International Association for Food Protection®
WEBINAR

Practical Applications of Microbial Modelling Webinar Series

May 22, 2018 10:00 a.m. CDT

Practical Applications of Microbial Modelling Webinar Series

Webinar Series:

Part III of III

- This IAFP webinar is sponsored by the following Professional Development Groups:
  - Microbial Modelling and Risk Analysis
  - Meat and Poultry Safety and Quality

International Association for Food Protection® WEBINAR

3

Practical Applications of Microbial Modelling

Webinar Series: Part III of III

Dr. Bala Kottapalli, moderator

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WEBINAR HOUSEKEEPING

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WEBINAR HOUSEKEEPING

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Agenda

- Introduction
  - Dr. Bala Kottapalli
- *Salmonella* – Sprouts Risk Assessment, with a general overview
  - Dr. Yuhuan Chen
- Interactive Panel Discussion
  - Dr. Betsy Booren
  - Dr. Tom Ross
  - Dr. Peter Taormina
  - Dr. Marcel Zwietering
- Audience Questions and Answers

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U.S. FOOD & DRUG ADMINISTRATION

An Overview of Risk Assessment

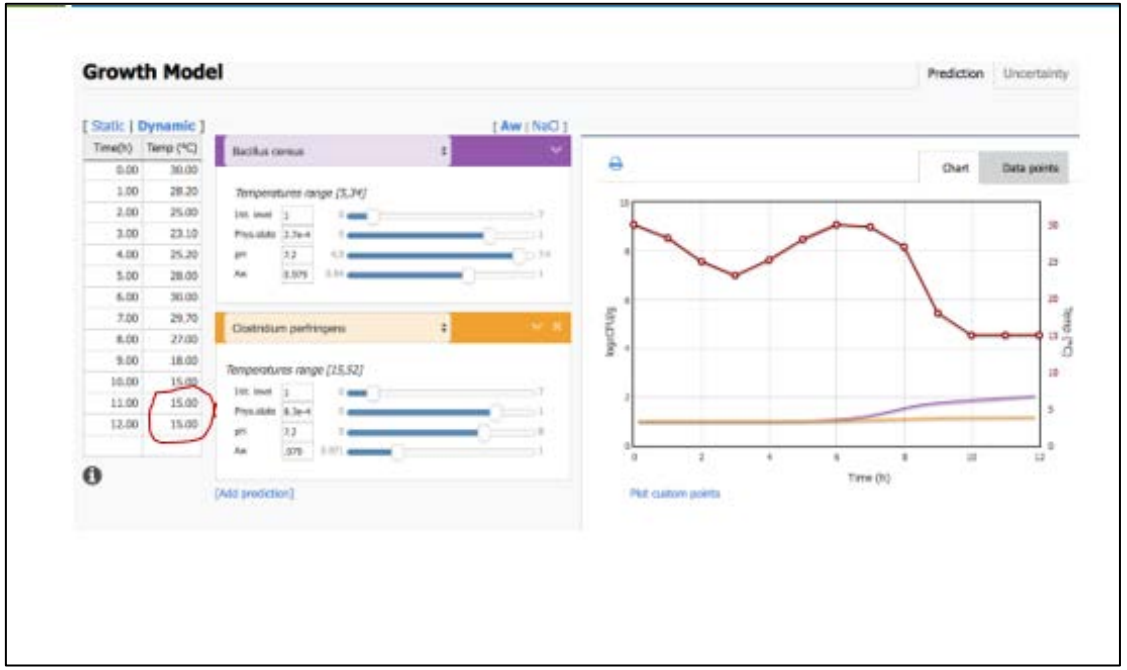
What comes before and after predictive modeling of growth and inactivation?

Dr. Yuhuan Chen, FDA CFSAN

Before I start…

The information and conclusions presented in this webinar do not necessarily represent Agency policy nor do they imply an imminent change in existing policy.

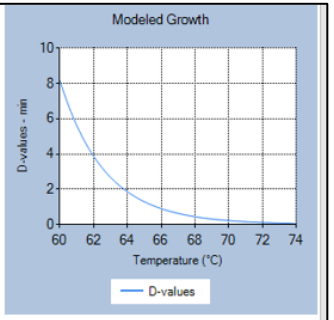
Review: Webinar Parts I and II



- Used Heat Inactivation Model
  - *L. monocytogenes* – ground beef
    - No sodium lactate or sodium diacetate

MODELED INACTIVATION

Temperature (°C)	D-value (min)
73.7	0.06
73.8	0.06
73.9	0.06



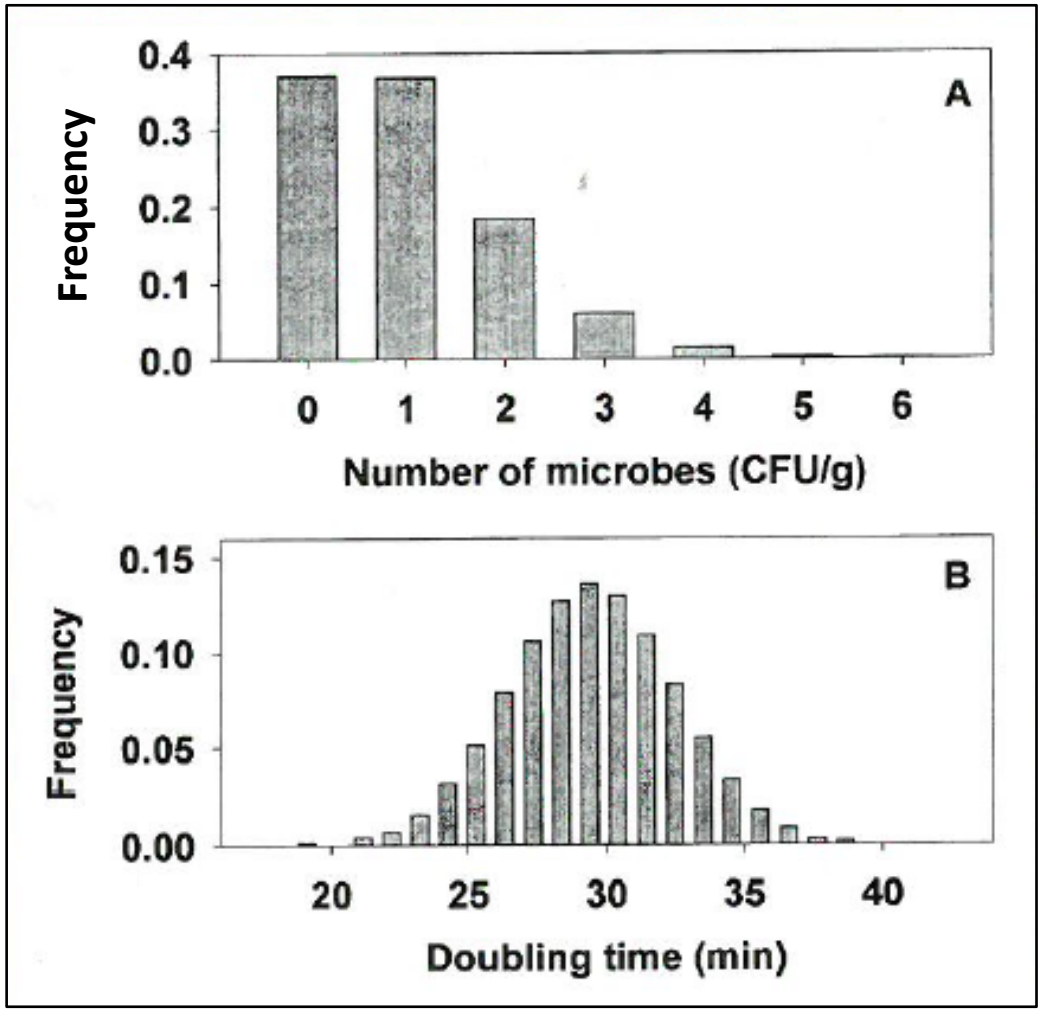
  - Assume 5-log reduction is adequate for safety, approximately 0.3 minutes are needed to achieve a 5 log reduction.
    - Actual product temperature at geometric center is 74°C for 1.5 minutes.

Predicted results from growth modeling and inactivation modeling (discussed in Webinars I&II) together with knowledge of pathogen initial level & level of concern, and other factors, inform determination of food safety risk.

Biological and Process Variability

- Webinar part II showed variability in thermal resistance among *L. monocytogenes* strains (Aryani et al., 2015)
- "The average" does not adequately capture, as examples:
  - the behavior of pathogen in food, e.g., growth
  - the effect of the pathogen reduction process
  - the initial levels of pathogen

Variability Matters: an Example



A) Poisson distribution for the initial level of pathogen

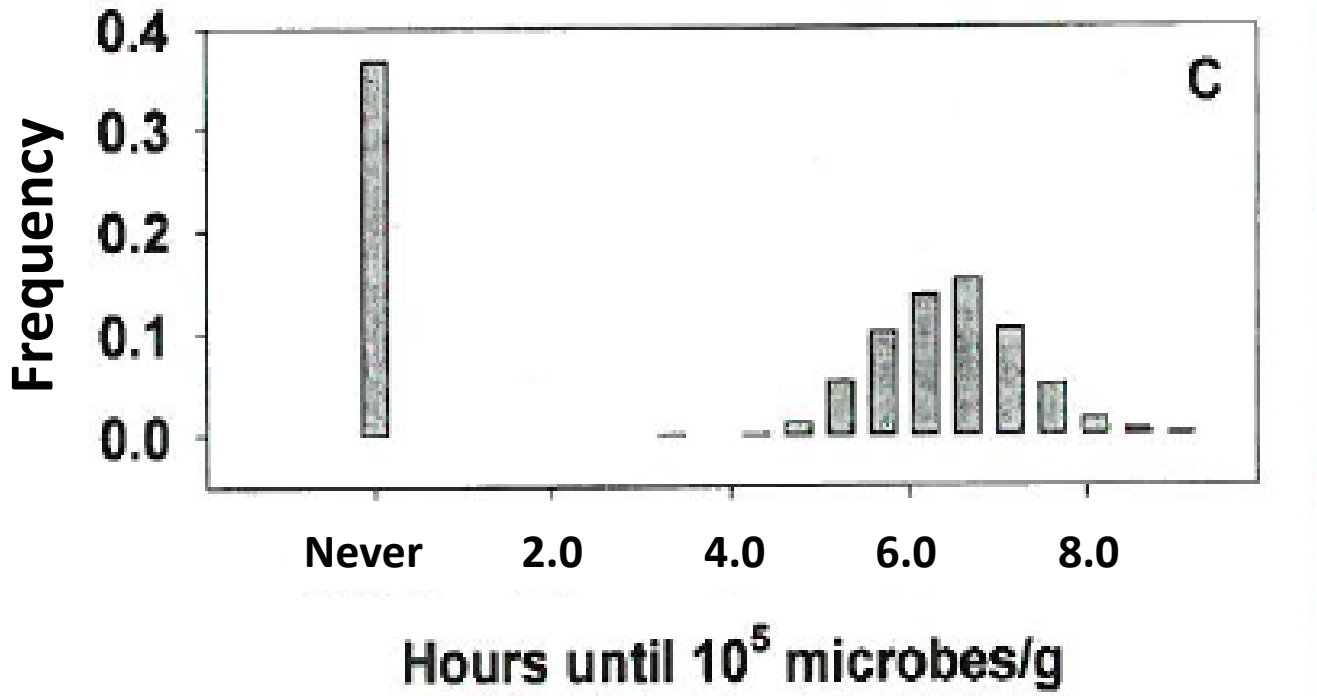
B) Normal distribution of doubling time

Assumption: level of concern 5 log CFU/g

Variability incorporated into exposure assessment through Monte Carlo simulation

(Schaffner and Chen, 2001)

Variability Matters: an Example (cont.)



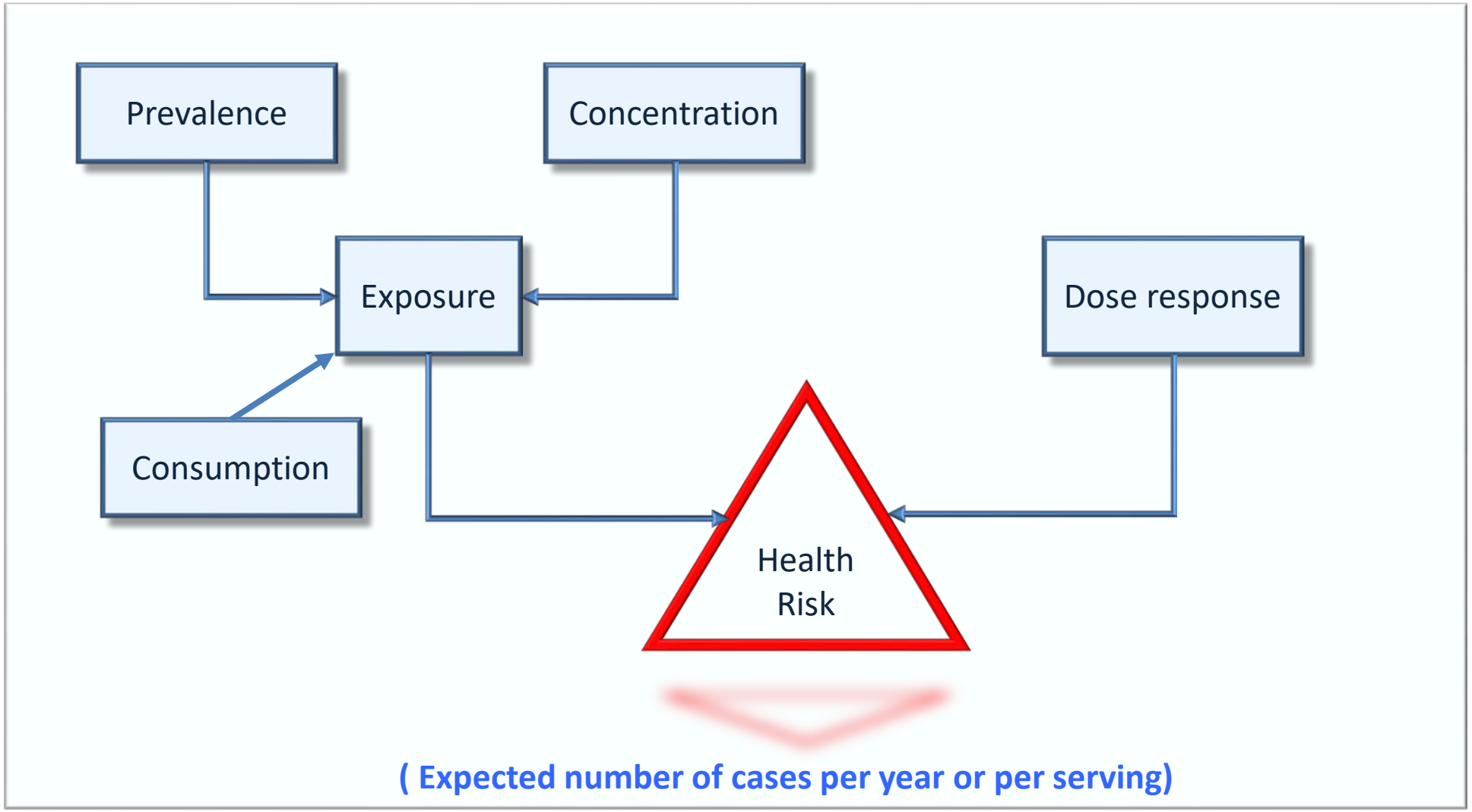
- A certain number of samples never reach 5 log CFU/g
- The time required to reach 5 log CFU/g varies (for positive samples)
  - average ~ 6.5 h
  - as little as 3.0 h
  - as long as 9.0 h
- Important to consider the variability in decision, e.g., for storage time, for in-process hold time.

(Schaffner and Chen, 2001)

What comes before and after predictive modeling of growth and inactivation?

- Before: initial prevalence and level, etc.
- Predictive modeling
  - growth
  - inactivation
  - cross-contamination
  - Other aspects of microbial behavior in foods
- After: connect contamination in food to other components of a risk assessment

Risk Assessment: Estimating Risk of Illness to Consumers

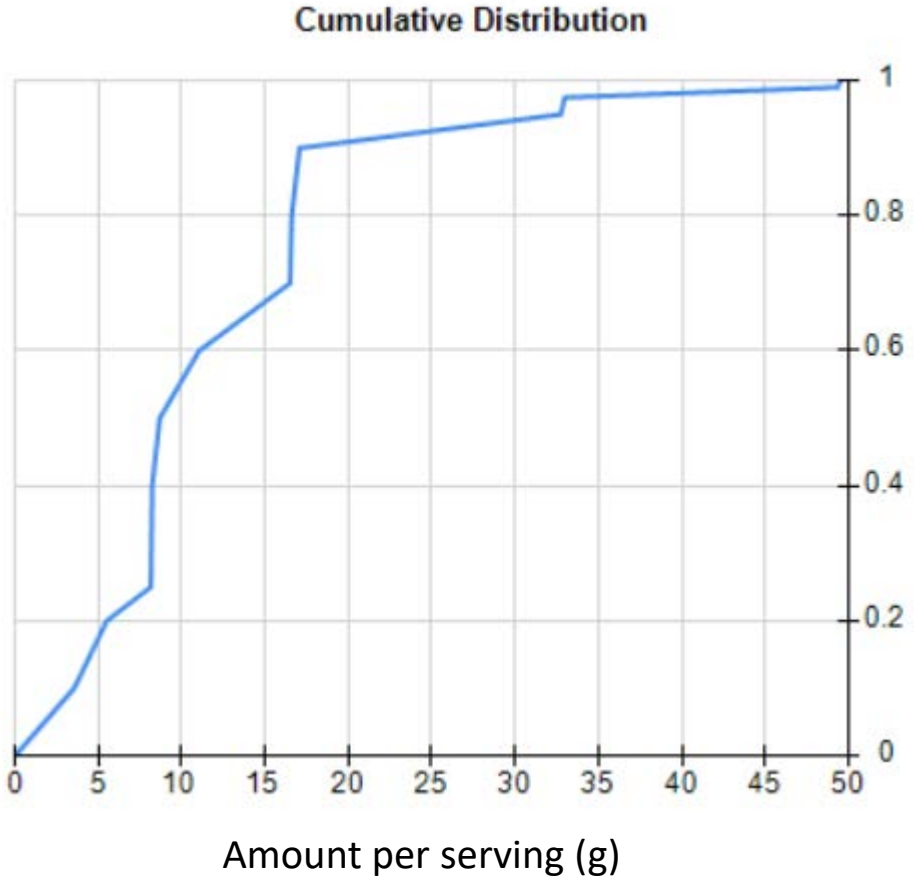


Consumption Example: Alfalfa Sprouts

- Eating occasions (servings) per year in the U.S. : 8.52×10^7 (85.2 million)
- Amount consumed per serving: variable

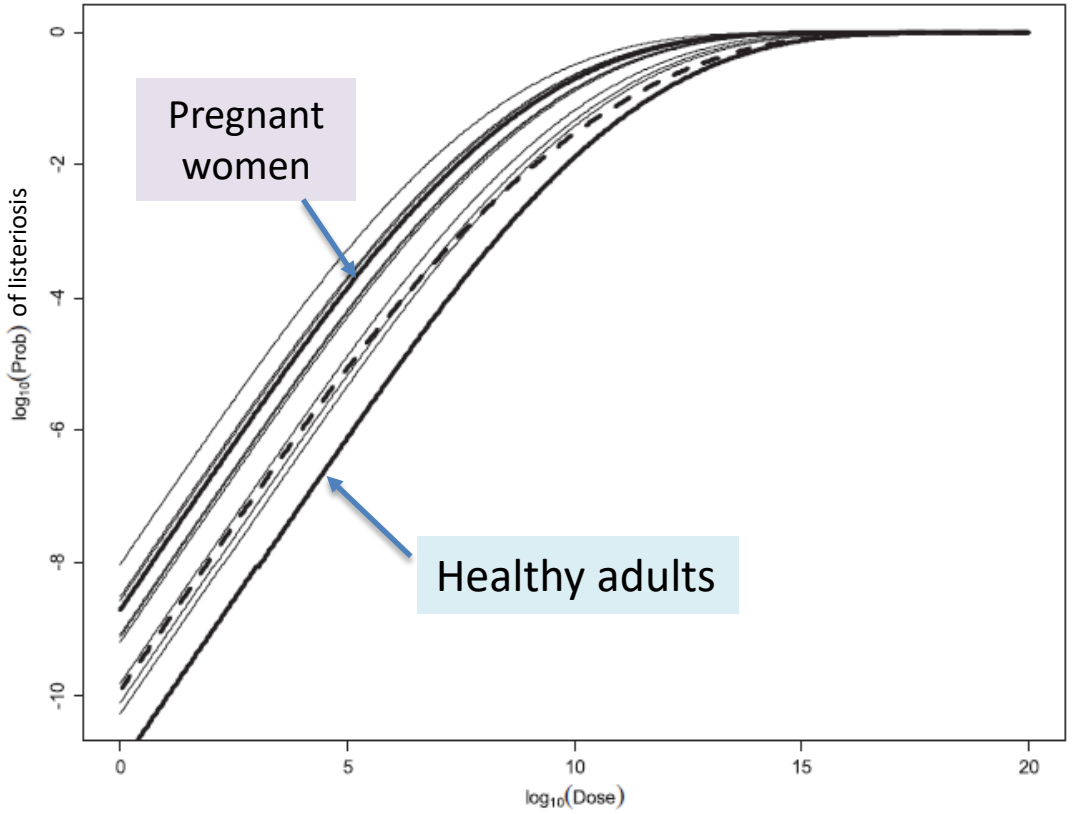
Source:

- NHANES What We Eat in America database



Dose-Response Relationship: Example 1

L. monocytogenes Dose-Response Variability

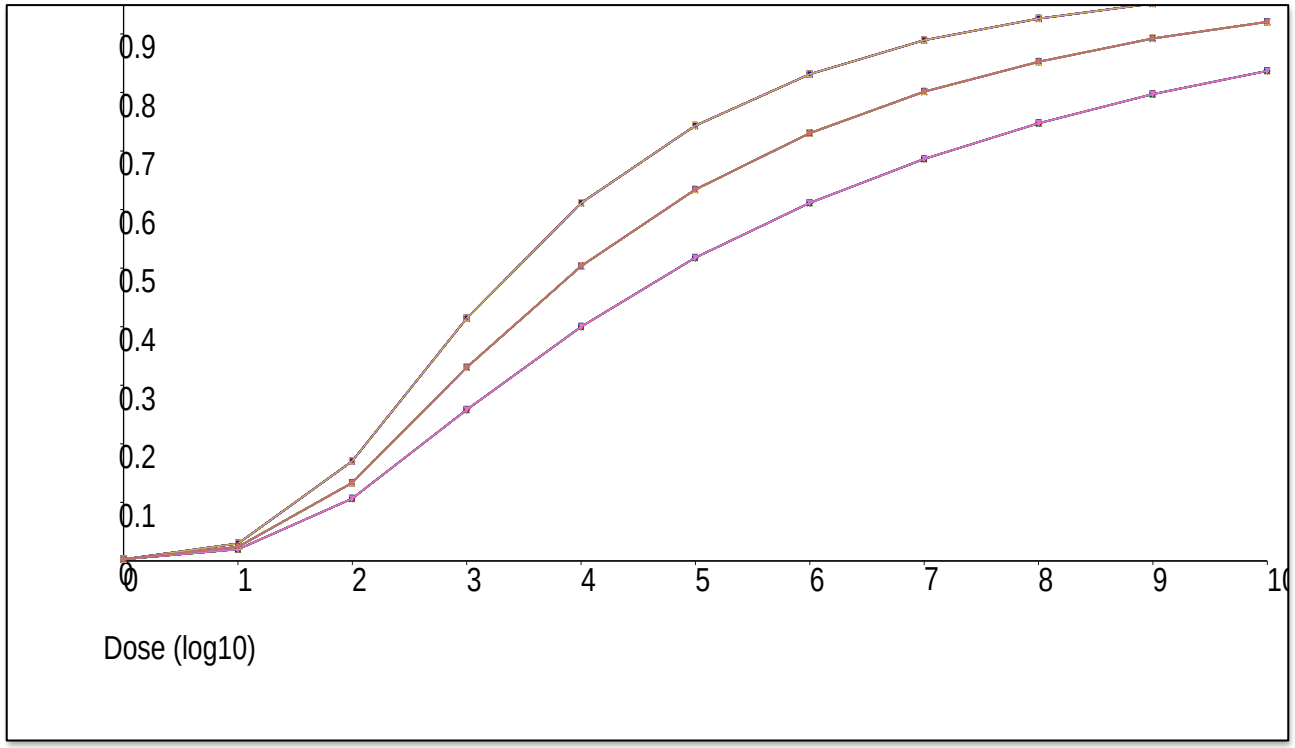


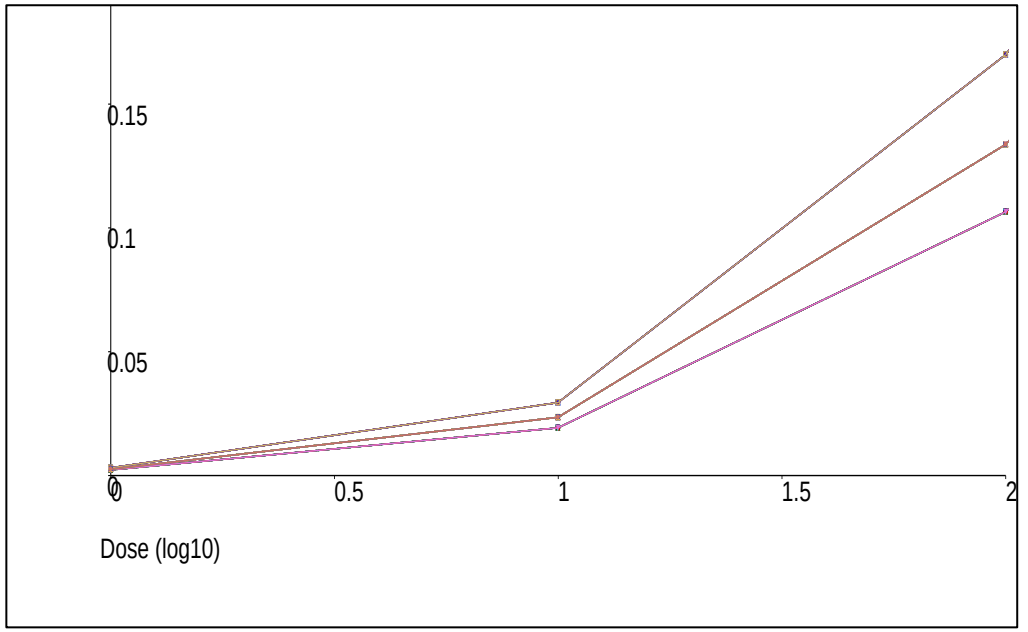
Lognormal-Poisson models for U.S. total population and sub-populations:

- 11 subgroups (solid lines)
- Total population (dashed line)

(Pouillot et al., 2015)

Dose-Response Relationship: Example 2



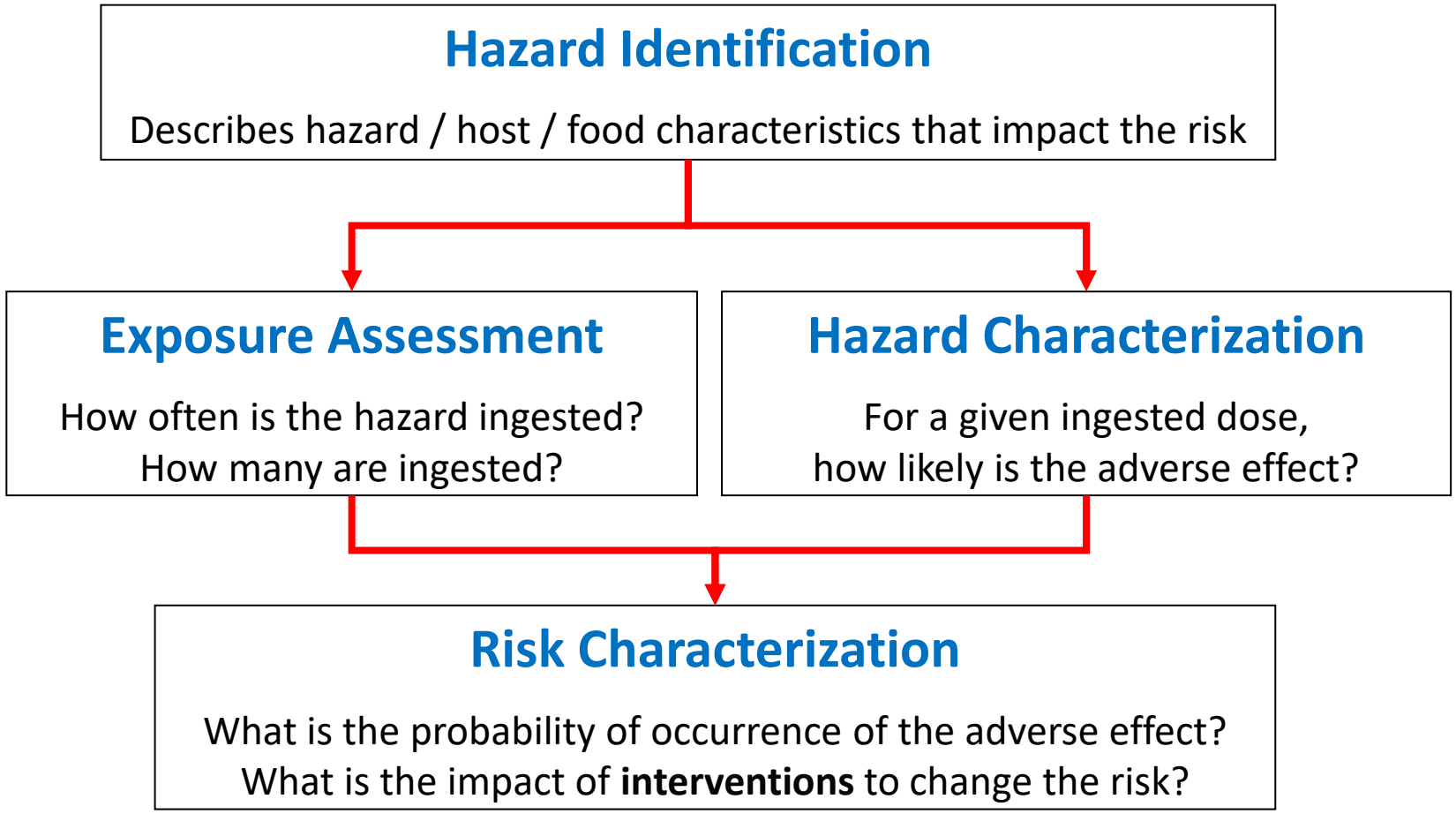


Salmonella dose response

median (middle curve) and 95% confidence interval (uncertainty, lower/upper curves)

(model parameters from WHO/FAO, 2002)

Risk Assessment Paradigm



(Codex working principles, 2007)

Salmonella – Sprouts Risk Assessment

Risk Analysis
AN INTERNATIONAL JOURNAL
An Official Publication of the Society for Risk Analysis

Original Research Article | Open Access

Risk Assessment of Salmonellosis from Consumption of Alfalfa Sprouts and Evaluation of the Public Health Impact of Sprout Seed Treatment and Spent Irrigation Water Testing

Yuhuan Chen, Régis Pouillot, Sofia M. Santillana Farakos, Steven Duret, Judith Spungen, Tong-Jen Fu, Fazila Shakir, Patricia A. Homola, Sherri Dennis, Jane M. Van Doren, ... See fewer authors

First published: 16 January 2018 | https://doi.org/10.1111/risa.12964

Salmonella – Sprouts Risk Assessment Policy Context

- Informs development of guidance to industry
  - Guidance provides recommendations to assist operations covered by Subpart M in complying with the requirements in the Produce Safety Rule
  - Draft Guidance announced in Federal Register Notice 01/23/17
  - FR Notice indicated developing a risk assessment model to evaluate the public health impact of seed treatment and testing of spent irrigation water in a sprout production system, and FDA's intention to make it available following peer review

Risk Assessment Charge

Evaluate risk of human salmonellosis associated with alfalfa sprouts consumption and the public health impact of different log pathogen reduction levels for treating seeds intended for sprouting, alone or in combination with spent irrigation water testing

Typical Sprout Production Process

Seed Receipt →Seed Storage
→Initial Seed Rinse →Seed Treatment
→Pre-germination Seed Soak →Germination and Growth
→Microbial testing of SIW (or in-process sprouts)
→Harvest →Wash/Drain Sprouts →Bulk Cool/Spin Dry
→ Pack and/or Package →Cooling & Storage →Distribution

(FDA draft guidance 2017,
Adapted from NACMCF 1999)

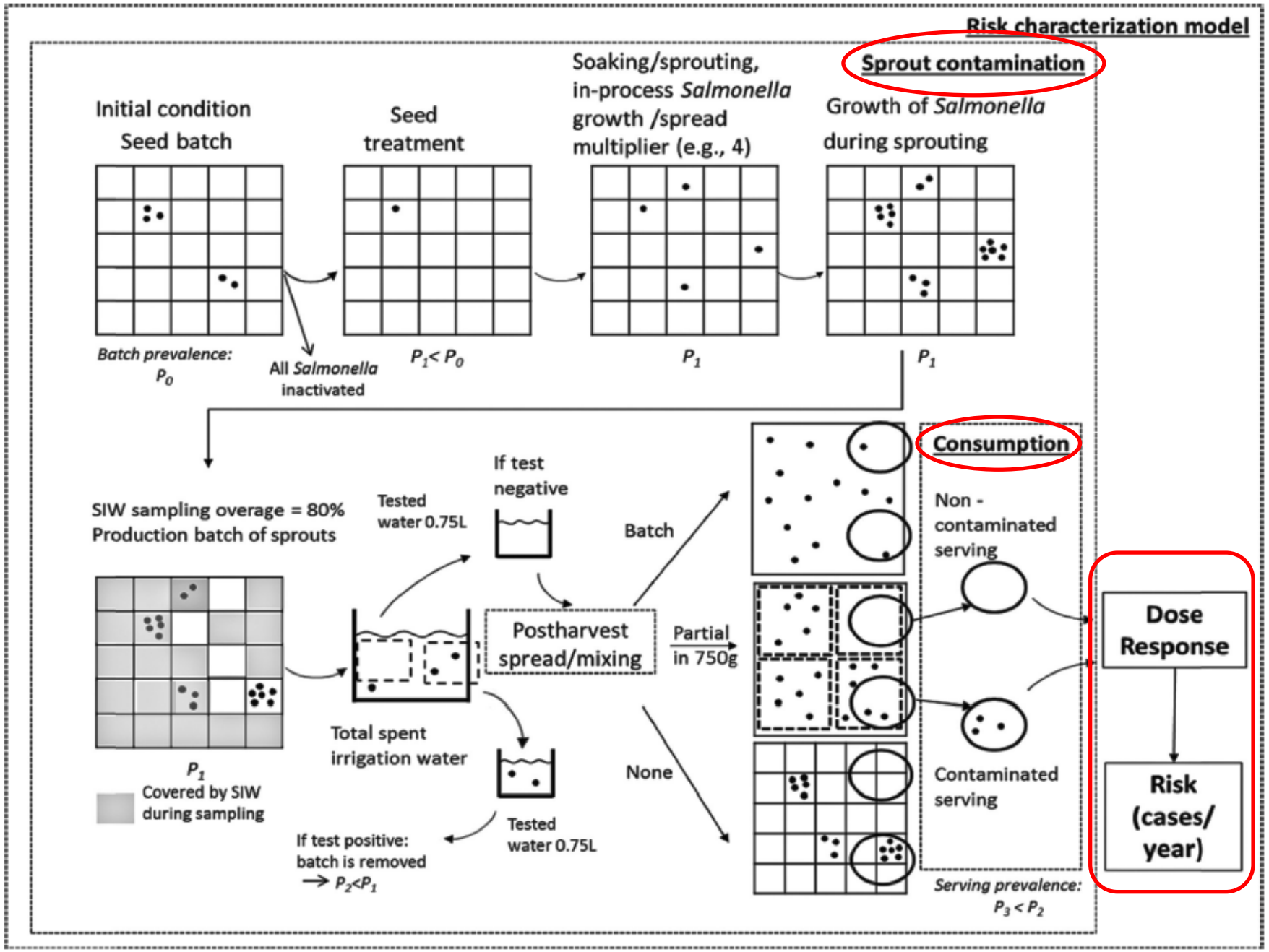
Public Health Concerns

- Outbreaks of foodborne illness attributed to the consumption of sprouts reported in the U.S. and worldwide, for example:
  - Worldwide: 15 outbreaks in eight countries between 1973-1998 (Taormina et al., 1999)
  - U.S.: 46 outbreaks, accounting for 2,474 cases, attributed to sprouts between 1996 and 2016 (Gensheimer and Gubernot, 2016)

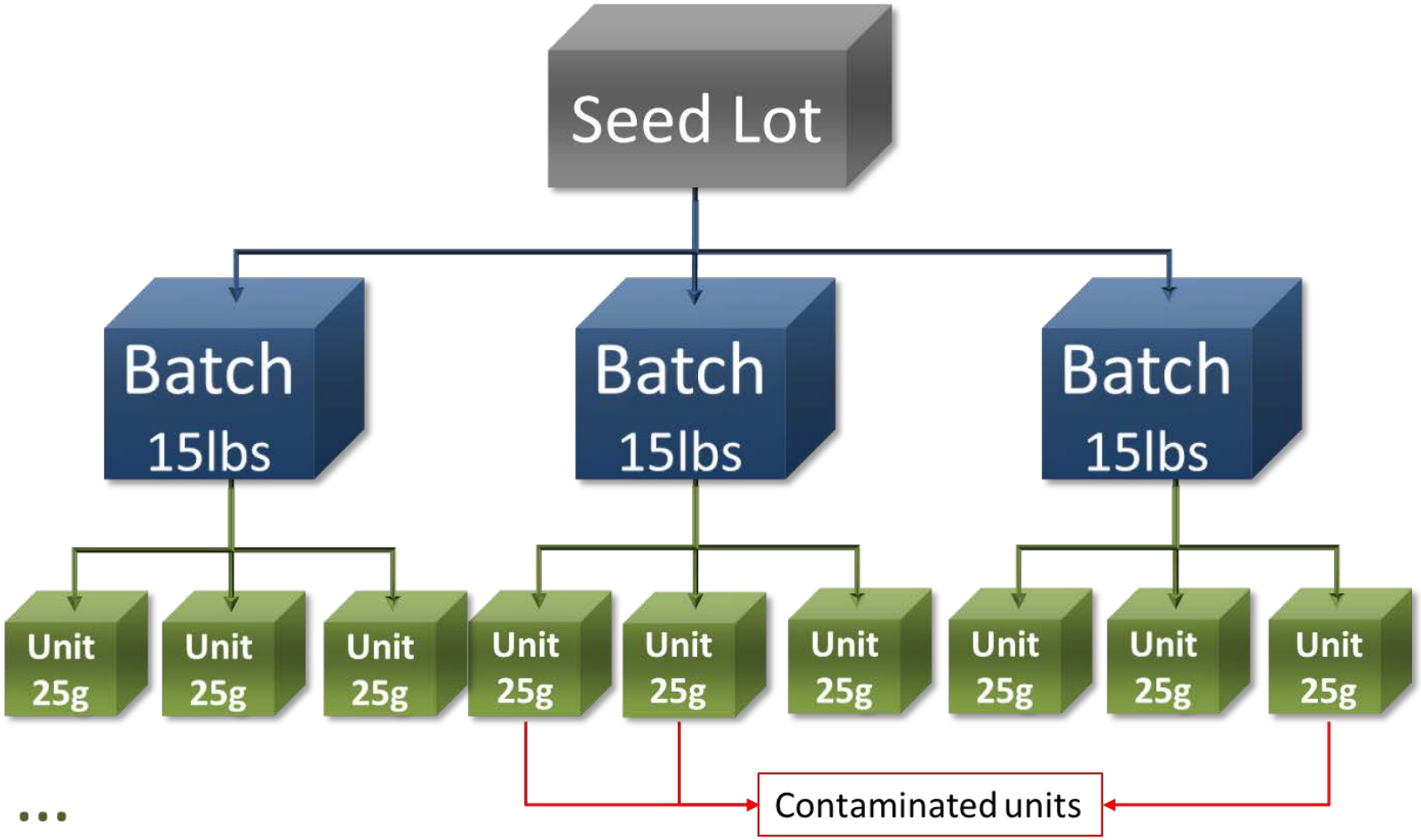
Public Health Concerns

- Sprouts produced under conditions that favor pathogen growth
- Sprouts are often consumed raw
- Outbreaks identified were diverse - associated with many different sprout varieties and attributed to a variety of pathogens
- *Salmonella* was the most common pathogen reported for sprout-associated outbreaks; the majority of the outbreaks were attributed to alfalfa sprouts.

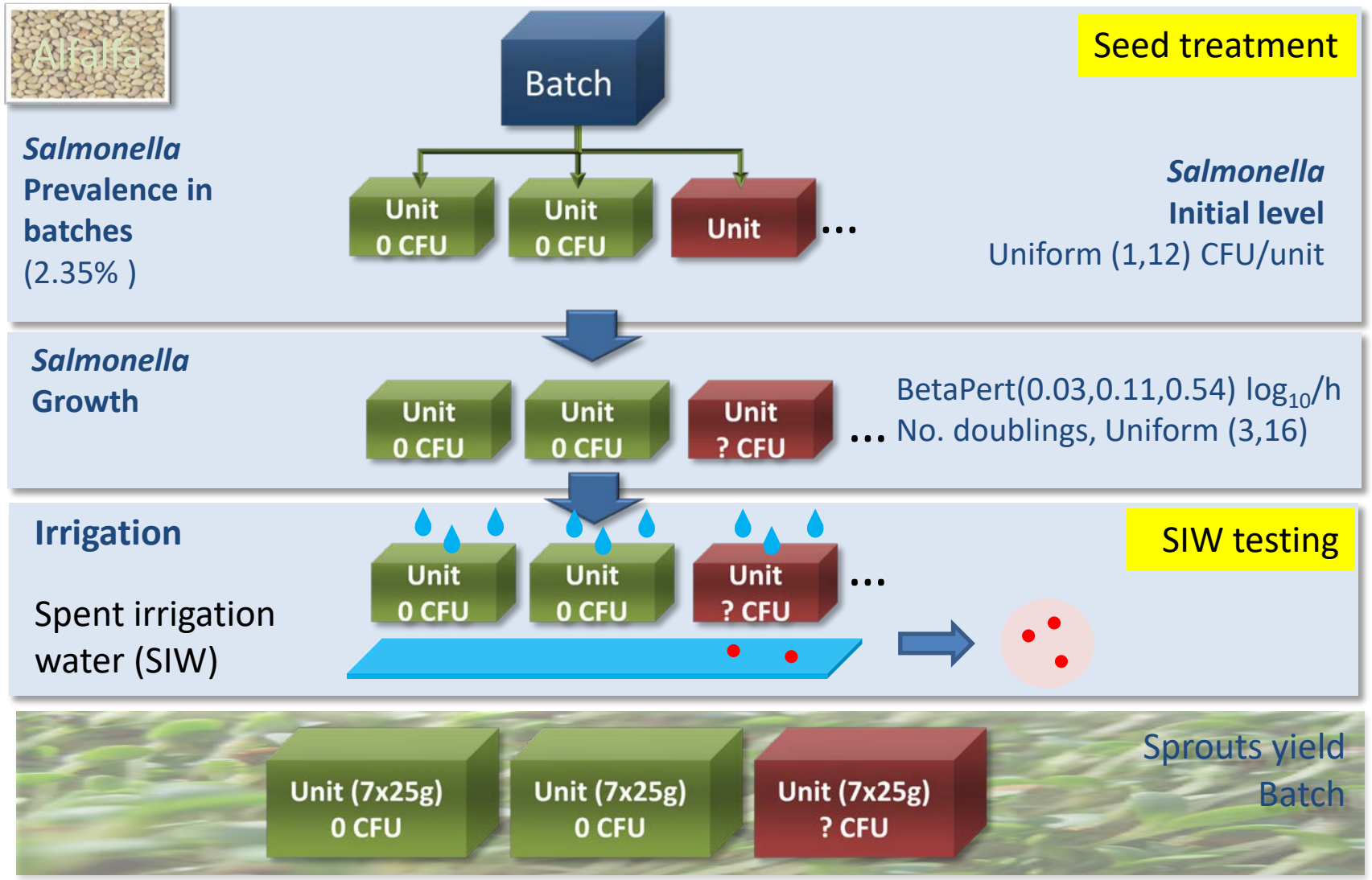
Components of the *Salmonella*-Alfalfa Sprouts Risk Assessment



Definitions: Size of Seed Batch and Seed Units



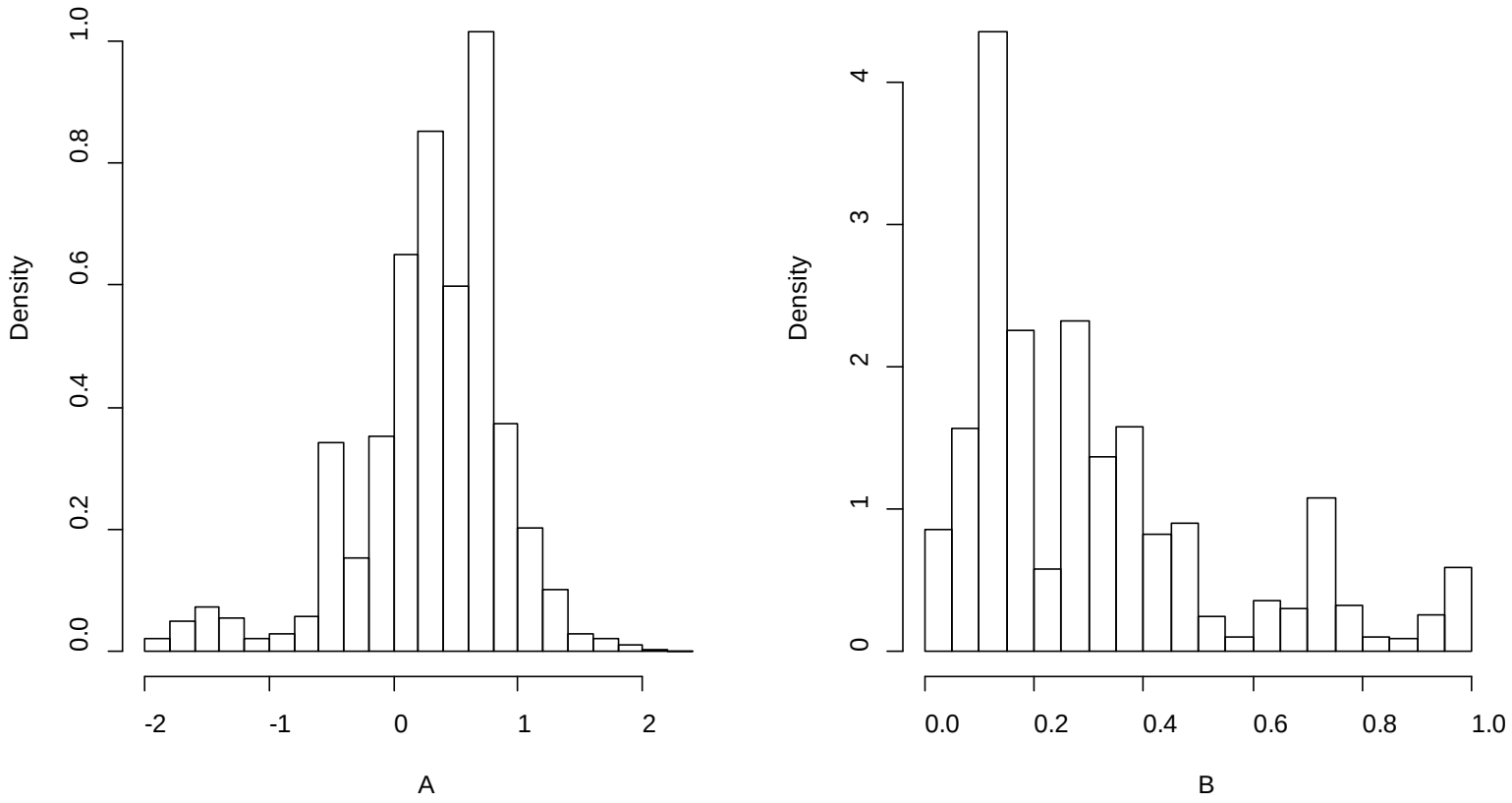
Process Model: *Salmonella* Dynamics during Sprout Production



(Cross-contamination)

(Adapted and expanded on process model by Montville and Schaffner 2005)

Model Inputs Example: Pathogen Transfer Distributions



A: differences in pathogen concentrations (log_{10} CFU/g) between in-process sprouts and SIW;
B: proportions of cells transferred from the sprouts to the SIW (spent irrigation water)

(Data extracted from literature; approach adapted from Montville and Schaffner, 2005)

Model Mathematical Notations and Equations

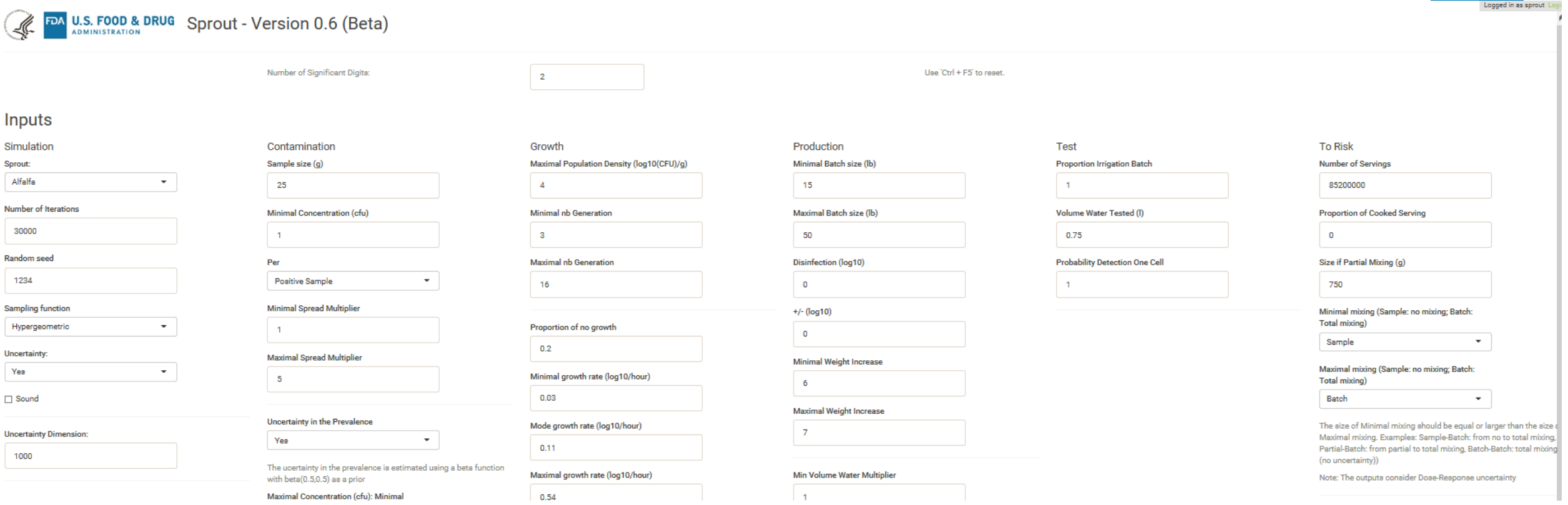
Exposure Module		
Initial conditions for seeds		
Initial unit size	M_u (g)	25 g
Batch size	M_b (g)	Uniform(6,810, 22,700) g, i.e., Uniform(15, 50) lb
Units per batches		
Units per 6.8 kg batches		
Initial batch prevalence		
Prevalence of contaminated units		
Initial number of positive units per		
Initial levels in positive unit		
Total number of cells in positive b		
Treatment		
Log_{10} reduction from seed treatme		
Probability of survival		
Number of cells in positive batch p		
Number of cells in positive units		
Number of positive units per conta disinfection		
Prevalence of contaminated batche		
Multiplier		
In-process pathogen spread multip		
Number of positive units		
Number of cells in posit		
Growth		
Pathogen growth: grow		
Maximum growth: number of gene		
Growth at time t		
Maximum population density		
Time of SIW sampling		
Duration of cell growth during pro		
Number of cells in each unit at the		
Number of cells in each unit at the		

Irrigation			
Coverage of production batch by irrigation water during sampling		δ	Choice: 0, 20, 40, 60, 80, or 100 (%)
Number of positive units touched by spent irrigation water		N_3	~ HypergeometricPos(N_2, $N_u - N_2$, $[N_u \times \delta]$)
Probability to have a positive unit touched by spent irrigation water		ρ_1	$P_1 \times (1 - \frac{(N_u-N_2)!(N_u-[N_u\times\delta])!}{N_u!(N_u-N_2)-[N_u\times\delta])!})$ (The fraction corresponds to the probability to observe 0 success in the hypergeometric process)
Corresponding number of cells in sampled in-process sprouts		C_s (CFU)	Sum of $C_{3,u}$ cells (at the time of SIW sampling) present in N_3 positive units randomly sampled among the N_2 positive units
Yield of sprouts		W	Uniform(6, 7) times
Ratio of volume of water to seeds/in-process sprouts per irrigation cycle		V_m	[volume Uniform(1, 5):1][b]
Differences in pat and sprouts			
Proportion of cell			
SIW testing			
Number of cells in			
Probability to hav			
Volume water use			
Volume of SIW te			
Number of cells in			
Probability to hav			
Probability to det			
Batch kept follow			
Prevalence post-te			
Size of a batch of			
Size of a unit of s			

Postharvest			
Extent of mixing/pathogen spread		M_p	[Ranging from no spread, to partial spread, to complete spread from one or more contaminated units [M_u] to the entire production batch (M_b)][b]
Size of partial mixing of sprouts		M_{ps}	$M_p \times W$
Number of contaminated units in the partial mixing		N_4	~HyperGeometricPos(N_2, N_u- N_2, $[M_p/M_u]$)
Corresponding number of cells in partial mix		C_p	Sum of cells $C_{4,u}$ (end of production) present in N_4 positive units randomly sampled among the N_2 positive units
Probability to have a contaminated partial mix		ρ_6	$P_2 \times (1 - \frac{(N_u-N_2)!(N_u-[M_p/M_u])!}{N_u!(N_u-N_2)-[M_p/M_u])!})$
Risk Prediction			
Parameter of the beta-Poisson dose–response model		α, β	Median estimates evaluated using $\alpha = 0.1{,}324$, $\beta = 51.45$, [lower CI bound (2.5th) evaluated using $\alpha = 0.094$, $\beta = 43.75$; upper CI bound (97.5th) evaluated using $\alpha = 0.1{,}817$, $\beta = 56.35$][b]
Risk per contaminated serving		R	1 – BetaFunction($\alpha, \beta + C_s$) / BetaFunction(α, β)
Eating occasions per year		N_s	8.52E+07
Expected number of cases			$N_s \times$ mean($P_3 \times R$)

The risk assessment considers separately variability and uncertainty in model inputs and predicts the risk of illness as well as uncertainty in the risk estimate

Web-based Model User Interface



Results

Batch Prevalence before water sampling (%)

mean median 1pct 99pct

Proportion of detected batches (%)

mean median 1pct 99pct

Proportion of contaminated batches that are detected (%)

mean median 1pct 99pct

Expected Number of Cases

	Cases
mean	13669.01
median	12128.80
Lower CI	2399.03
Upper CI	41265.59

Predicted Impact of Seed Treatment

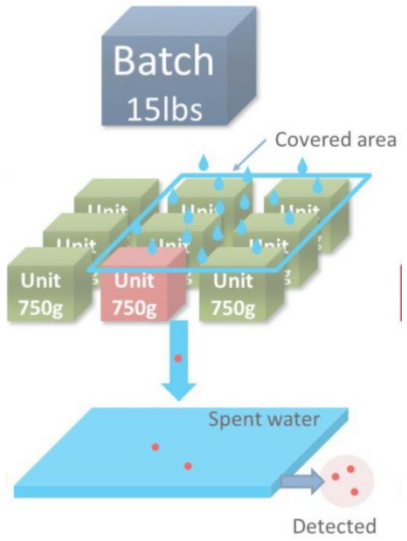
Predicted reduction in contaminated production batches, and reduction in risk to consumers

Scenario, seed treatment	% batches contaminated	Predicted cases/yr	% reduction in cases/yr
No treatment	5.2 [1.8, 12.0]*	76,600 [15,400, 248,000]	
1-log reduction	2.3 [0.81, 5.5]	12,100 [2,900, 39,300]	84 [80, 85]
3-log reduction	0.032 [0.011, 0.077]	139 [33, 448]	99.8 [99.76, 99.83]

* Confidence Interval (uncertainty in the risk estimate)

Predicted Impact of Spent Irrigation Water (SIW) Testing

Predicted reduction in contamination of sprout production batches



Test volume: 0.75L

Scenario, irrigation coverage	% batches contaminated	% reduction in batches contaminated	Predicted cases/yr
0 (no testing)	5.2 [1.8, 12.0]		76,600 [15,400, 248,000]
0.2	3.0 [1.1, 7.0]	42 [37, 44]	
0.4	1.9 [0.66, 4.5]	64 [54, 65]	
0.6	1.4 [0.45, 3.2]	75 [64, 77]	
0.8	1.0 [0.33, 2.7]	82 [69, 83]	
1	0.8 [0.26, 2.3]	86 [72, 87]	12,100 [2,400, 41,200]

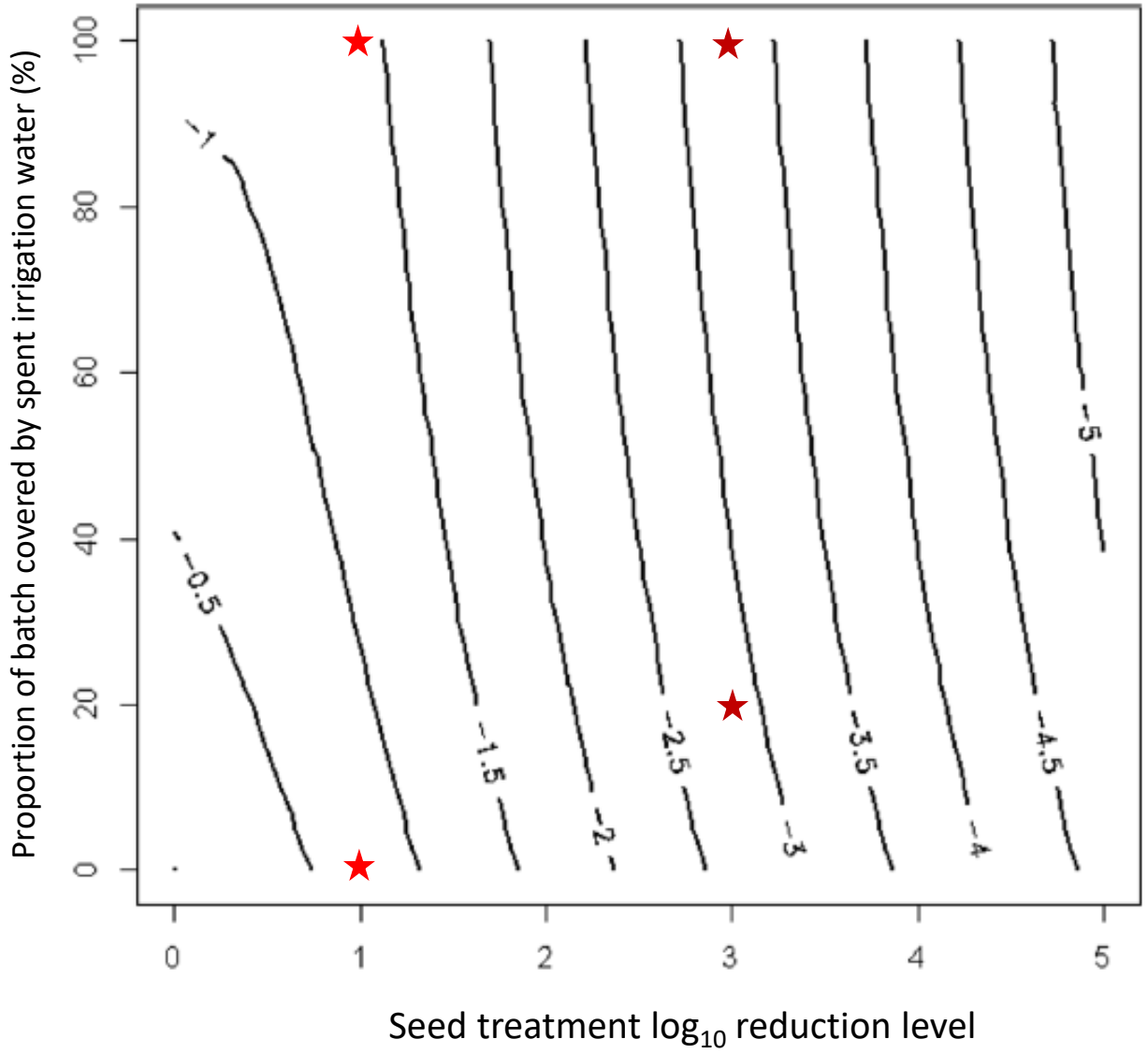
In SIW testing, how you take samples is important. Representative sampling is critical

Predicted Impact of Interventions: Combined Seed Treatment and SIW Testing

Scenario, seed treatment	% batches contaminated	Predicted cases/yr	% reduction in cases	Log_{10} change in cases
No treatment	5.2 [1.8, 12.0]	76,600 [15,400, 248,000]		
1-log reduction + SIW test	0.69 [0.22, 1.7]	3,560 [821, 11,400]	96 [93, 96]	-1.4 [-1.2, -1.4]
3-log reduction + SIW test	0.01 0 [0.0033, 0.026]	45 [10, 146]	--	-3.2 [-3.1, -3.3]
5-log reduction + SIW test	0.00010 [0.000033, 0.00026]	0.45 [0.10, 1.5]	--	–5.3 [–5.1, –5.3]

Predicted Impact of Interventions: Combined Seed Treatment and SIW Testing

Contour plot, $\log_{10}$ reduction in predicted cases/year



Concluding Thoughts

- Risk assessment provides a framework within which to
  - represent sprout production, and integrate a multitude of data and information on a large number of factors to predict effectiveness of control measures
  - understand the impact of seed treatment and SIW testing on reducing a microorganism of public health significance
  - quantify the impact of variability and uncertainty in the outcomes of the risk assessment
- Web-based user interface can be useful to make a complex model more accessible
  - provides a means to evaluate assumptions and alternative scenarios, and to engage SMEs and risk managers during and after model development

Acknowledgements

- Régis Pouillot, Sofia Santillana Farakos, Steven Duret, Judith Spungen, Tong-Jen Fu, Fazila Shakir, Patricia Homola, Joy Johanson, Sherri Dennis and Jane Van Doren

Panel Discussion

Summary from Webinar I

- Various Tertiary Models Exist
  - Some of which were demonstrated in this webinar series
- Select Model Based Upon Your Unique Situation and Parameters
- Be Careful with Assumptions and Interpretation
  - Read and follow guidelines and disclaimers
- Validate and Verify

Summary from Webinar II

- Predictive Modeling is a valuable tool for the food industry to use.
  - It can be used in a variety of situations to access food safety risk.
  - It is important to understand the limitations of predictive modeling to make the best food safety assessment.

Panel Discussion: Question 1

- When can we use predictive modeling as part of validation?
- How can we use predictive modeling as part of validation?
  - Hazard analysis
  - Design of critical limits
  - Corrective actions
  - Reassessment of HACCP and/or Food Safety Plan
  - Other aspects

Panel Discussion: Question 2

- How much variability is there between the responses of the strains of the same bacteria, e.g., growth rate, or death rate?

Panel Discussion: Question 3

- Given the various sources of variability and uncertainty in modelling, how confident can we be in the model predictions and how do we incorporate that into decisions?

Panel Discussion: Question 4

- When can stakeholders engage in the risk assessment process?

Panel Discussion: Question 5

- How much do we know about the relative susceptibility to infection from food-borne pathogens of different groups of people in society, e.g., immunocompromised, pregnant, aged, other factors? Where do we find this information?

Panel Discussion: Question 6

- Where can we find more information about practical applications of predictive modeling and risk assessment?

Dr. Tom Ross

Dr. Peter Taormina

Dr. Marcel Zwietering

Dr. Betsy Booren

Dr. Yuhuan Chen

Audience Questions & Answers

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- David Tharp
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- Tamara Ford

Practical Applications of Microbial Modelling Webinar Series

Recordings of webinar series

- ✔ Part I – Overview & Practical Applications
  - November 29, 2017 (Q&A Document Now Available!)
    https://www.foodprotection.org/upl/downloads/library/qa-11-29-webinar.pdf
- ✔ Part II – Inactivation
  - March 5, 2018 (Q&A Document Now Available!)

    https://www.foodprotection.org/upl/downloads/library/3-5-18-webinar-slides.pdf
- ✓ Part III – Risk Modeling
  - Recording to be posted on IAFP website
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