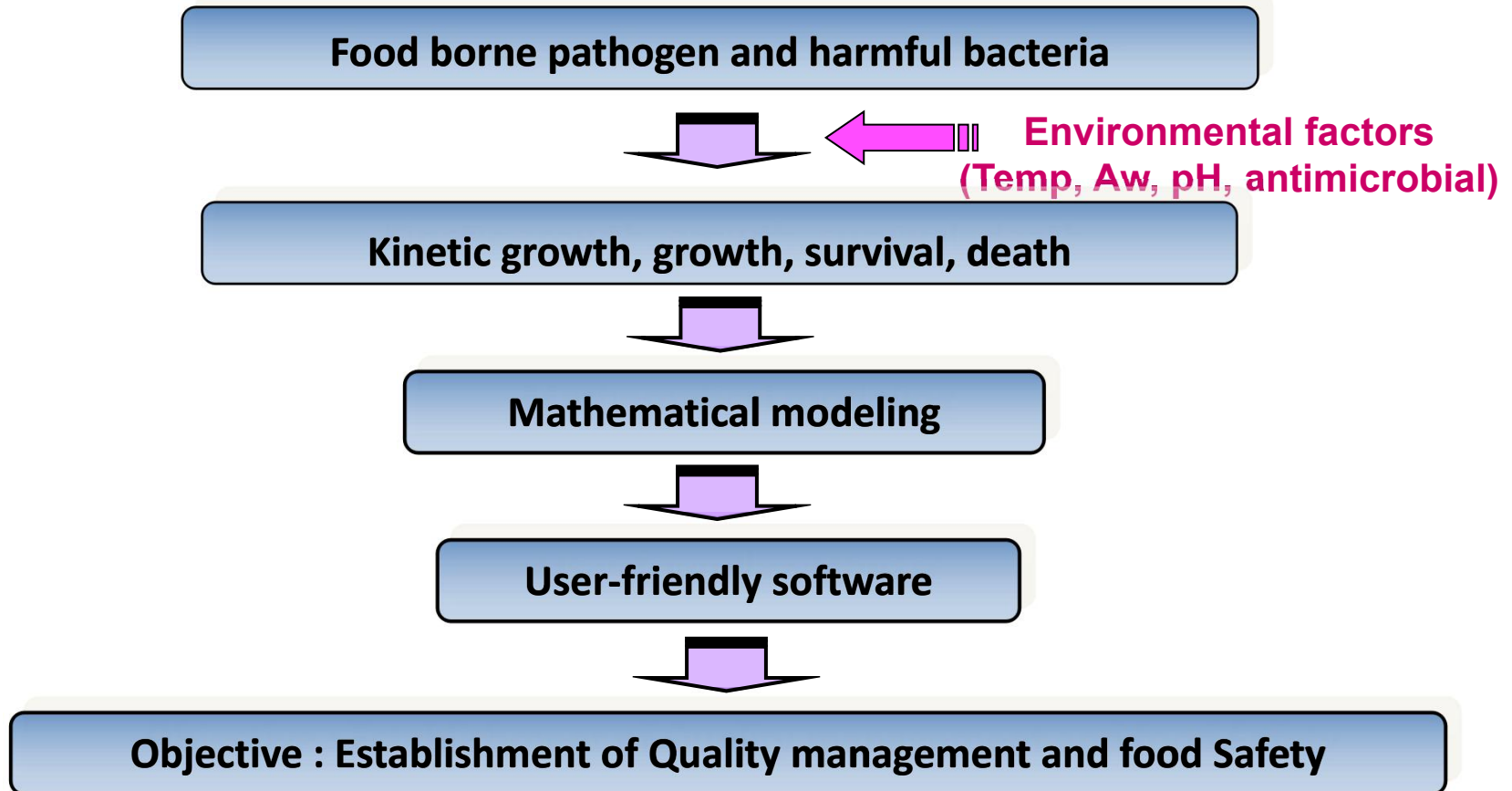
预测微生物学

More methods has been involved from beginning of 21 century



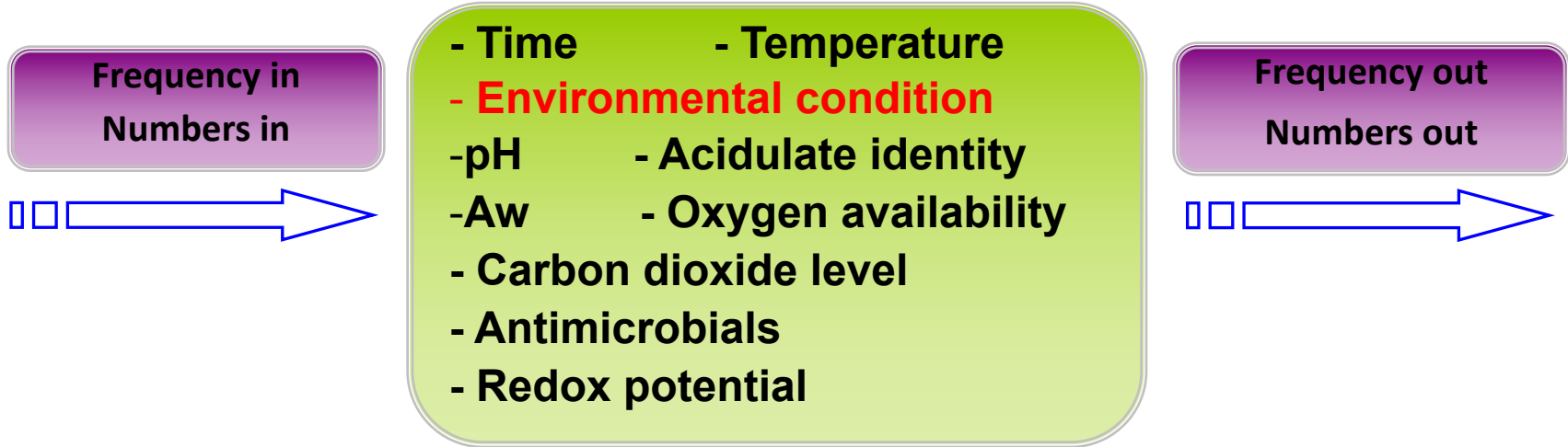


预测微生物学





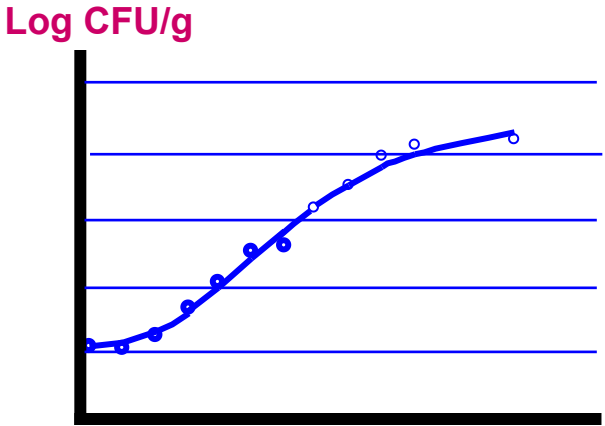
Microbial unit operations in food



Growth, Survival, Death

Predictive Model
(Gompertz Equation)

$$\text{Log CFU/ml} = A + C e^{(- e^{-B(t - M)})}$$





Several Famous Units Worldwide





Types

- kinetic models for spoilage prediction and probability models to address risks posed by *C. botulinum* and other toxigenic microorganisms (Roberts *et al.*, 1981)
- Devlieghere *et al.* (2006) suggested that the modelling needs to shift from a kinetic model, when the environmental conditions still support growth, to a probability model when the growth/no growth interface is reached, demonstrating the near link between the two modelling types.



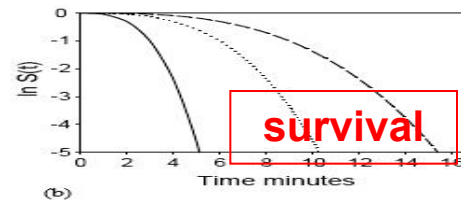
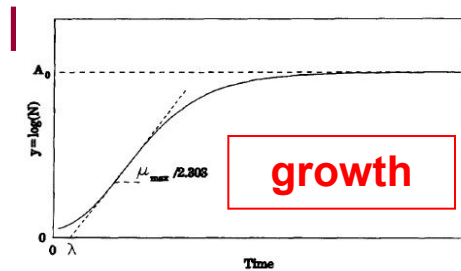
Types continued

- Devlieghere *et al.* (2006) also gave a succinct account of model types: **empirical versus mechanistic models**,
- **primary, secondary and tertiary models**,
- **thermal inactivation and non-thermal inactivation models.**

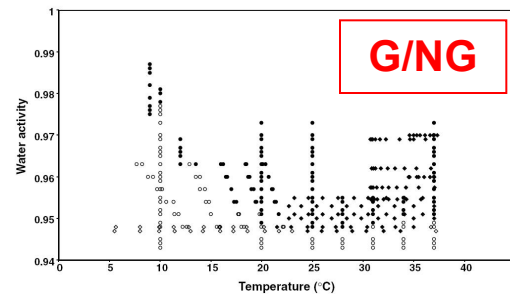
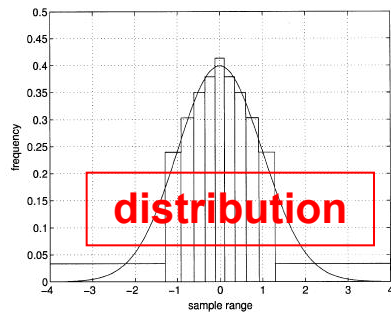


Types of PFM models

- deterministic



- probability models



Relation of two models?



More usual classification of PFM models

- primary, secondary and tertiary models
- Primary to describe the response of microorganisms to a single set of conditions *over time*;
- Secondary to describe the effect of environmental conditions, e.g., physical, chemical, and biotic features, on the values of the parameters of a primary model
- Tertiary refers to some computer database, e.g. USDA Pathogen Modelling Program, UK Food MicroModel, and now into a common database named ComBase <http://www.combase.cc/>





PFM models

Primary models	Secondary models	Tertiary models
Gompertz function	Belehradek model (<i>square-root model</i>)	USDA Pathgen Modelling Program
Modified Gompertz	Rathkowsky model (<i>square-root model</i>)	Food MicroModel
Logistic model	Arrhenius model	Pseudomonas Predictor
Baranyi model	Modified Arrhenius models (<i>Davey or Schoolfield</i>)	Expert Systems
First-order monod model	Probability models	
Modified monod model		
D values of thermal inactivation	Z values	
Growth decline model of Whiting and Cygnarowicz	Polynomial or response surface models	
Three-phase linear models	Williamw-Landel Ferry model	



Primary, Secondary and Tertiary models

Primary models

Limited conditions

$$y_{T1} = f_{T1}(t)$$

$$y_{T2} = f_{T2}(t)$$

⋮

$$y_{Tn} = f_{Tn}(t)$$

$\{\mu\}$

$$y_{Tn-1} = f_{Tn-1}(t)$$

$$y_{Tn} = f_{Tn}(t)$$

Secondary models

General information

$$\mu(T) = g(T, pH, A_w)$$

General application

Tertiary models

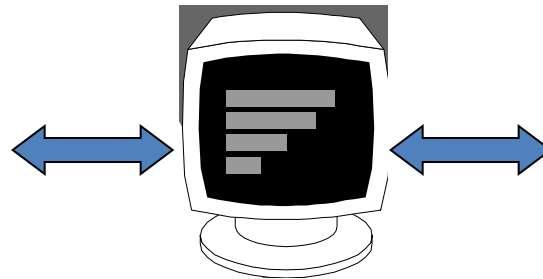




预测模型有什么用？

- 以简化等式来模拟微生物的实际动态变化
- 用于预测非检测条件下的可能情况

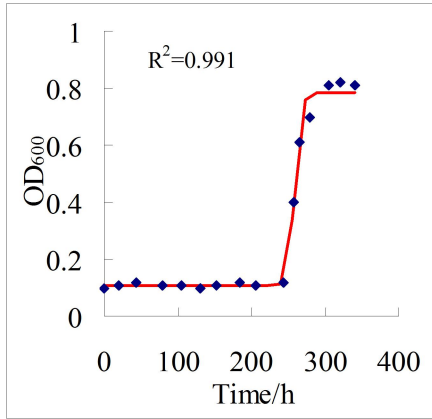
to turn data into knowledge



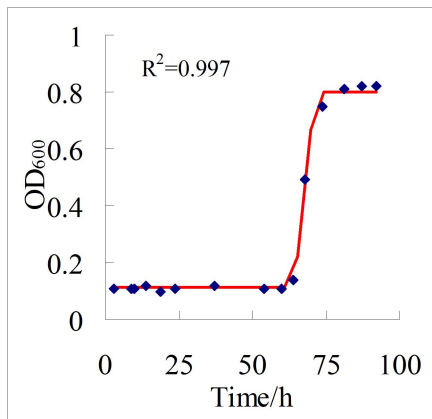


不同试验条件下生孢梭菌的生长模拟

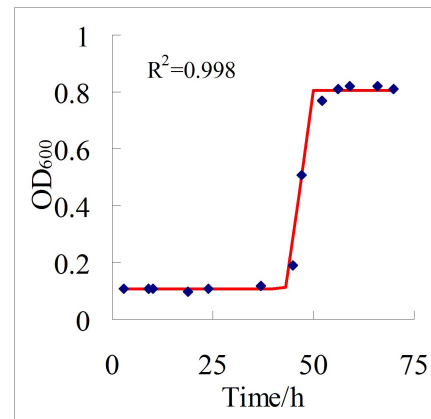
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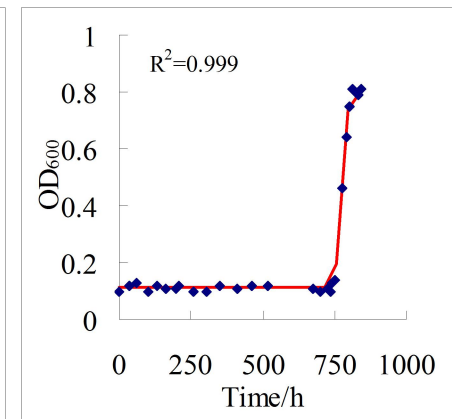
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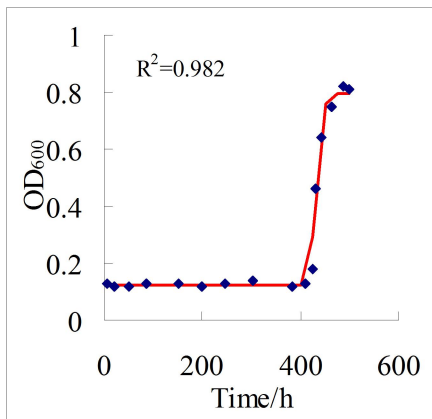
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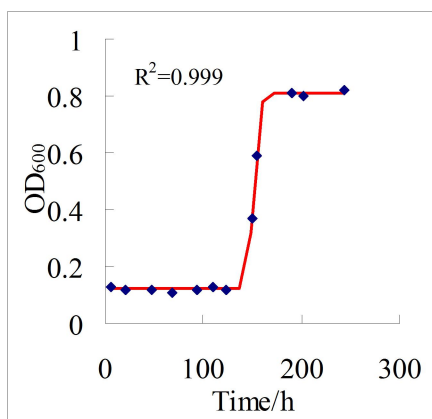
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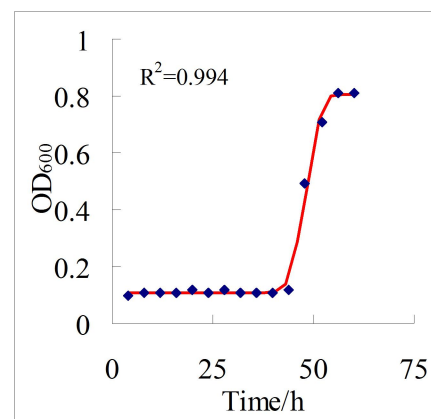
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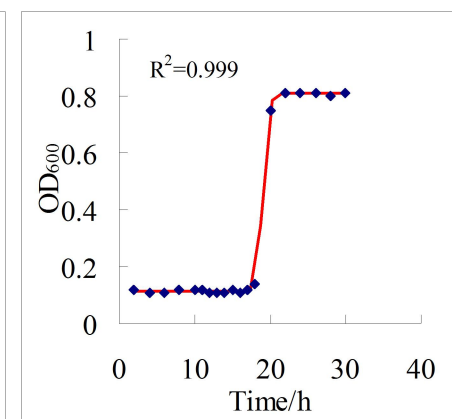
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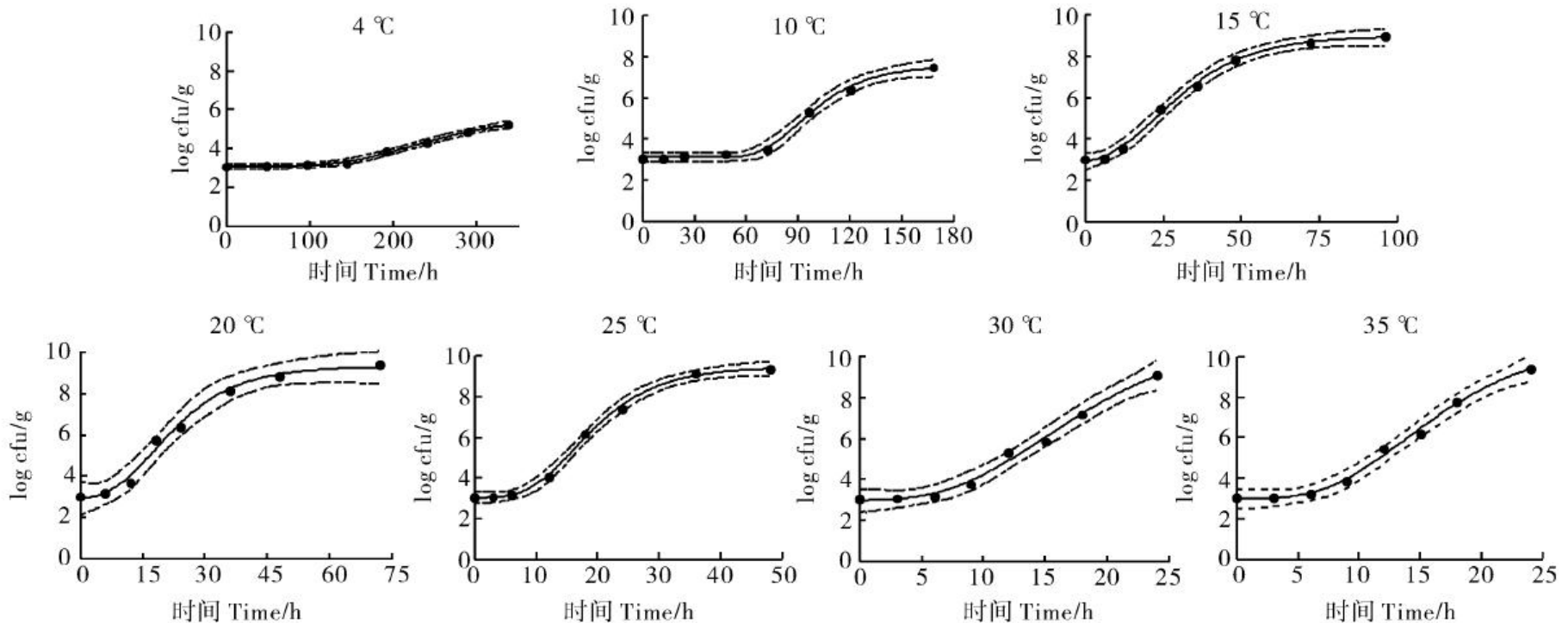


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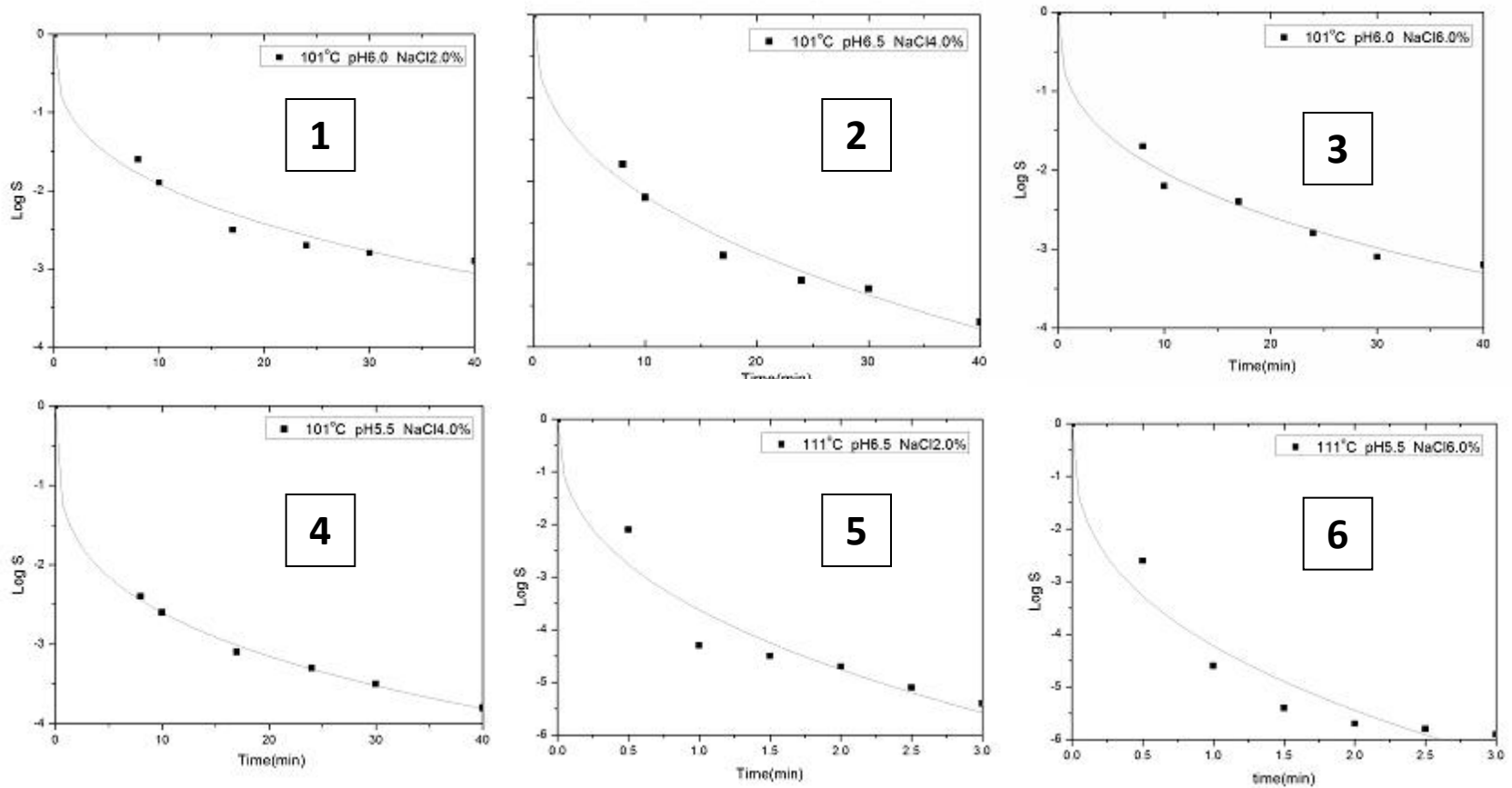


不同温度下单增李斯特菌的生长模拟



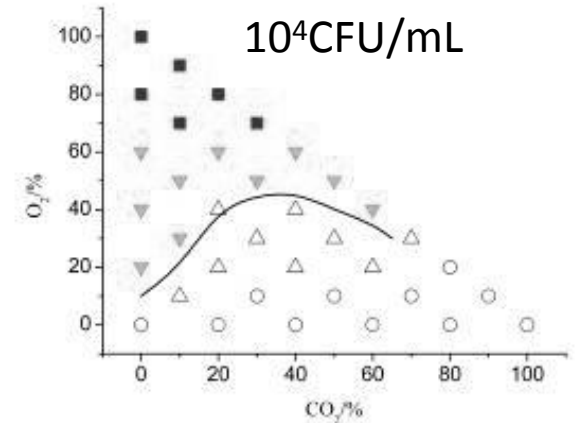
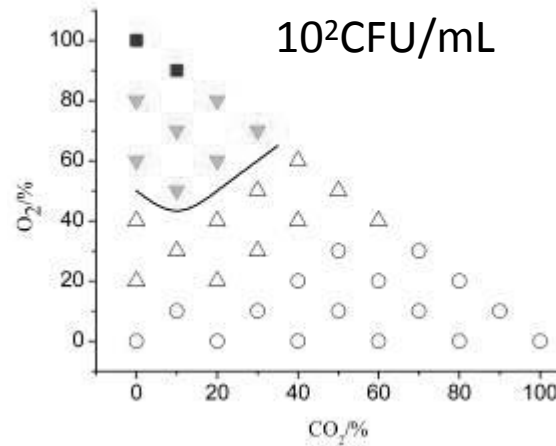
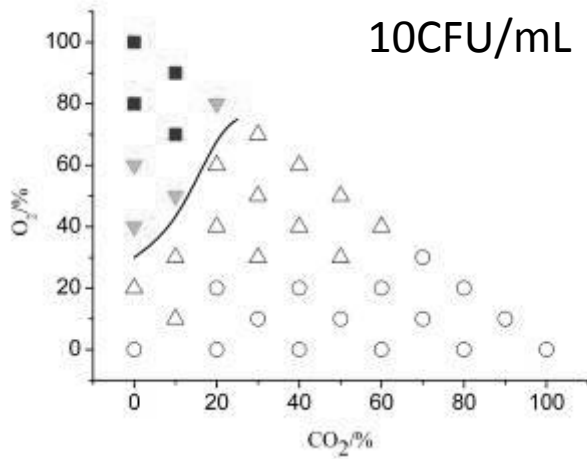


生孢梭菌孢子热失活的模拟





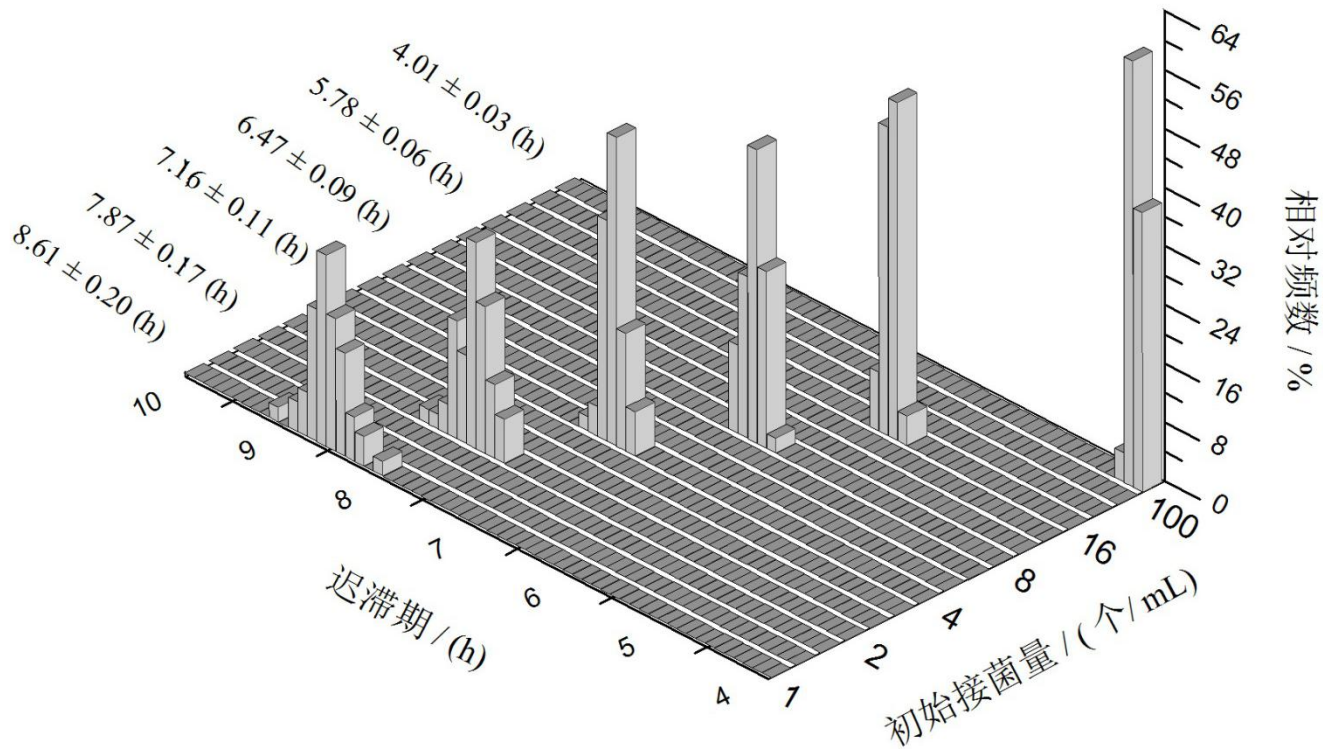
气调包装条件下假单胞菌的G/NG模拟



■ $P=1$, ▼ $P=0.66$, △ $P=0.33$, ○ $P=0$, — $P=0.5$



铜绿假单胞菌的迟滞期概率分布



Effect of different inoculum size on the lag time of *P. Aeruginosa* growth under 35°C

植物乳杆菌和单增李斯特菌单独及混合培养生长结果:

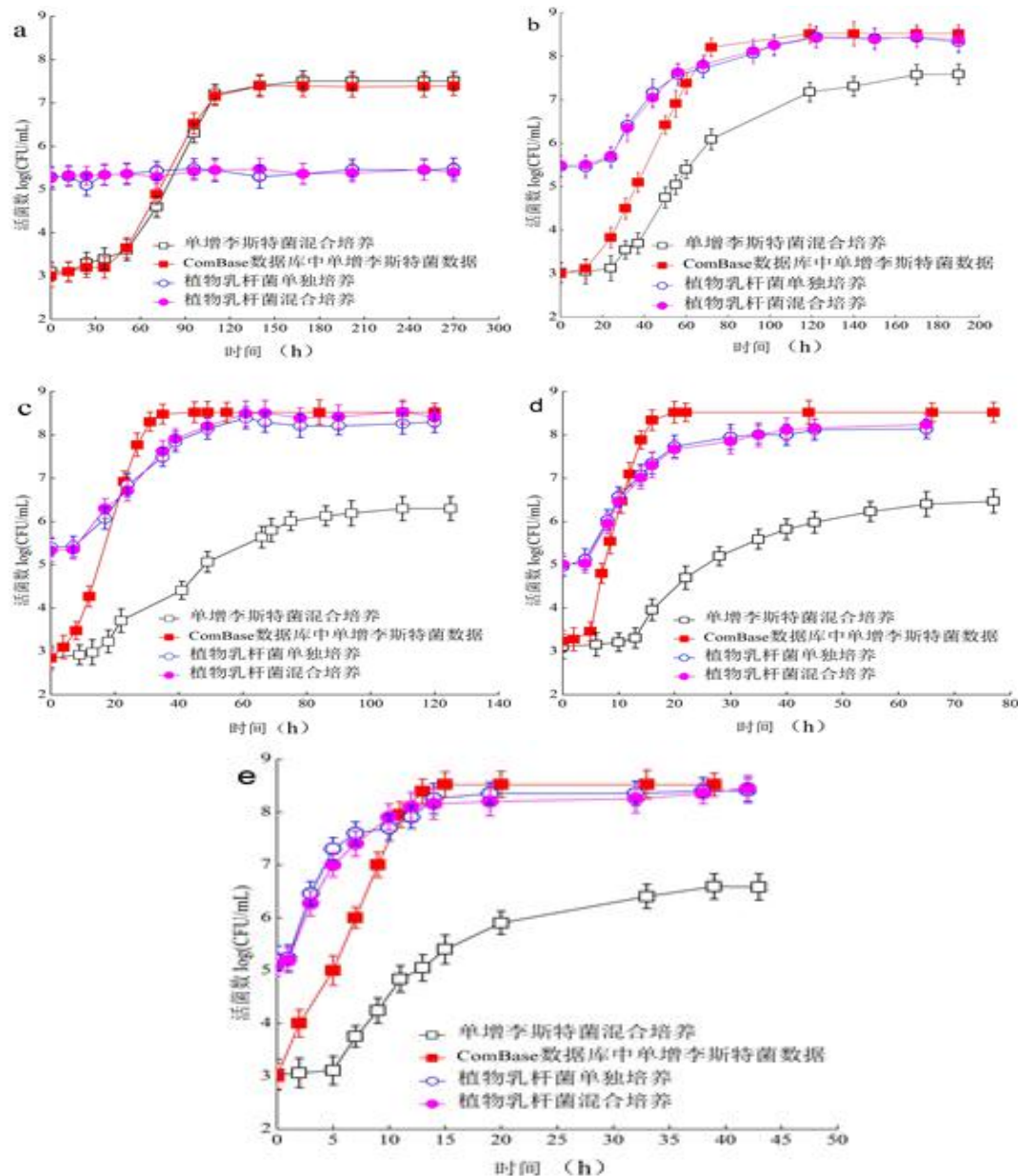
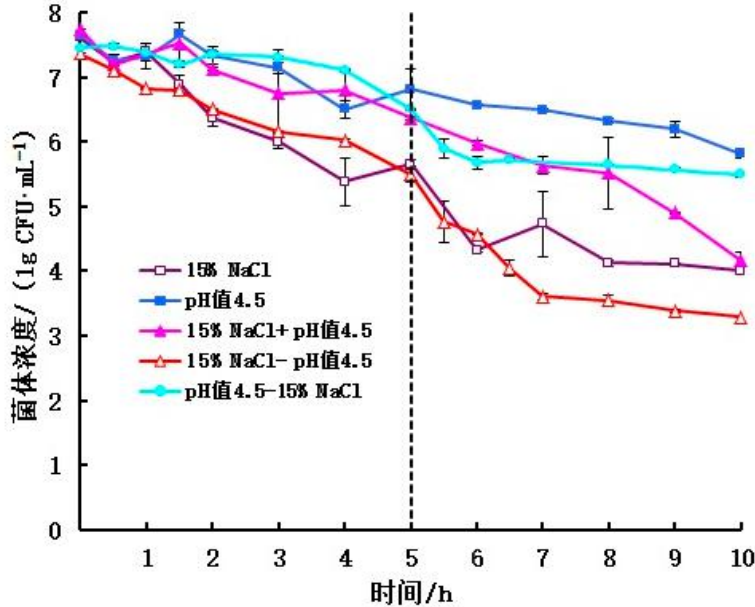


图1 不同温度下单独及混合培养时植物乳杆菌和单增李斯特菌的生长曲线, (a)7°C, (b)14°C, (c)21°C, (d)28°C和(e)35°C

Fig.1 Growth curves of *Lac. plantarum* and *Lis. monocytogenes* in pure- and mixed- culture at different temperatures



酸化和渗透压的不同处理顺序对肠炎沙门菌失活的影响



不同处理条件下肠炎沙门菌失活曲线拟合
失活动力学一级模型的参数估计值

处理条件	第1阶段 (0~5h)		第2阶段 (5~10h)			
	D/h	R ²		D/h	R ²	
		最大值	最小值		最大值	最小值
15% NaCl	2.086±0.157 ^a	0.971	0.941	2.487±0.020 ^a	0.861	0.850
pH 值 4.5	5.156±0.841 ^b	0.824	0.806	5.555±0.172 ^b	0.984	0.978
15% NaCl+pH 值 4.5	3.727±0.808 ^{bc}	0.917	0.819	2.585±0.116 ^c	0.996	0.912
15% NaCl-pH 值 4.5	2.652±0.085 ^c	0.989	0.987	1.767±0.079 ^d	0.913	0.887
pH 值 4.5-15% NaCl	7.832±0.116 ^d	0.785	0.783	3.912±0.193 ^e	0.807	0.798

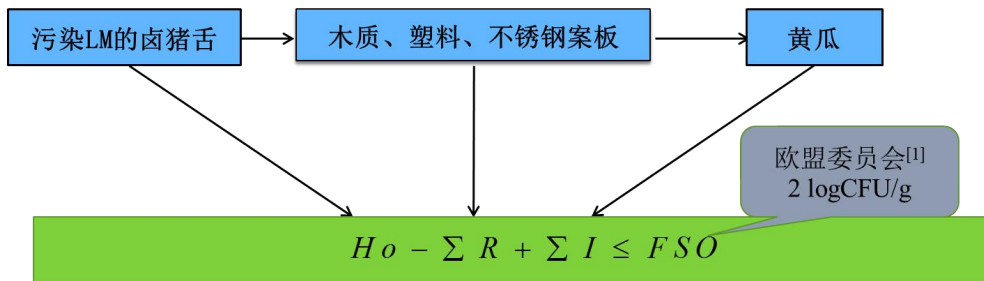
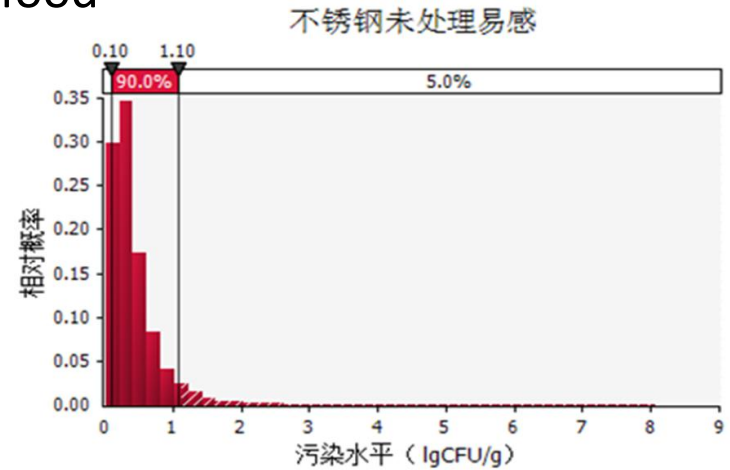
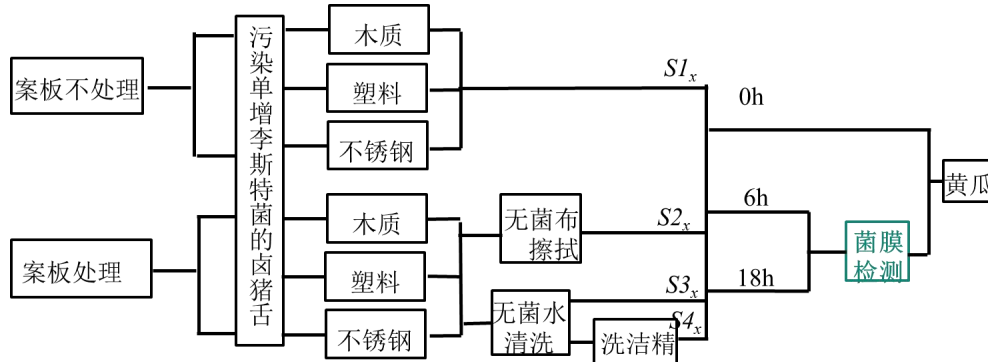
注：同列不同字母表示差异显著($p < 0.05$)。

不同处理下条件下沙门菌失活曲线图

通过分析5种不同处理条件对肠炎沙门菌失活效果的显著性、失活模型参数D值影响的显著性，进而得到15%NaCl-pH4.5是使肠炎沙门菌在25°C和10h内失活的最优处理条件。



Risk assessment of cross-contamination of *Listeria monocytogenes* from raw to cooked food



案板材质	处理方式	易感人群中单增李斯特菌发病概率 (每人/事件)	非易感人群中单增李斯特菌发病概率 (每人/事件)
木质	未处理表面	4.55×10^{-12}	4.15×10^{-12}
	处理表面	3.61×10^{-12}	3.28×10^{-12}
塑料	未处理表面	4.10×10^{-12}	3.74×10^{-12}
	处理表面	3.37×10^{-12}	3.06×10^{-12}
不锈钢	未处理表面	4.26×10^{-12}	3.89×10^{-12}
	处理表面	3.37×10^{-12}	3.07×10^{-12}

董庆利等, 农业机械学报, 2016
陆冉冉等, 现代食品科技, 2016

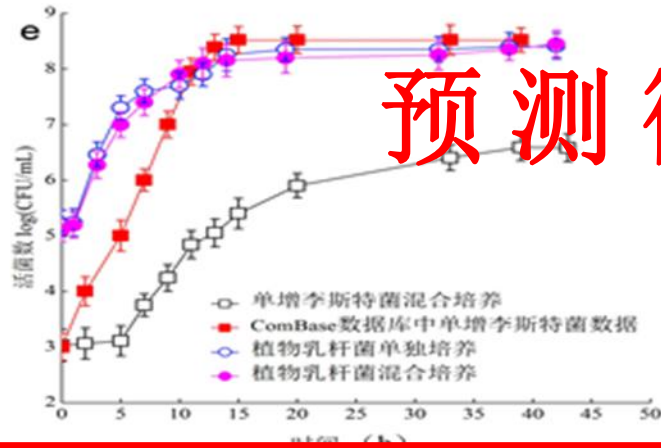
我们团队近年来的微生物建模进展

Strains	Media or foods	Sources
<i>L.monocytogenes</i>	LB	J. Huazhong Agri. Univ. (2010) (Chn)
	RTE lettuce	Sci. Tech. of Food Ind. (2010) (Chn)
	RTE salads	Food Science (2013a) (Chn)
	Cross-contamination of cutting board	Transactions of the CSAM (2016a) (Chn)
	Transfer modelling based FSO	Modern Food Sci. & Tech. (2016a) (Chn)
	Cc from meat to vegetables	Chinese J. Bioprocess Eng. (2016) (Chn)
	Jameson-effect with LAB	Modern Food Sci. & Tech. (2016b) (Chn)
<i>Salmonella</i> spp.	pH & NaCl treat sequence	Transactions of the CSAM (2016b) (Chn)
	Different physical conditions	Food Science (2016)
<i>Aeromonas</i> spp.	Chilled pork	Food Sci. (2011) & Ch J. Bioprocess Eng. (2012) (Chn)
	Injured in media	Food Sci. (2012a) (Chn)
	Shelf-life prediction in media	Food Sci. (2012b) (Chn)
	RSM in media	J. South China Agri. Univ. (2012) (Chn)
	Uncertainty and Variability	Food Sci. (2014a) (Chn)
	Cross contamination scenarios	Food Sci. (2014b) (Chn)
	Survival model & transfer matrix	Transactions of the CSAM (2015a, 2015b)
<i>Pseudomonas</i> spp.	Inoculum size & Temp. abrupt	Transactions of the CSAM (2015c) & J. Food Safety
	Chilled pork in MAP	Transactions of the CSAE (2012) (Chn)
	Time-to-detection	Food Sci. (2013b) (Chn)
	Cardinal Parameters Models	Transactions of the CSAM (2014a)
	Single cell probability	Transactions of the CSAM (2014b)
	RSM of NaL, teapolyphenol and chitosan	Chinese J. Bioprocess Eng. (2014) (Chn)
<i>C. Sporogenes</i>	RSM in media	Food Micro. (2007)
	Weibull heat survival	Int'l Food Process Eng. (2011)
	Combined growth and inactivation	Food Sci. (2010) (Chn)

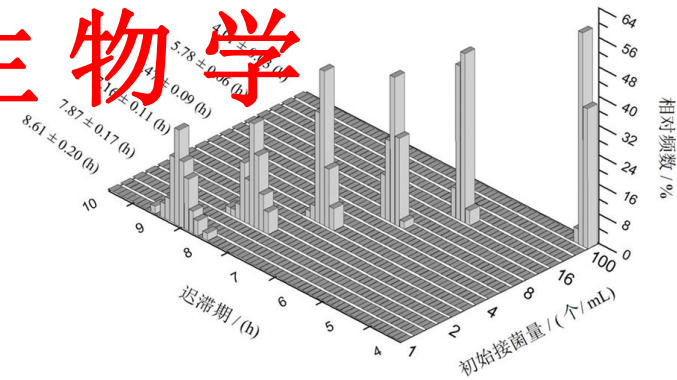
我们团队的在研课题

预测建模领域中的多菌竞争建模

微生物单细胞观测和建模

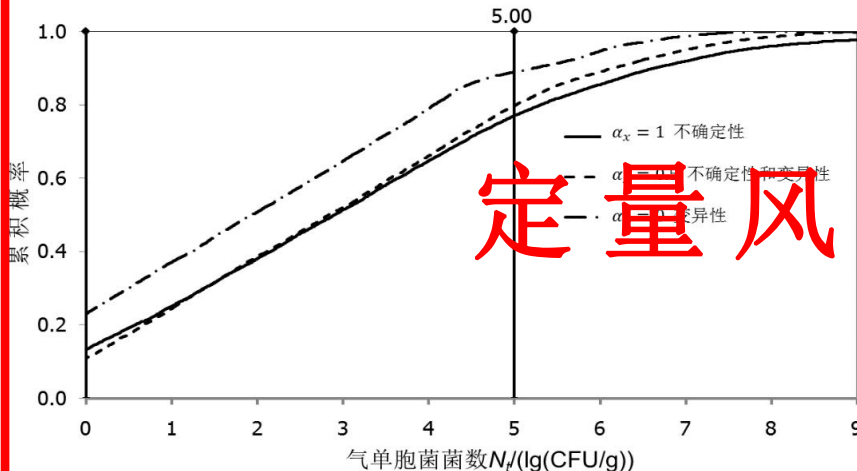


预测微生物学



定量风评中的概率建模和评价体系

全程产业链的交叉污染建模



定量风险评估

$$T_{CM} = \begin{bmatrix} 1-T_1-T_2-T_3 & T_1 & T_2 & T_3 & 0 \\ 0 & 1 & 0 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 0 & 1 \end{bmatrix}$$

场景 1

$$\begin{aligned} T_1 &= T_{MB}'(1-T_{MH}')(1-T_{MK}') \\ T_2 &= T_{MH}'(1-T_{MB}')(1-T_{MK}') \\ T_3 &= T_{MK}'(1-T_{MB}')(1-T_{MH}') \end{aligned}$$

$$b_{M1} = b_0 \cdot T_{CM} = \{b_1, b_2, b_3, b_4, b_5\}$$

$$b_{L1} = b_0 \cdot T_{CM} \cdot T_{CL1}$$

$$T_{CL1} = \begin{bmatrix} 1 & 0 & 0 & 0 & 0 \\ 0 & 1-T_{BL1}' & 0 & 0 & T_{BL1}' \\ 0 & 0 & 1-T_{HL1}' & 0 & T_{HL1}' \\ 0 & 0 & 0 & 1-T_{KL1}' & T_{KL1}' \\ 0 & 0 & 0 & 0 & 1 \end{bmatrix}$$



目 录

- 食品安全风险
- 预测微生物学概念和发展
- 基于预测微生物学的定量风险评估
- 结论与展望



微生物风险评估的研究进展

- 截至2016年，WHO/FAO已连续发表了19部微生物风险评估的系列报告，内容涉及到禽肉、即食食品、水、婴儿配方粉、水产品、蔬菜等不同食品基质中的沙门氏菌、单增李斯特菌、阪崎肠杆菌、弧菌、病毒等，成为指导全球开展相关评估和制定相关标准的权威性技术文件。



World Health
Organization





国内外的研究进展

- 我国在此领域起步较晚，但自2000年以来，先后开展了带壳鸡蛋中沙门氏菌、生食牡蛎中副溶血性弧菌、即食肉制品中单核细胞增生性李斯特菌及婴儿配方粉中阪崎肠杆菌的风险评估。
- 近年来，上海市食药监局组织食品安全风险评估专项，对常见食品中的单增李斯特菌、沙门氏菌、金黄色葡萄球菌、大肠杆菌、蜡样芽胞杆菌等进行了评估。



Status and future of Quantitative Microbiological Risk Assessment in China

Q.L. Dong^{a,b,*}, G.C. Barker^b,
L.G.M. Gorris^c, M.S. Tian^{d,e},
X.Y. Song^f and P.K. Malakar^b

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to initiate QMRA, effectiveness of microbial risk assessment utility for risk management decision making, and application of QMRA to establish appropriate Food Safety Objectives.

Introduction

Several food safety issues that occurred in China in recent years have contributed to a decrease in public confidence concerning the domestic food supply. These incidents include the melamine contamination in milk powder in 2008, clenbuterol hydrochloride contamination of meat products in 2010, widespread usage of tainted cooking oil in Chinese restaurants in 2011, and plasticizer contamination in moon-cakes (traditional food for Mid-Autumn Festival in China) in 2013.

Keywords: Quantitative microbiological risk assessment; Food safety; China



MRA

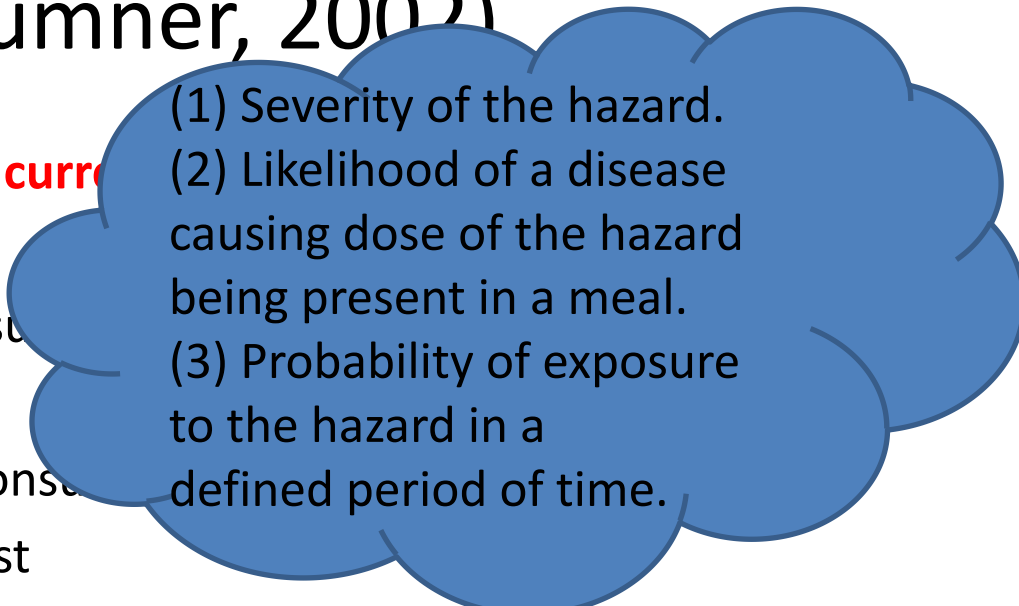
- Microbial risk assessment (MRA) is a process used to evaluate the likelihood of adverse human health effects occurring after exposure to a pathogenic microorganism.
- Qualitative vs. Quantitative?

Excel-in Risk Ranger

(Ross and Sumner, 2002)

Weighting values used in the current

1. Hazard severity
2. How susceptible is the consumer?
3. Frequency of consumption
4. Proportion of population consumed
5. Size of population of interest
6. Proportion of product contaminated?
7. Effect of process
8. Is there a potential for recontamination?
9. How much increase from level at processing is required to reach an infectious or toxic dose for the average consumer?
10. How effective is the post-processing control system?
11. Effect of preparation for meal

- 
- (1) Severity of the hazard.
 - (2) Likelihood of a disease causing dose of the hazard being present in a meal.
 - (3) Probability of exposure to the hazard in a defined period of time.

A. SUSCEPTIBILITY AND SEVERITY

1 Hazard Severity

SEVERE hazard - causes death to most victims
 MODERATE hazard - requires medical intervention in most cases
 MILD hazard - sometimes requires medical attention
 MINOR hazard - patient rarely seeks medical attention

2 How susceptible is the population of interest ?

GENERAL - all members of the population
 SLIGHT - e.g., infants, aged
 VERY - e.g., neonates, very young, diabetes, cancer, alcoholic etc
 EXTREME - e.g., AIDS, transplants recipients, etc.

B. PROBABILITY OF EXPOSURE TO FOOD

3 Frequency of Consumption

daily
 weekly
 monthly
 a few times per year
 once every few years

4 Proportion of Population Consuming the Product

all (100%)
 most (75%)
 some (25%)
 very few (5%)

5 Size of Consuming Population

Australia
 ACT
 New South Wales
 Northern Territory
 Queensland
 South Australia
 Tasmania
 Victoria
 Western Australia
 OTHER

Population considered:

19,500,000

If "OTHER" please specify:

270,000,000

C. PROBABILITY OF FOOD CONTAINING AN INFECTIOUS DOSE

6 Probability of Contamination of Raw Product per Serving

Rare (1 in a 1000)
 Infrequent (1 per cent)
 Sometimes (10 per cent)
 Common (50 per cent)
 All (100 per cent)
 OTHER

If "OTHER" enter a percentage value between 0 (none) and 100 (all)

5.0000%

7 Effect of Processing

The process RELIABLY ELIMINATES hazards
 The process USUALLY (99% of cases) ELIMINATES hazards
 The process SLIGHTLY (50% of cases) REDUCES hazards
 The process has NO EFFECT on the hazards
 The process INCREASES (10 x) the hazards
 The process GREATLY INCREASES (1000 x) the hazards
 OTHER

If "OTHER" enter a value that indicates the extent of risk increase

1.00E-01

8 Is there potential for recontamination after processing ?

NO
 YES - minor (1% frequency)
 YES - major (50% frequency)
 OTHER

If "OTHER" enter a percentage value between 0 (none) and 100 (all)

2.90%

9 How effective is the post-processing control system?

WELL CONTROLLED - reliable, effective, systems in place (no change)
 CONTROLLED - mostly reliable systems in place (3-fold increase)
 NOT CONTROLLED - no systems, untrained staff (10-fold increase)
 GROSS ABUSE OCCURS - (e.g. 1000-fold increase)
 NOT RELEVANT - level of risk agent does not change

10 What increase in the post-processing contamination level would cause infection or intoxication to the average consumer ?

none
 slight (10 fold increase)
 moderate (100-fold increase)
 significant (10,000-fold increase)
 OTHER

If "other", what is the increase (multiplicative) needed to reach an infectious dose ?

7.E+01

11 Effect of preparation before eating

Meal Preparation RELIABLY ELIMINATES hazards
 Meal Preparation USUALLY ELIMINATES (99%) hazards
 Meal Preparation SLIGHTLY REDUCES (50%) hazards
 Meal Preparation has NO EFFECT on the hazards
 OTHER

If "other", enter a value that indicates the extent of risk increase

5.00E-01

RISK ESTIMATES

probability of illness per day per consumer of interest ($P_{inf} \times P_{exp}$) 6.32E-01

total predicted illnesses/annum in population of interest 2.25E+07

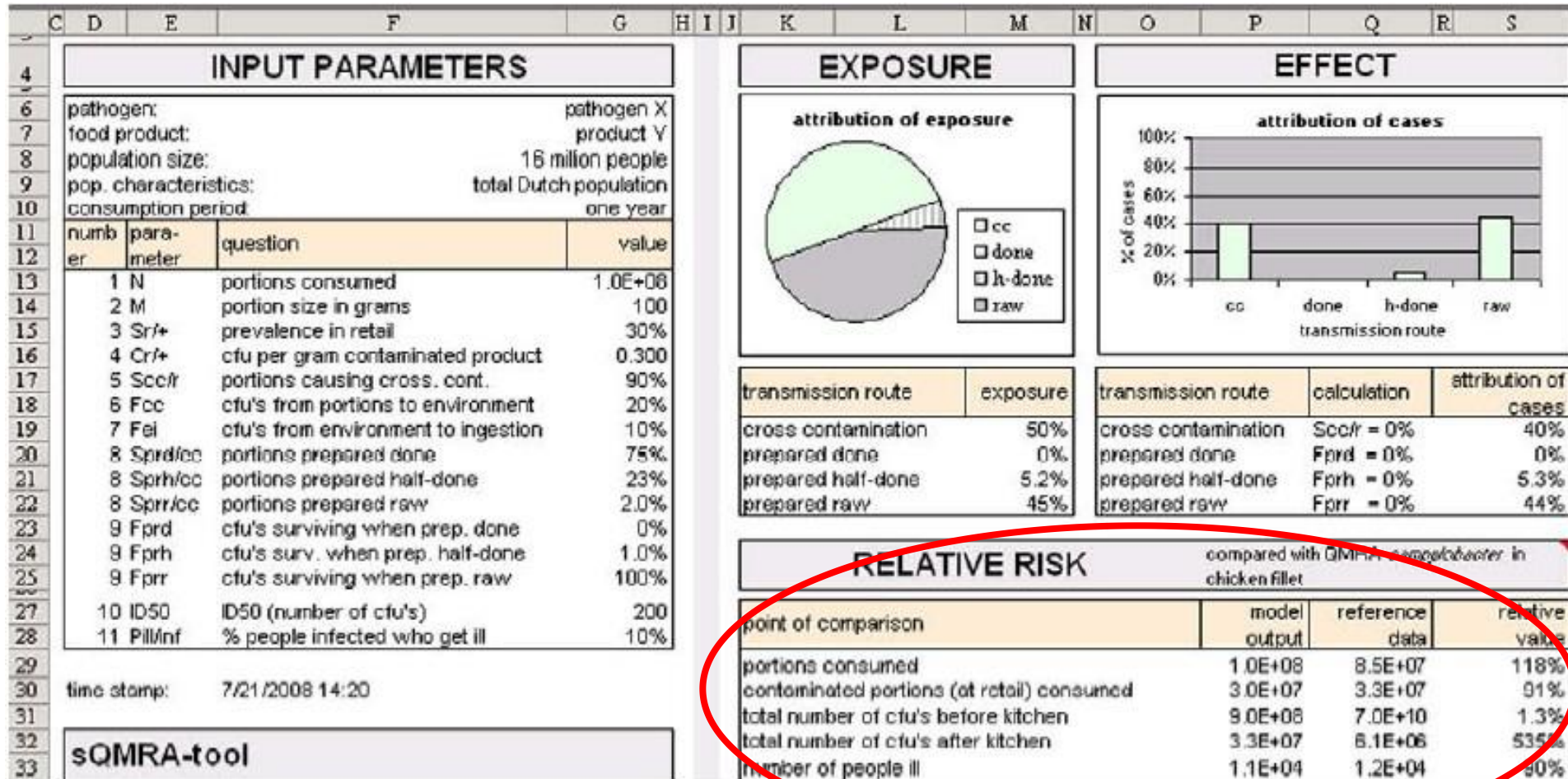
"COMPARATIVE RISK" in population of interest (severity*proportion consuming*prob.illness per consumer per day) 3.16E-01

RISK RANKING
 (0 to 100)

52

A swift Quantitative Microbiological Risk Assessment (sQMRA) tool

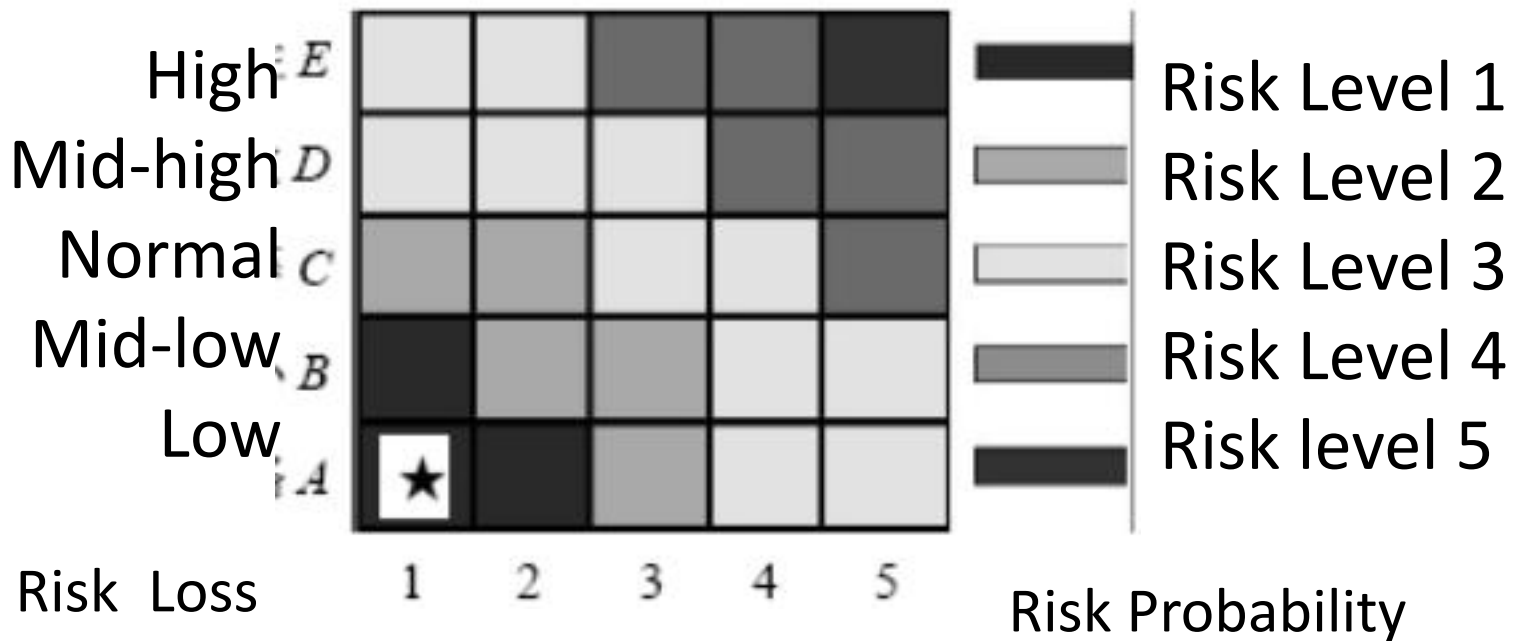
(Evers and Chardon, 2010)





Risk Matrix

Dong et al., (2012)





QMRA tool

食品微生物风险评估工具

简介 初级模型 二级模型 风险评估

本软件是由VBA编写的EXCEL宏程序，通过输入初始菌数，可简单快速的得到初级模型及二级模型的拟合图形及相关参数，并用于风险评估，估计易感人群的患病风险。

注意：使用该程序时，请确保已安装相应插件，否则程序将不能正常启动。

食品微生物风险评估工具

简介 初级模型 二级模型 风险评估

Time Log CFU

粘贴 删除

计算 (Gompertz) 计算 (Logistic)

复制结果 复制结果

Gompertz					
N_0	C	U	Lag	R^2	
Logistic					
N_0	C	U	Lag	R^2	

食品微生物风险评估工具

简介 初级模型 二级模型 风险评估

Square Root
Modified SR
Ratkowsky
Modified RK
Polynomial

2 factors
3 factors

Factor Observed

删除

食品微生物风险评估工具

简介 初级模型 二级模型 风险评估

No	U	Lag	t	r	计算	N_t	<input type="text"/>
N_t					计算	p	<input type="text"/>
删除							



致病菌定量风险评估示例

- 某市冷却猪肉中气单胞菌的风险评估（2011）
- 某市米饭中蜡样芽胞杆菌的风险评估（2012）
- 全国即食食品中单增的李斯特菌（2014）



米饭中蜡样芽胞杆菌的风险评估

- 剂量-反应模型（dose–response model, DRM）是用数学表达式来表示剂量和反应的关系，主要描述摄入的致病菌数量和最终发生疾病的数量关系。
- 指数模型 $P = 1 - \exp(-r * N)$
- Beta-Poisson模型 $P = 1 - (1 + N / \beta)^{-\alpha}$
- Weibull模型 $P = -(N / \beta)^{\alpha}$



米饭中蜡样芽孢杆菌的风险评估

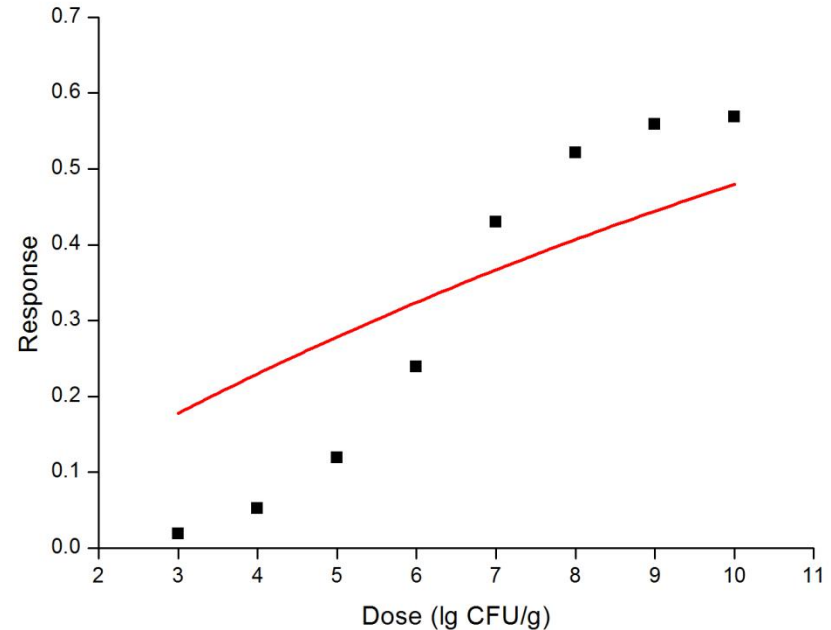
- 米饭的蜡样芽孢杆菌的**摄入剂量** (N) 通过Kramer和Gilbert在1989年对1971~1985年间107起英国米饭中蜡样芽孢杆菌导致食物中毒的分析数据。含量为 $1.0 \times 10^3 \sim 5.0 \times 10^{10}$ CFU/g, 中值约为 1×10^7 CFU/g。
- **发病概率** (P) 的数据计算公式: $P = P_E \times A_R \times S_E$
- 其中 P_E 为特定含量的蜡样芽孢杆菌导致的呕吐发生率, 通过摄入人群中的疾病爆发率求得。 A_R 为致病菌的攻击率, 为独立于剂量的变量, 流行病学统计表明此变量符合正态分布, 均值为**63.7%**。 S_E 为致呕吐型蜡样芽孢杆菌的比例, 根据Gilbert和Parry (1977)、Shinagawa等 (2001) 的研究, 此比例取值为**75%**。



米饭中蜡样芽胞杆菌的风险评估

$$P = 1 - \exp(-0.065N)$$

- 只有指数模型非线性收敛（ $R^2=0.66, p<0.05$ ），Beta-Poisson模型和Weibull模型均不理想。



- 指数模型是一种典型的非阈限模型，这意味着没有“最低感染剂量”。指数型剂量反应模型在模拟严重的致病菌上具有公认的适用性，以及当外推到低剂量影响范围时的线性特点，在Bahk等（2007）开展的韩国传统食品紫菜包饭（Kimbab）中蜡状芽孢杆菌定量风险评估也曾成功应用过。



米饭中蜡样芽孢杆菌的风险评估

- **Beta** ($s+1, n-s+1$), 其中 n 指样品总数, s 代表阳性样品数.
- **Cumulative** ($min, max, \{x_1, x_2, \dots, x_n\}, \{p_1, p_2, \dots, p_n\}$), 其中 min 和 max 分别为阳性样品检测量 x_1, x_2, \dots, x_n 的最小值和最大值, p_1, p_2, \dots, p_n 为各检测量的累积概率。
- 阴性样品中污染水平参考 $M = -(2.303 / V) \times \log(Z / N)$
- 式中 M 表示样品中的真实浓度, V 表示检测时所用样品量, Z 表示阴性样品的数量, N 为检测样品的总数。
- **Normal** ($mean, std-dev$), 其中销售时间的平均值 $mean$ 和标准差 $std-dev$.
- **Pert** ($min, most, max$), 其中销售温度的最小值 min 、最可能值 $most$ 和最大值 max .
- 预测微生物模型分别选用修正的Gompertz模型和Ratkowski平方根模型, 数据参考McElroy等在1999年监测中式米饭中蜡样芽孢杆菌和2000年监测煮米饭中蜡样芽孢杆菌孢子, 以及Heo等2009年监测蒸煮米饭中蜡样芽孢杆菌的生长数据。

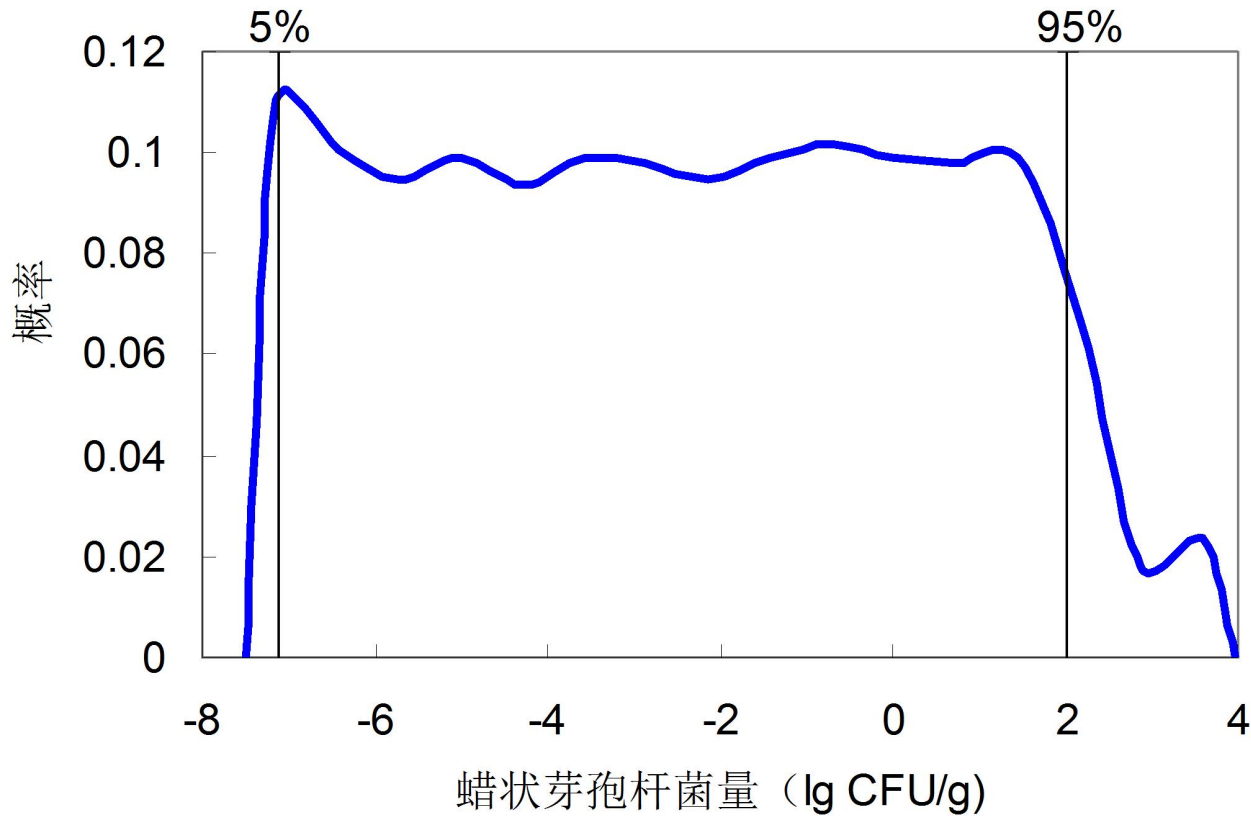


米饭中蜡样芽孢杆菌的风险评估

- 构建暴露评估模型后，以蜡样芽孢杆菌预测模型的最终菌数为输出变量，选用美国Palisade公司@Risk 4.5软件中的Monte Carlo取样方法运行迭代10000次。参照Notermans和Batt于1998年完成的食源性蜡样芽孢杆菌风险评估报告，以蜡样芽孢杆菌导致食物中毒的经验值4 log CFU/g为风险阈值。



米饭中蜡样芽胞杆菌的风险评估



米饭中蜡样芽孢杆菌的初始污染水平



米饭中蜡样芽胞杆菌的风险评估

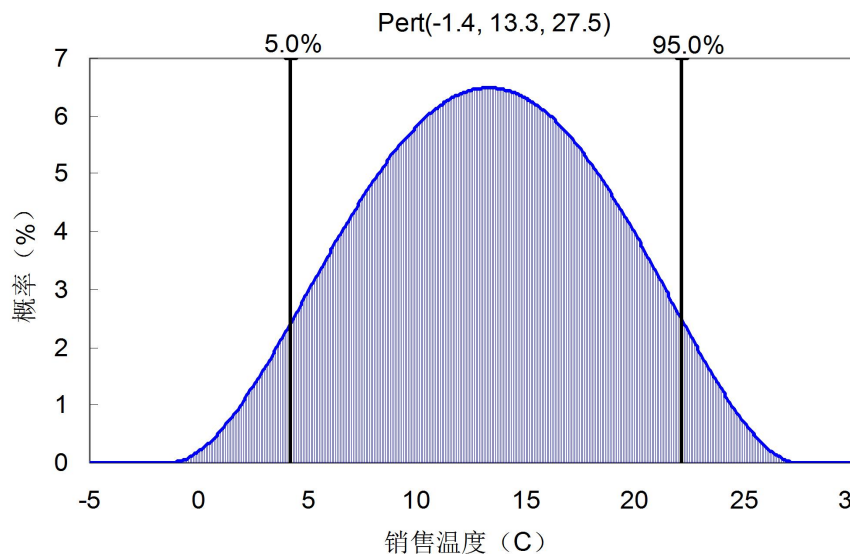
米饭中蜡样芽孢杆菌的初始污染概率分布数据

	阳性样品	阴性样品	初始污染率	初始污染水平
最小值 (lg CFU/g)	2.00	-7.51	0	-7.51
最大值 (lg CFU/g)	4.00	2.00	1	3.96
中值 (lg CFU/g)	2.60	-2.76	0.05	-
平均值 (lg CFU/g)	2.81	-2.76	0.06	-2.46
标准差	0.60	2.80	0.03	3.00
5%置信区间	2.06	-7.12	0.02	-7.11
95%置信区间	3.87	1.61	0.11	2.01

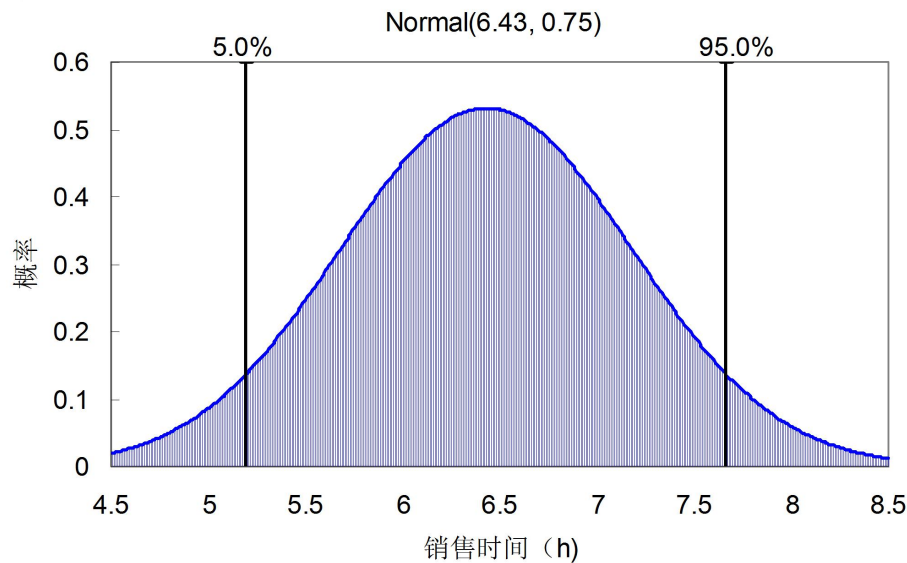
米饭销售时气单胞菌量最低为-7.51 lg CFU/g，最高为3.96 lg CFU/g，平均值为-2.46 lg CFU/g。



暴露评估-仿真运行



米饭销售至食用前的温度概率分布

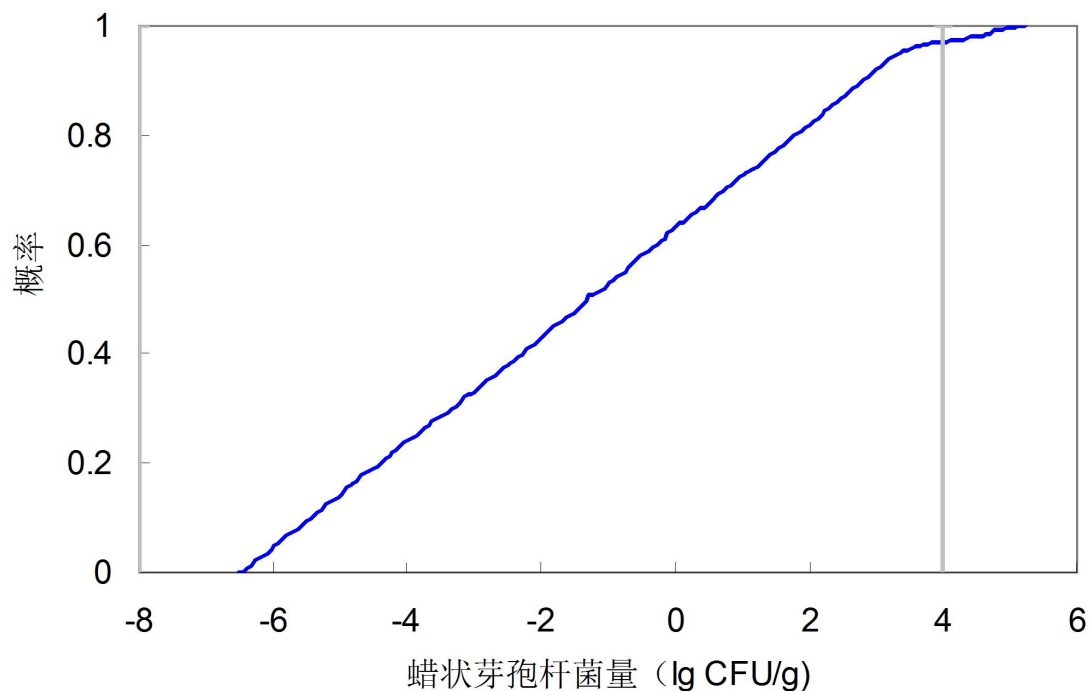


米饭销售至食用前的时间概率分布

销售温度和时间分别为4.20°C（5%置信区间）至22.16°C（95%置信区间），
5.20 h（5%置信区间）至7.66 h（95%置信区间）。



米饭中蜡样芽胞杆菌的风险评估

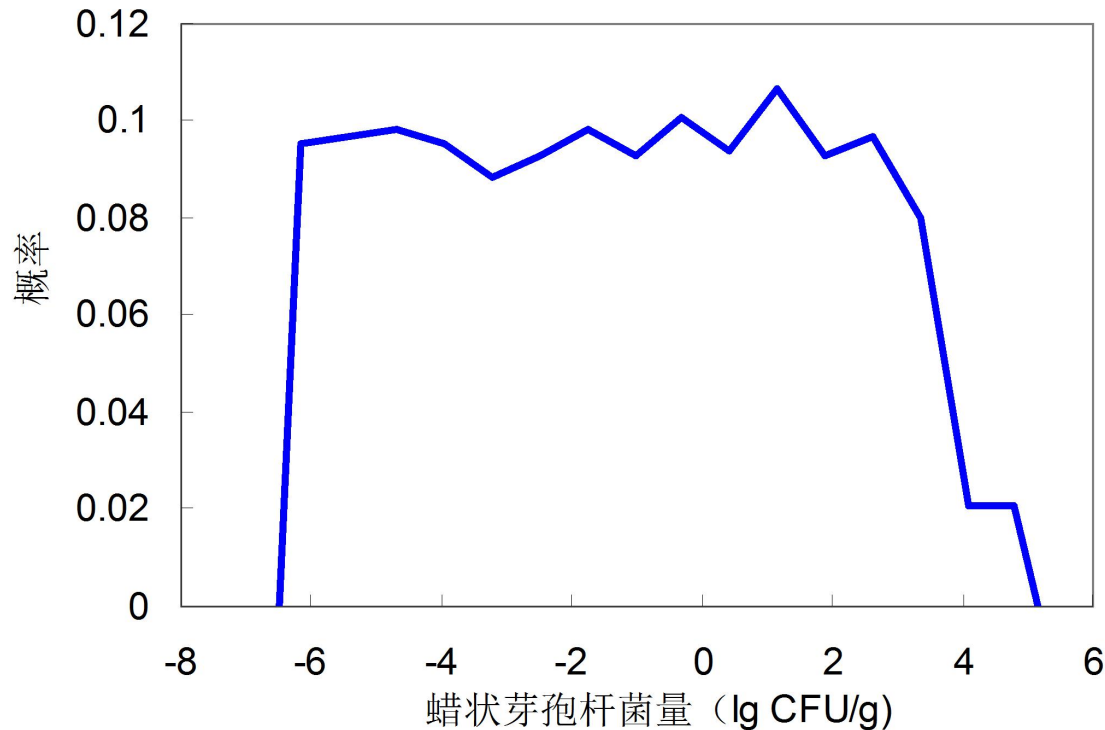


米饭销售至食用阶段蜡样芽孢杆菌的概率分布

米饭在销售阶段增长超过风险阈值（4 lg CFU/g）的概率较低（3.07%）



米饭中蜡样芽胞杆菌的风险评估



米饭中蜡样芽孢杆菌的食用前最终污染水平

最终污染范围从-6.51 lg CFU/g（5%置信水平）至5.17 lg CFU/g（95%置信水平），均值为-1.26 lg CFU/g。



米饭中蜡样芽胞杆菌的风险评估

- 通过小样本调研确定1100人样本量进行每日米饭摄入量膳食调查，基于某市统计局 2011年5月公布的某市第六次全国人口普查主要数据，某市实际膳食调查样本构成和调查结果如下：

	年龄0-14	年龄组15-64	年龄组65-	合计
性别 男	48	470	48	566
性别 女	43	439	52	534
合计	91	909	100	1100

	年龄组 0-14	年龄组 15-64	年龄组 65-	合计
性别 男	155.00 ± 110.53 Aa	369.91 ± 102.62 Ba	328.10 ± 95.45 Ca	348.14 ± 118.80 a
性别 女	94.77 ± 72.43 Ab	254.44 ± 92.67 Ba	261.02 ± 97.69 Ba	242.22 ± 101.47 b
合计	126.54 ± 98.70 A	314.15 ± 113.65 B	293.22 ± 101.86 B	296.72 ± 122.69

数据表示形式为平均值±标准差，同行上标不同大写字母者差异显著 ($p < 0.05$)，同列上标不同小写字母者差异显著 ($p < 0.05$)



米饭中蜡样芽孢杆菌的风险评估

- 某市居民平均每日米饭类产品的总摄入量均值为**296.72 g**，则根据风险阈值（ $4 \lg \text{CFU/g}$ ）求得某市居民食用米饭中蜡样芽孢杆菌的风险量为 **$2.97 \times 10^6 \text{CFU}$** ；
- 根据暴露评估中米饭中蜡样芽孢杆菌的食用前最终污染水平为 **$-6.51 \lg \text{CFU/g}$** （5%置信水平）至 **$5.17 \lg \text{CFU/g}$** （95%置信水平），均值为 **$-1.26 \lg \text{CFU/g}$** ；求得某市居民平均每日米饭中的蜡样芽孢杆菌量为 **$9.17 \times 10^{-5} \sim 4.29 \times 10^7 \text{CFU}$** ，均值为 **$16.31 \text{CFU}$** ，可将此作为某市居民每日的蜡样芽孢杆菌耐受摄入量（TI），即因摄入米饭不可避免的摄入蜡样芽孢杆菌的允许量，超过风险阈值的概率仍为**3.07%**。
- 代入剂量反应方程，推测某市居民由于摄入米饭中蜡样芽孢杆菌的发病率（ P ）为 **-5.48%** （5%置信水平）至 **28.5%** （95%置信水平）。



米饭中蜡样芽孢杆菌的风险评估

米饭中蜡样芽孢杆菌暴露评估的敏感性分析

	相关系数	回归系数	等级
销售温度	-0.015	-0.004	1
初始污染水平	0.009	0	2
销售时间	-0.004	0	3

敏感性指的是哪些数据输入对最终结果影响最大，取决于数据中的不确定性，以获得降低风险的有效控制措施。米饭中蜡样芽孢杆菌的食用前最终污染水平与与销售温度绝对值最高（-0.015），其次是初始销售水平和销售时间，采用必要的冷藏措施降低米饭销售时的贮藏温度有助于减少最终污染的风险。



展望

- 预测微生物的基础体系日渐完善，英美等国构建的ComBase全球数据库免费共享。
- 预测微生物学从理论到实践任重而道远。
- 预测微生物学在微生物定量风险评估领域应用潜力深远。
- 国内的相关研究急需整合和完善。





MicroRisk workshop

MicroRisk-2014 Beijing



MicroRisk-2014 Wuhan



MicroRisk-2015 Shanghai



MicroRisk-2016 Shanghai



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<http://www.microrisk.cn>

谢谢请指正！

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