

*“Challenges in the
holistic risk
assessment of the
21st century”*

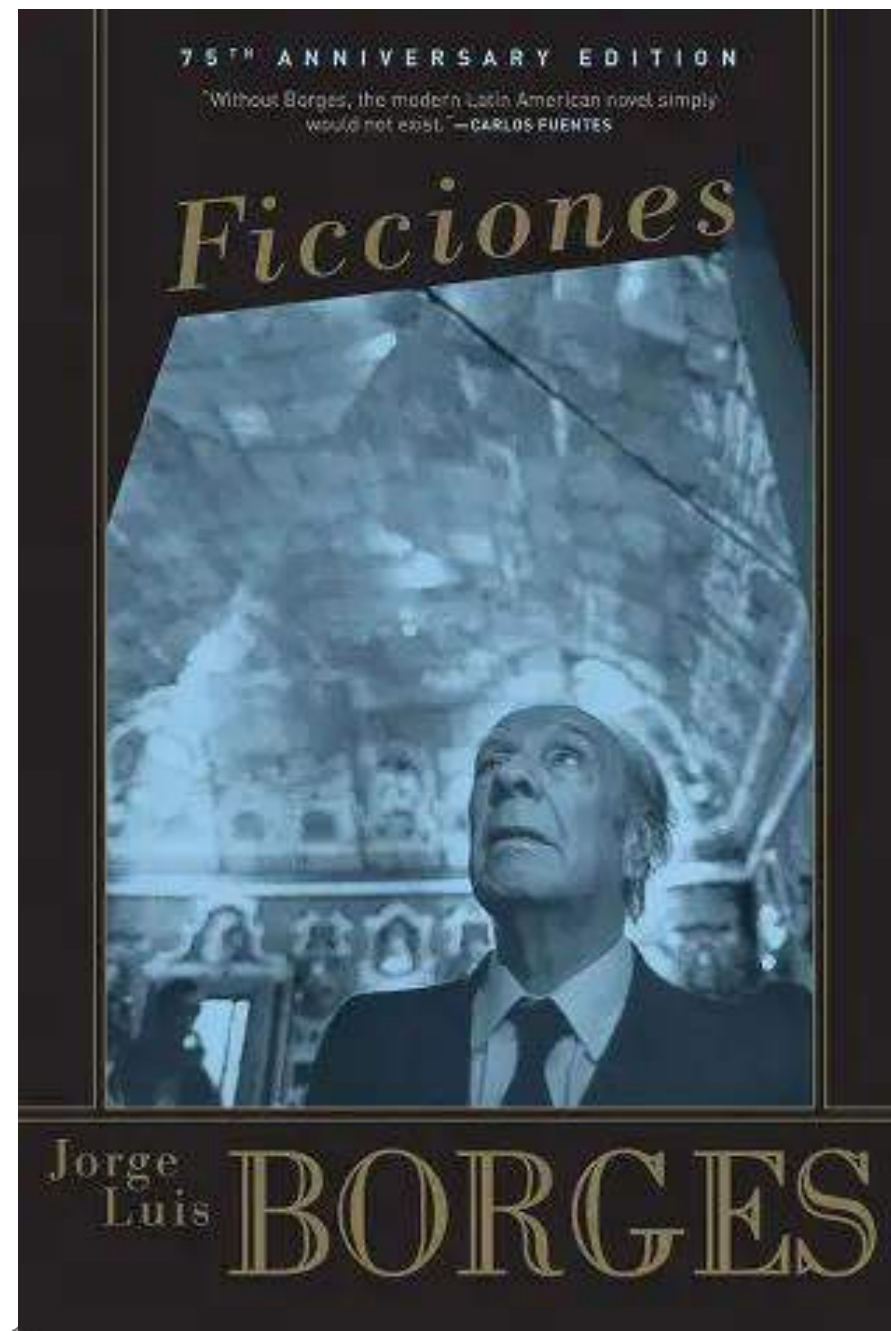
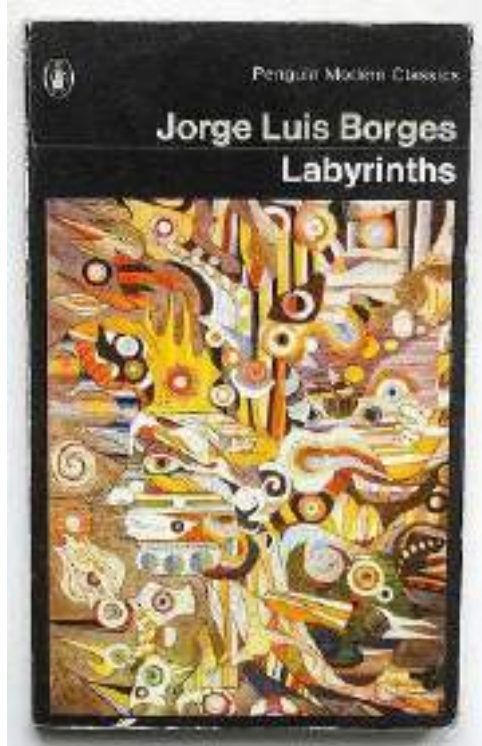
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Professor

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Knowledge is memory
Novelty is oblivion

Francis Bacon (adopted from Plato,
the ancient Greek!)

The One-Health Challenge

Emerging risks Resilience
Drivers Early warning
Food supply chain analysis
Combined exposure
Cost-benefit



SCIENTIFIC OPINION

ADOPTED: 25 October 2018

doi: 10.2903/j.efsa.2018.5482

Evaluation of the safety and efficacy of the organic acids lactic and acetic acids to reduce microbiological surface contamination on pork carcasses and pork cuts

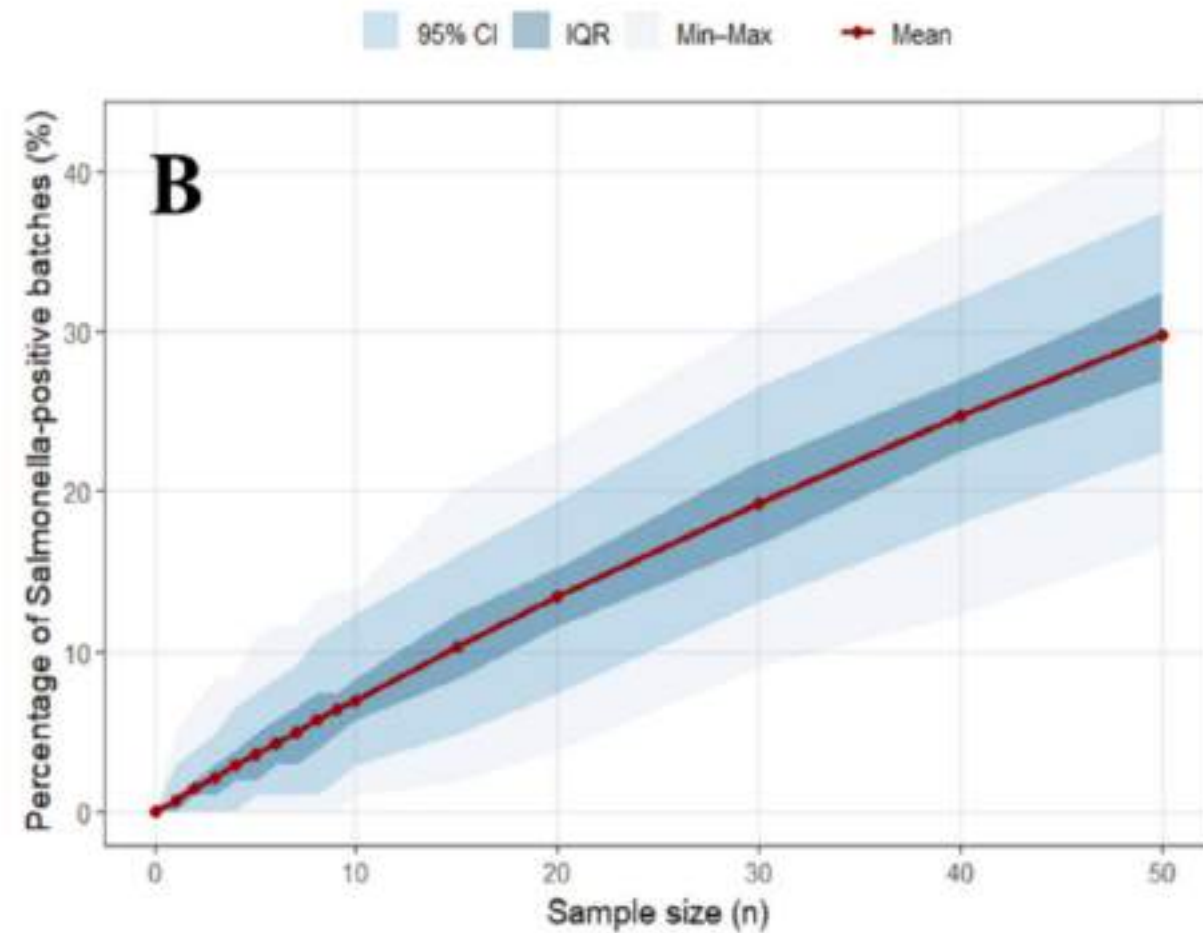
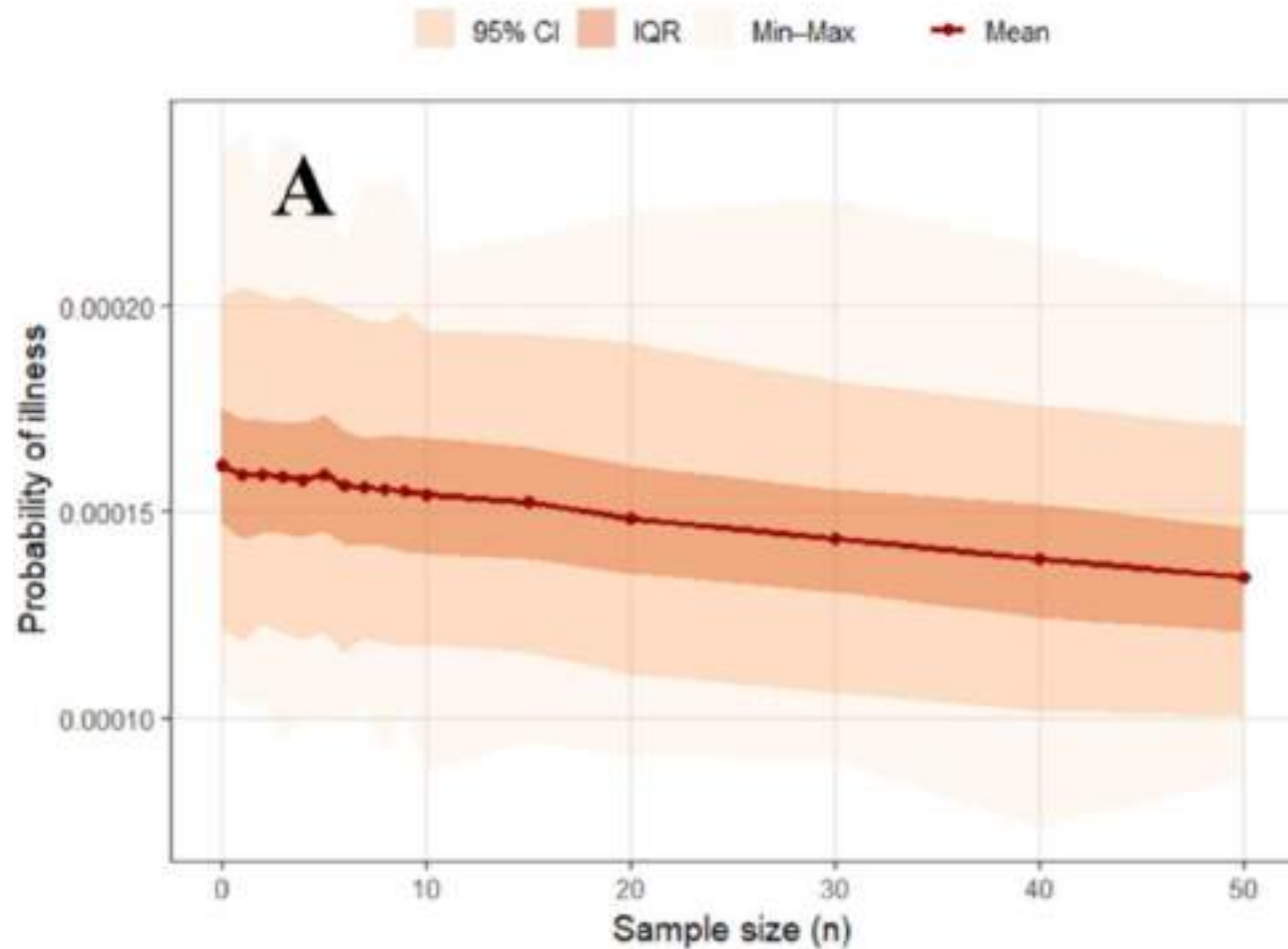
TERMS OF REFERENCE

- The **toxicological safety** of the substance
- The **efficacy**, i.e., does the individual use of the substance significantly reduce the level of contamination of pathogens on the target food
- The **potential emergence of reduced susceptibility to biocides and/or resistance to therapeutic antimicrobials** linked to the use of the substance
- The risk related to the release of the processing plant effluents, following the use of the substances **in the environment**

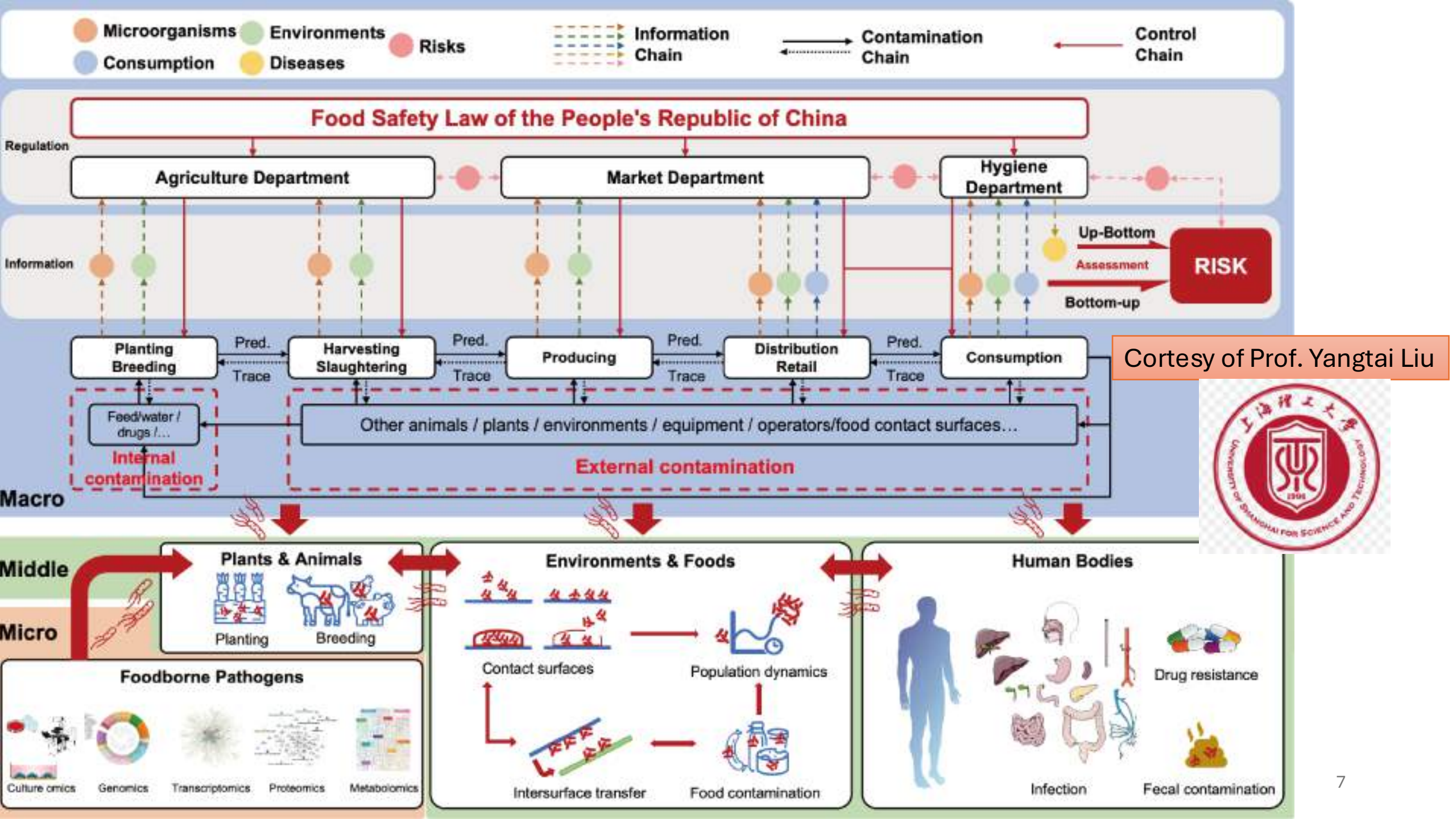
Well-known cost-benefit dilemmas

- Nitrites in cooked meat products ->
 - Nitrosamines vs control of *Clostridium botulinum*
- Chlorination in fresh produce washing ->
 - Tri-halogenes vs control of microbial contamination in washing water
- Thermal processing of tomato-based products ->
 - Formation of furans vs control of spore-formers

Cost-benefit in sampling to prevent diseases



Stathas et al. (2026) IJFM 111709



Courtesy of Prof. Yangtai Liu



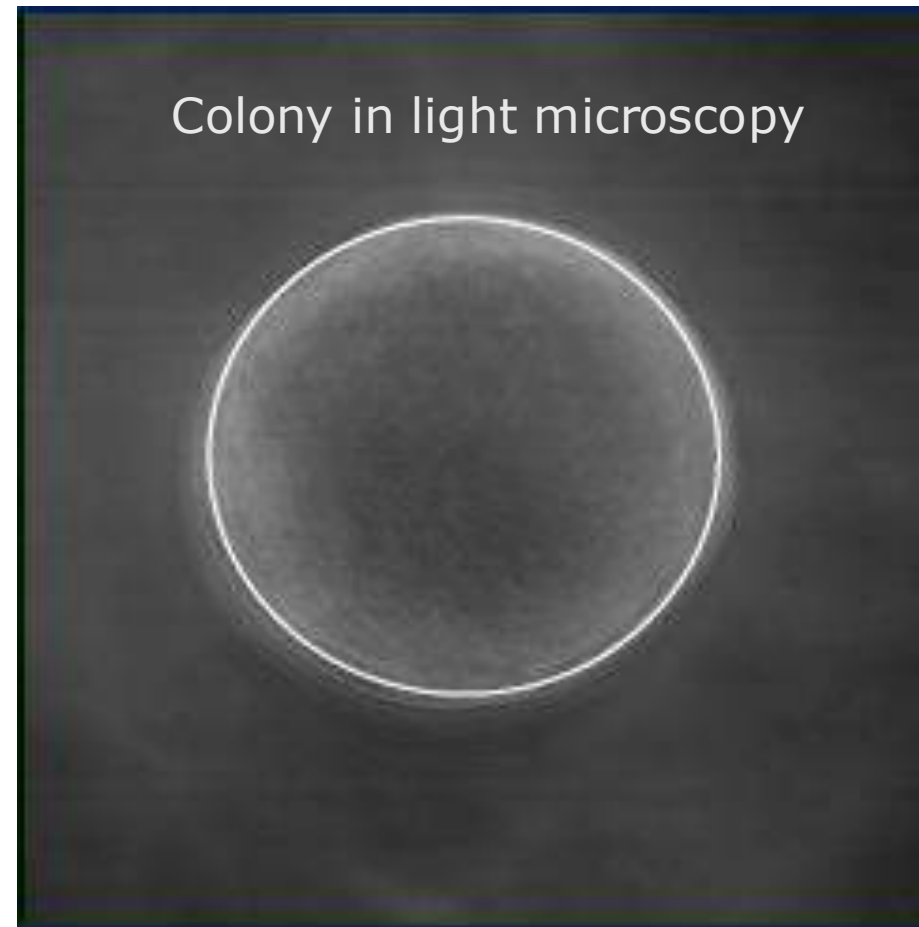
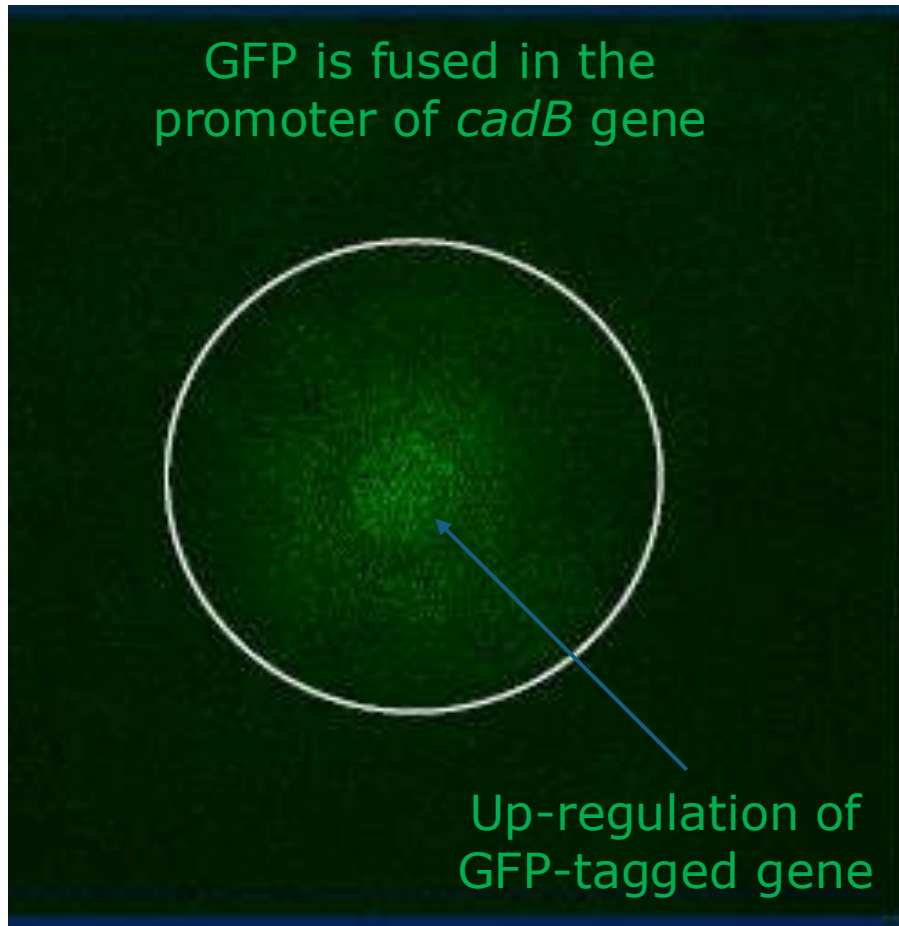
Microbiological challenges

Challenge 1: Variability

- Variability exists everywhere
- Variability makes our life beautiful
- But variability causes (food safety) “headaches”

Variability in cellular physiology within a single colony!

Courtesy of D. Wilson and T. Brocklehurst. Institute of Food research, Norwich (slide from 2000!!!!)





Defining bacterial heterogeneity and dormancy with the parallel use of single-cell and population level approaches

Marianna Arvaniti and Panagiotis N Skandamis



RESEARCH ARTICLE



Listeria monocytogenes Sublethal Injury and Viable-but-Nonculturable State Induced by Acidic Conditions and Disinfectants

Marianna Arvaniti,^a Panagiotis Tsakanikas,^b Vasiliki Papadopoulou,^a Artemis Giannakopoulou,^a Panagiotis Skandamis^a

Arvaniti et al.
2022, 2023, 2024



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International Journal of Food Microbiology

journal homepage: www.elsevier.com/locate/ijfoodmicro



Deciphering the induction of Listeria monocytogenes into sublethal injury using fluorescence microscopy and RT-qPCR



Marianna Arvaniti^a, Panagiotis Tsakanikas^b, Spiros Paramithiotis^{a,c}, Vasiliki Papadopoulou^a, Athanasios Balomenos^a, Artemis Giannakopoulou^a, Panagiotis Skandamis^{a,*}

PN Skandamis: Joined FoodSafeR HoliFood meeting, 9-12 June 2026

Variability in generation times among cells in different microcolonies

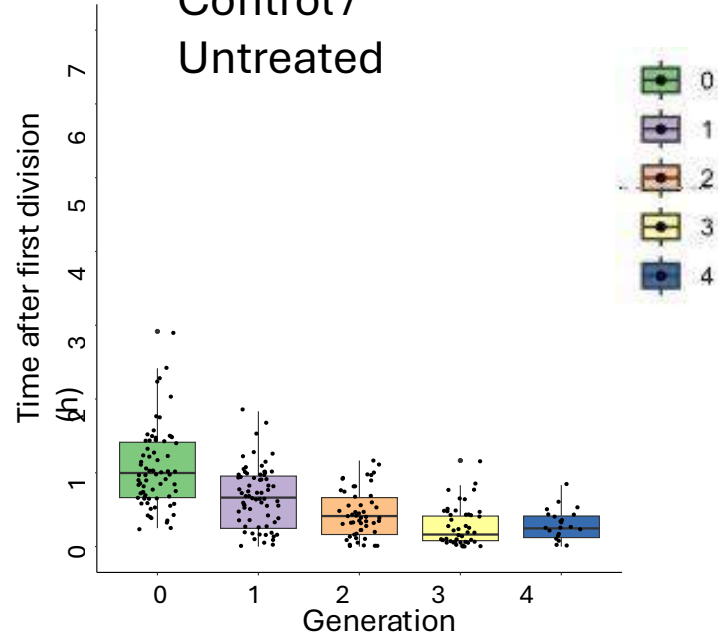


Treatment with 30 ppm PAA → 2 sub-populations with different growth dynamics:

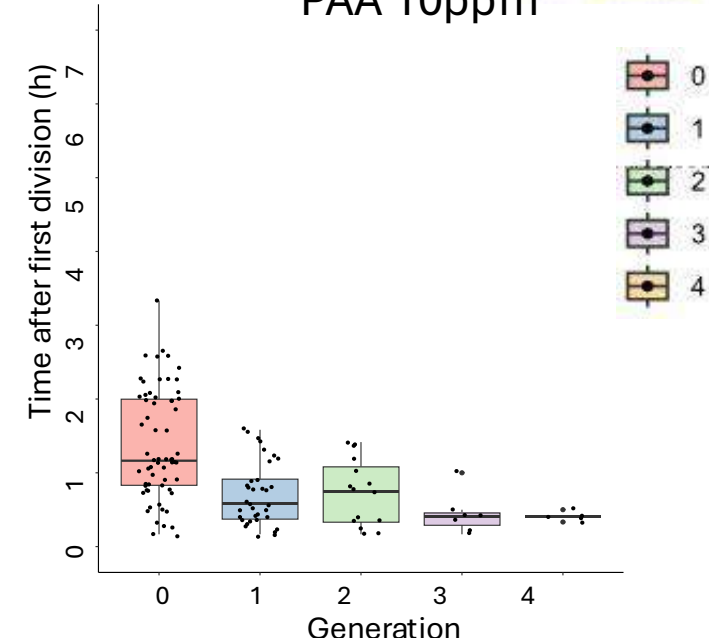
bimodal probability distribution of the lag times.

3. Division time variability

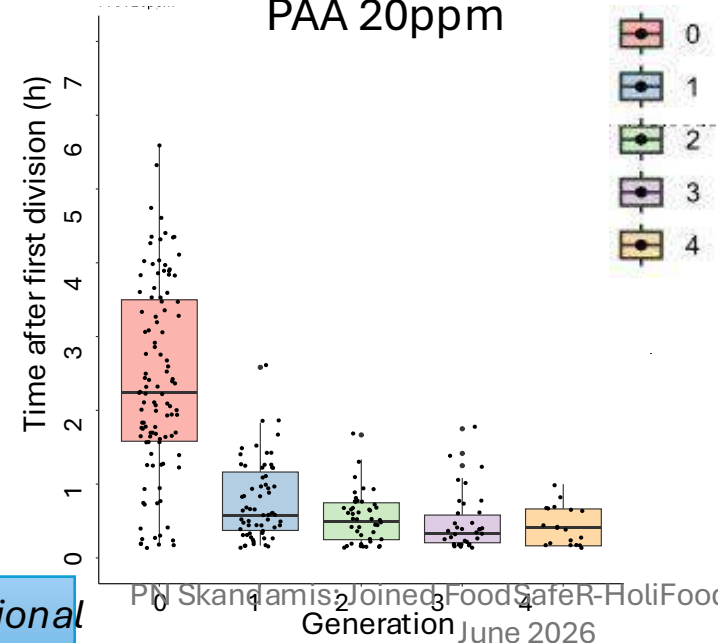
Control /
Untreated



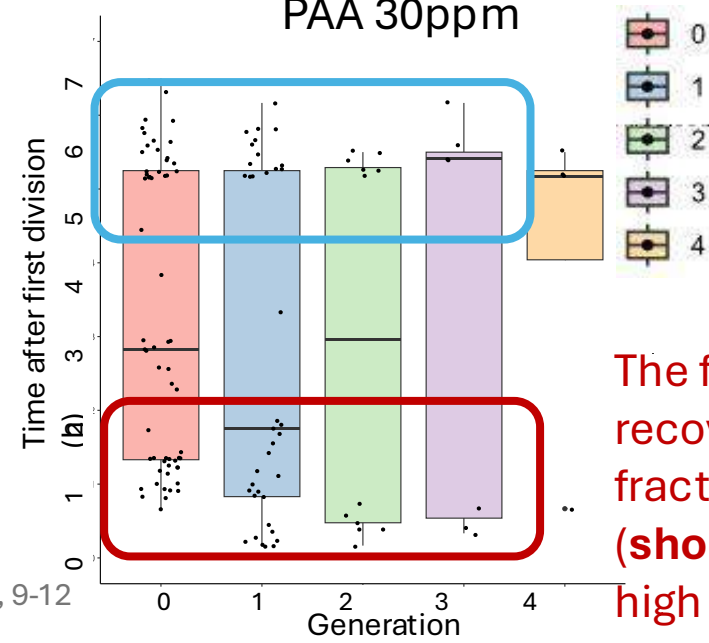
PAA 10ppm



PAA 20ppm



PAA 30ppm



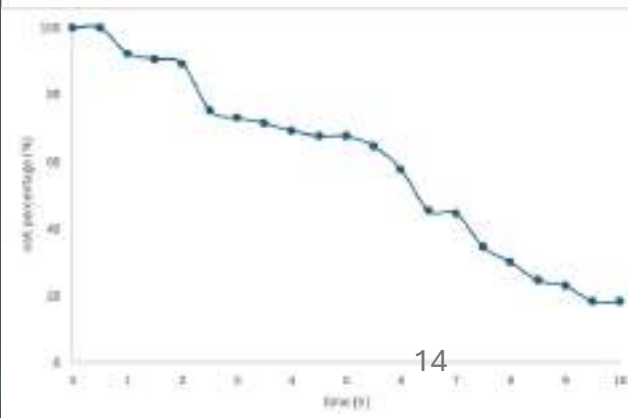
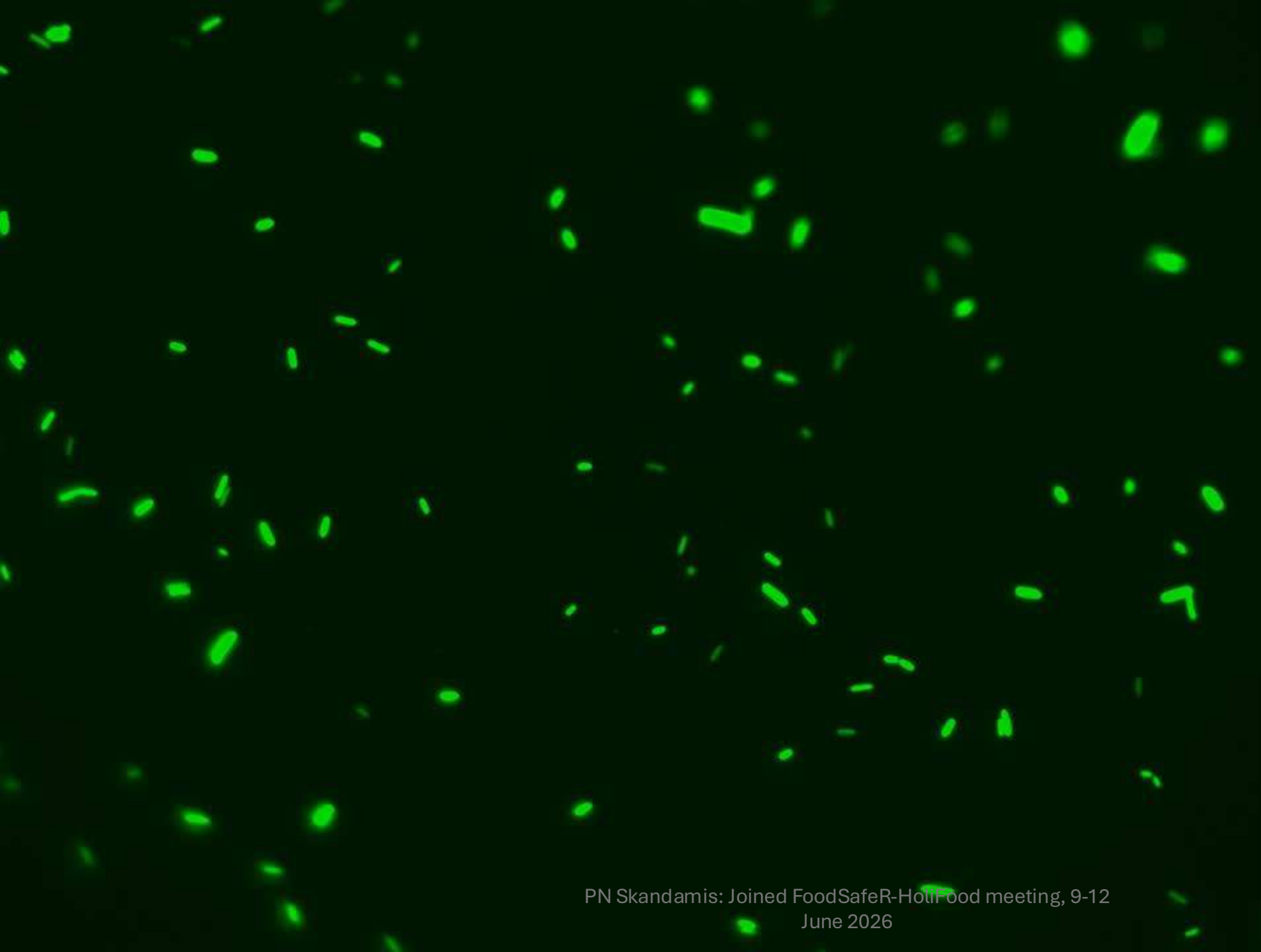
Severely injured (long lag) = Low risk

The fastest recovering fraction (short lag) = high risk

1.30h

Stochastic
inactivation of
L. monocytogenes
by lytic phages

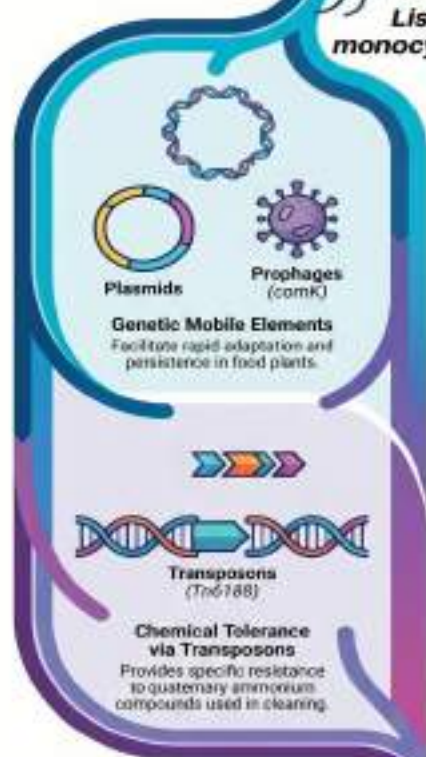
**Alive *Listeria* cells after
exposed to PGL for 10h**



Survival Strategies of *Listeria monocytogenes*: Understanding Microbial Variability

Strain Diversity

Variation Between Different Strains
- Diversity across different genetic lineages determines long-term survival in specific environments.



Genetic Elements
Plasmids & Prophages (comK)
Supports niche-specific adaptation and persistence in processing plants.

Population Heterogeneity

Phenotypic vs. Genotypic Variants
- Heterogeneity includes transient "persister" cells and stable, inheritable stress-resistant mutants.



System Responses
SOS Response & TA Systems
RecA, HipA, or MazEF
Triggers dormant states to survive lethal stress.

Targeted Mutations
ctsR & *rspU*
Increases resistance to heat, pressure, and acidic environments.

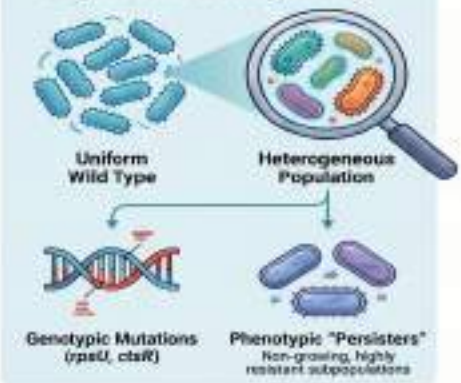
Why Pathogens Survive: The Science of Microbial Variability

How food characteristics, strain diversity, and processing parameters combine to create unpredictable microbial growth and survival patterns

1) Product Characteristics (Intrinsic Factors)

Food Matrices
Intrinsic factors like milk and meat composition dictate growth.

2) Strain Diversity & Population Heterogeneity



3) Extrinsic Properties

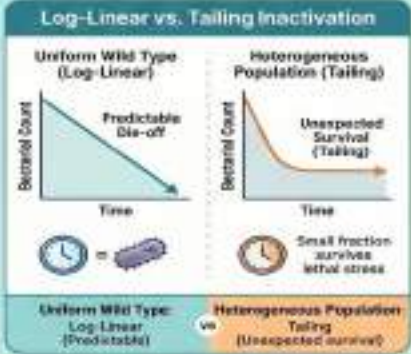
Temperature Variability
Storage temperatures significantly affect growth rates.

4) Food Processing Parameters

Thermal Pasteurization
Inadvertently selects for heat-resistant variants

High Hydrostatic Pressure (HHP)
Pressure treatment can leave robust variants unharmed

OUTCOME: Unpredictable Survival Patterns & Persistence



The Growth-Survival Trade-off



Persistence in the Food Chain

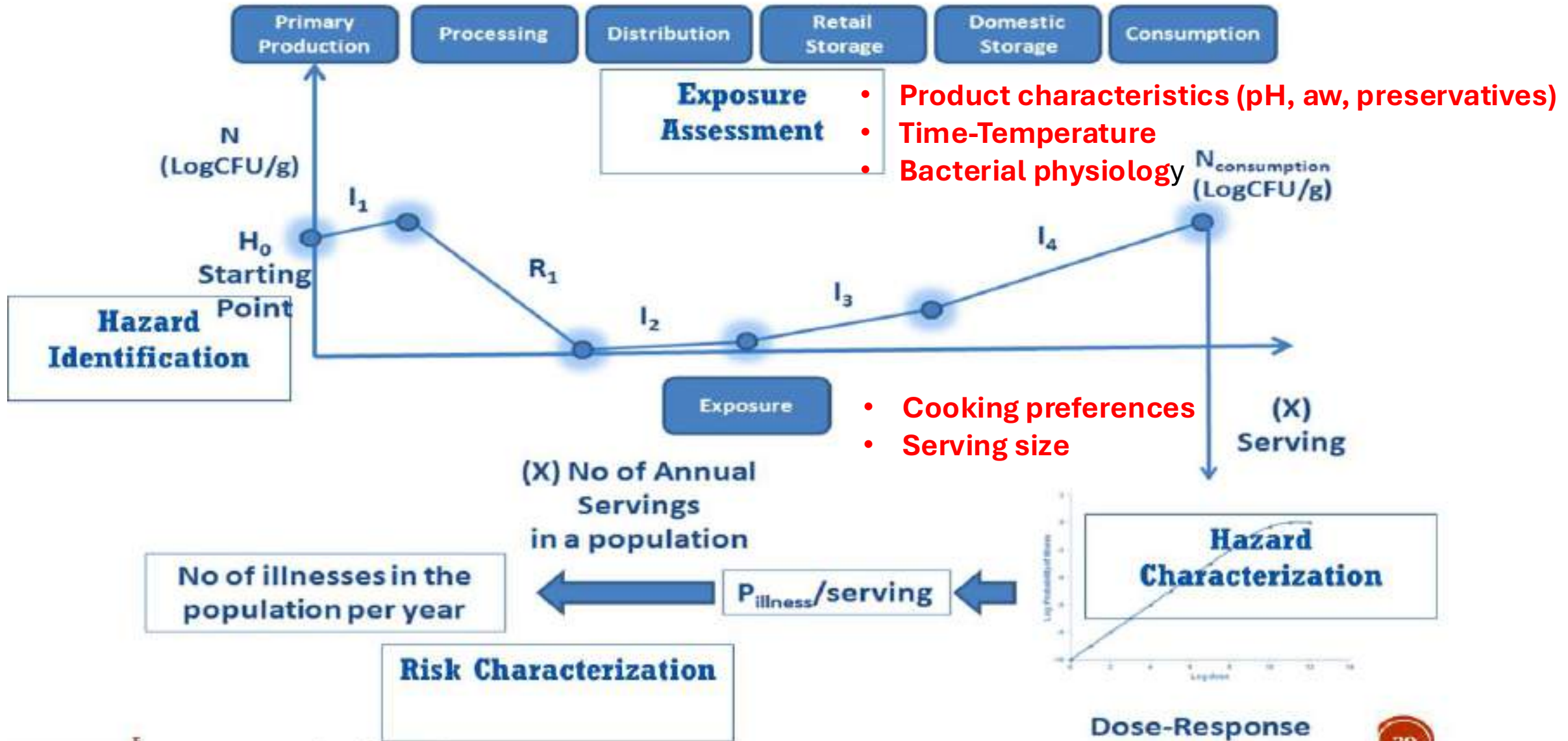


Challenge 2: Probabilities matter

- Probabilistic (risk-based) way of thinking is the only way
- This is how we can accommodate variability and uncertainty

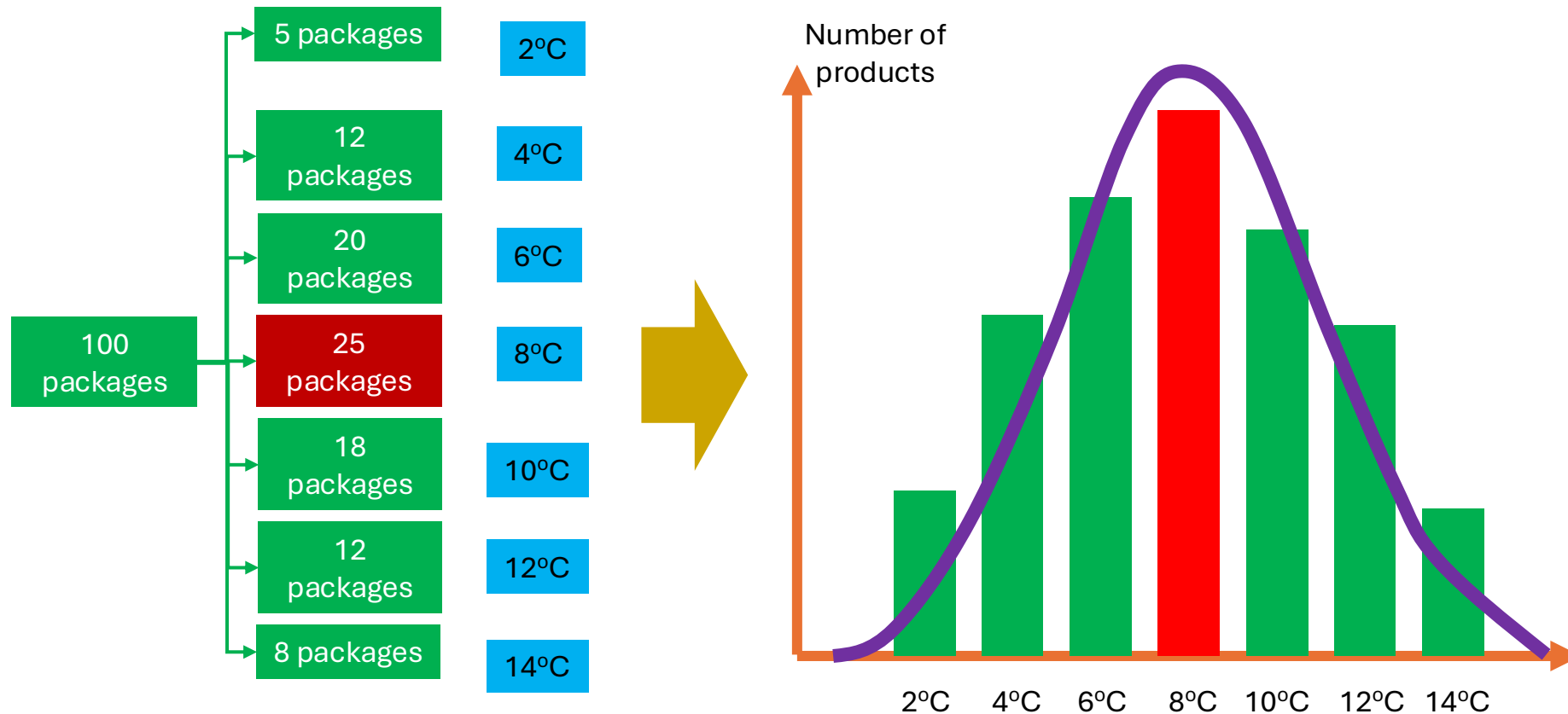
Risk Assessment Infograph:

Variability sources

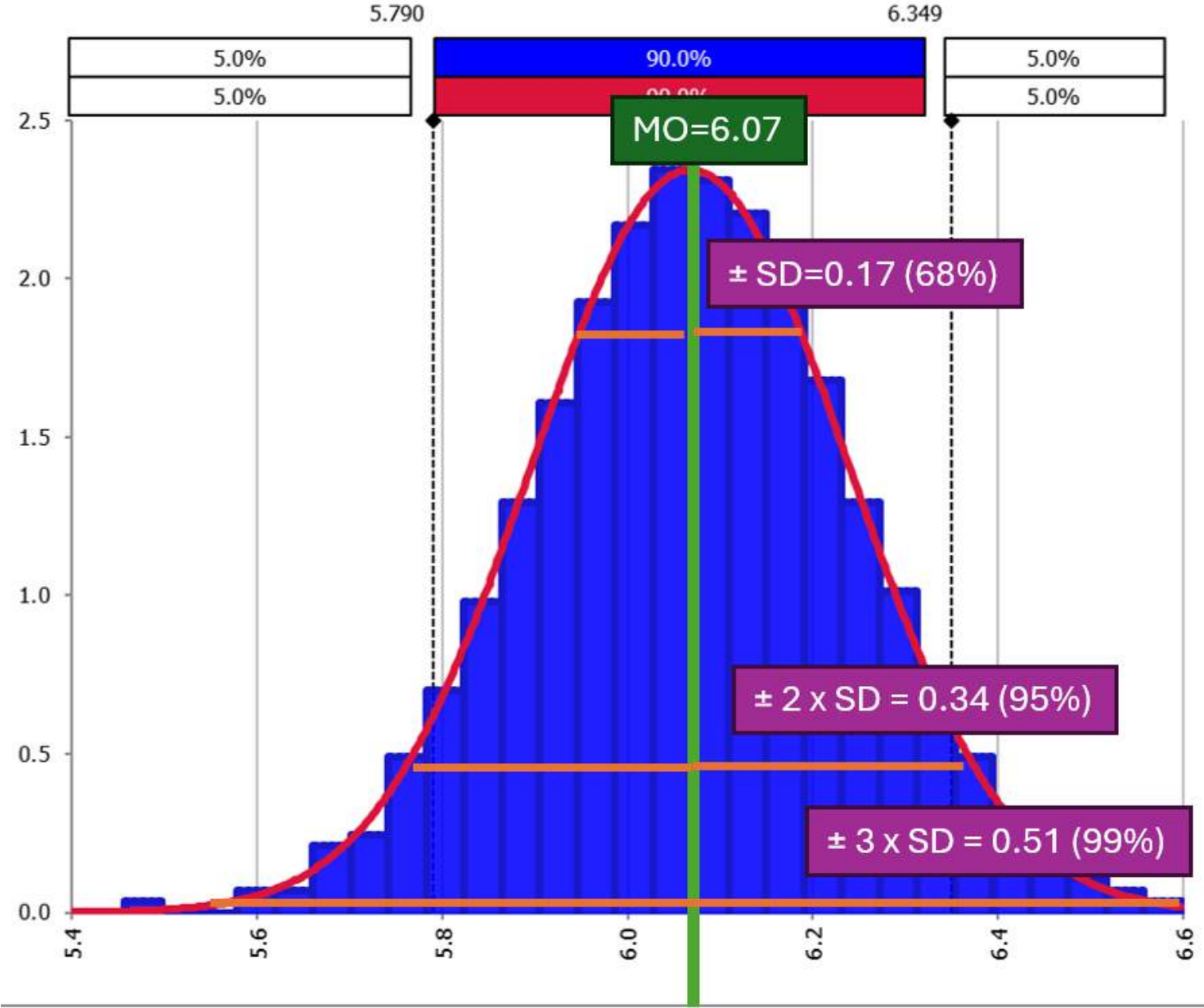


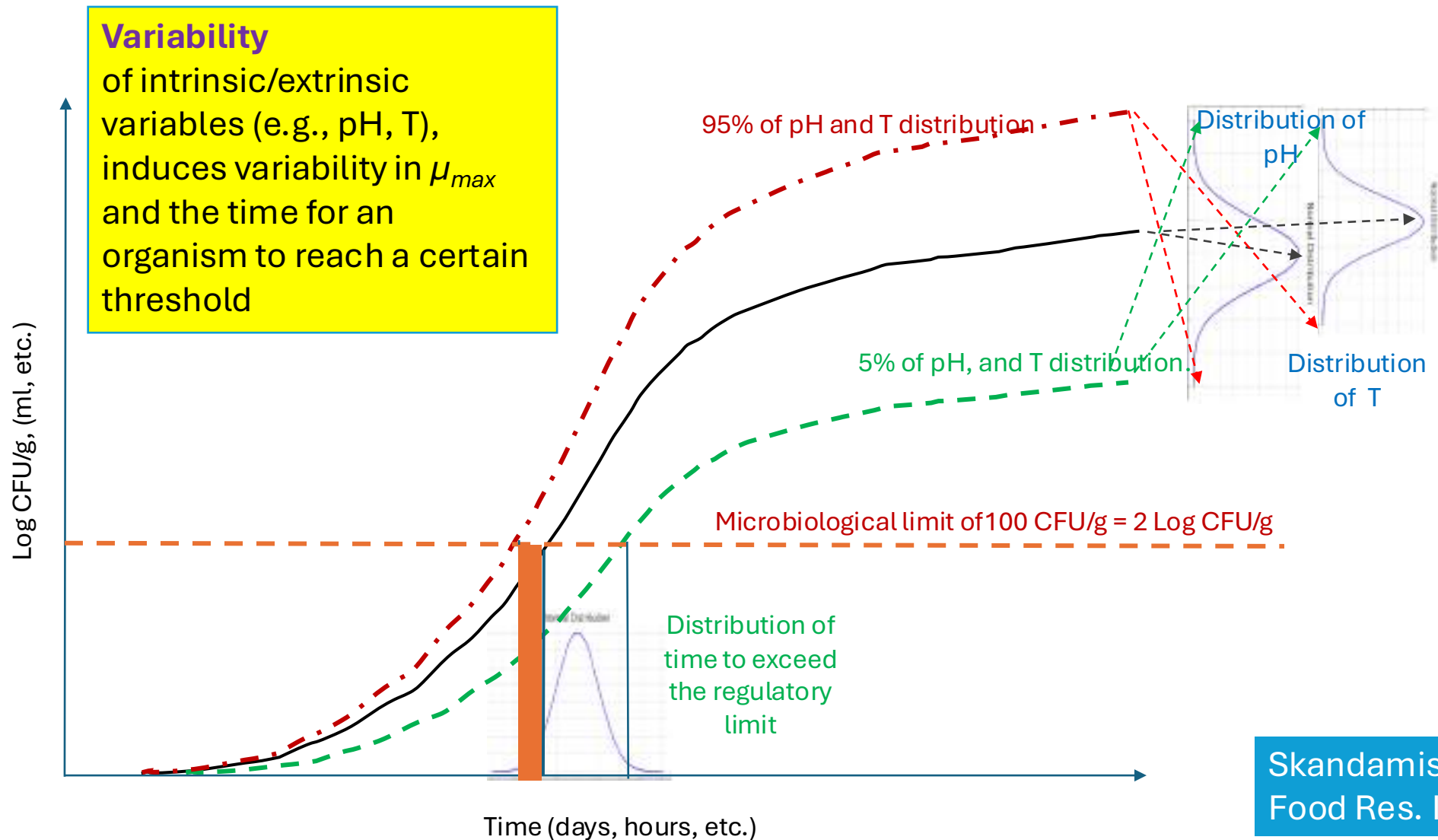
Stochasticity in the food supply chain

Extrapolate this example by replacing Temperature with:
time on the shelf, product characteristics, etc.



Variability of intrinsic properties (pH)

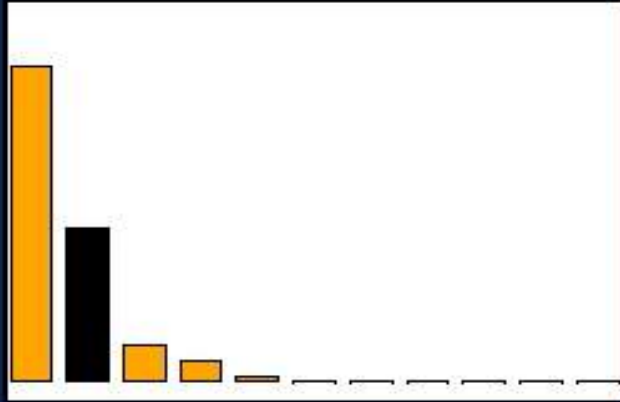




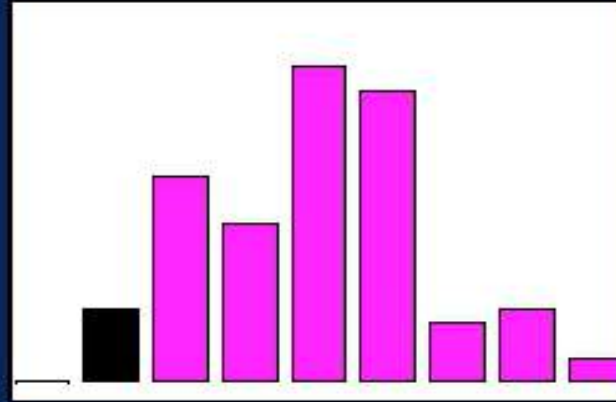
<https://skandamis.shinyapps.io/Microbial-Growth-Predictor-Dashboard/>

UNCERTAINTY challenge: How sure we are about the shape of each distribution, or the mathematical formula of a Dose-Response model?

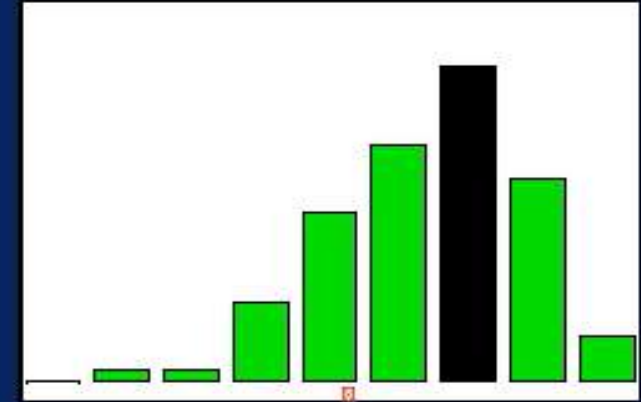
INPUT: Concentration



INPUT: Storage

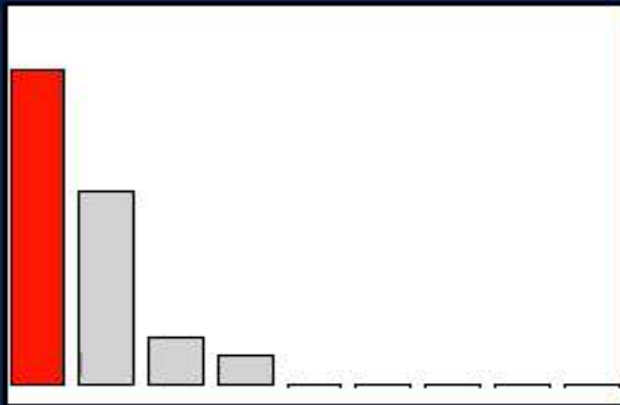


INPUT: Cooking

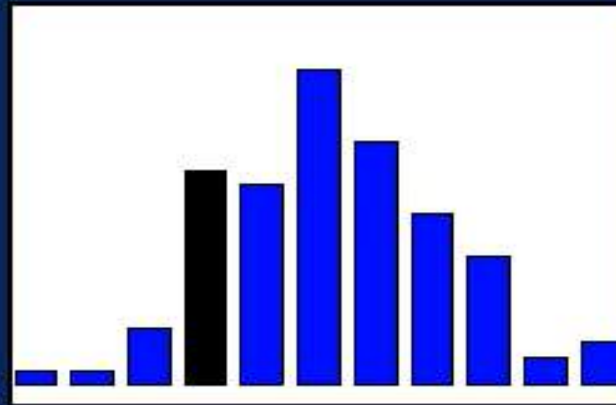


Monte Carlo simulation

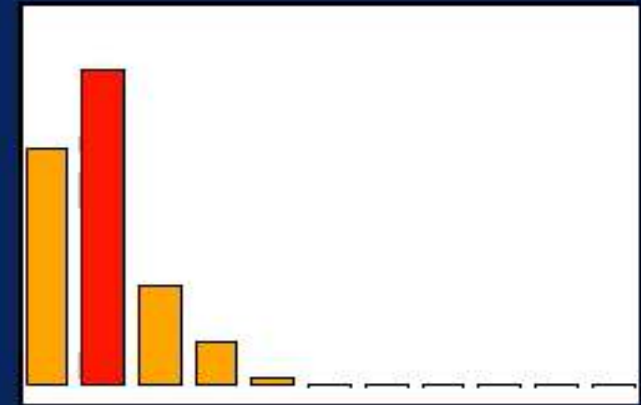
OUTPUT: Dose



INPUT: Amount



OUTPUT: Cooked Product



Challenge 3: Make the most out of data

Big data = big challenges

From *Data Collection* to *Data Connection*

"To those who devote their lives to science, nothing can give more happiness than making discoveries, but their cups of joy are full only when the results of their studies find practical applications".

Louis Pasteur

Selection Page

Extract Table

Videometer

SAMPLE TYPE GROUPS

- FINAL PRODUCTS
- INTERMEDIATE PRODUCTS
- MEAT PREPARATIONS
- RAW MATERIALS
- SURFACES

Sample Condition

All

Sample Description

All

Sample Code

All

Producer

All

Listeria

All

Salmonella

All

Year, Month, Day

All

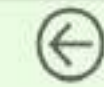
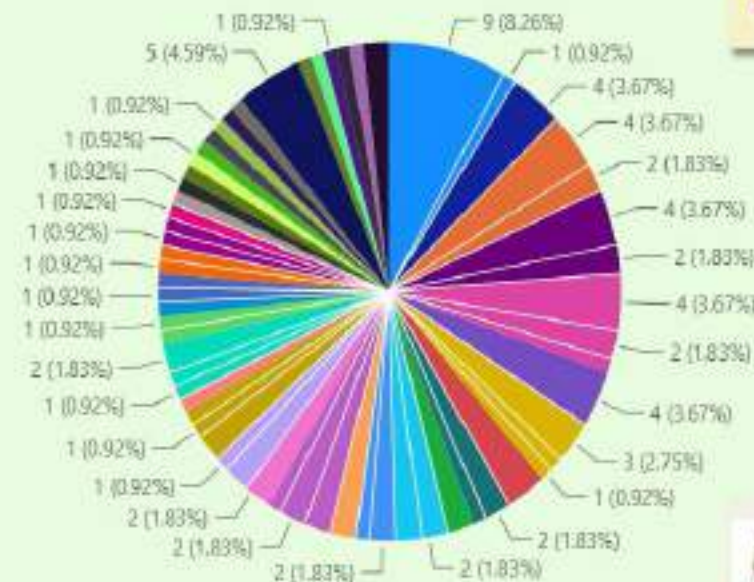
Summary table

a/a	SAMPLE TYPE GROUPS	Producer
1	FINAL PRODUCTS	
1	FINAL PRODUCTS	
1353		

Sample Description

- ΒΟΕΙΟ
- ΓΑΛΟΠΟΥΛΑ
- ΓΕΜΙΣΜΕΝΟ ΣΑΛΑΜΙ ΑΕΡΟΣ
- ΜΠΙΦΤΕΚΙ ΒΟΕΙΟ
- ΜΠΙΦΤΕΚΙ ΚΟΙΤΟ
- ΣΤΙΣΙΑ ΒΟΕΙΟ
- ΣΟΥΒΛΑΚΙ ΚΟΤΟΠΟΥΛΟ
- ΤΡΑΧΗΛΟΣ ΒΟΕΙΟ
- TRIMMINGS
- ΚΟΤΣΙ
- ΚΟΥΤΕΡ
- ΛΟΥΚΑΝΙΚΑ ΚΑΡΑΜΑΝΛΙΔΙΚΑ
- ΜΟΣΧΑΡΙ
- ΣΠΑΛΑ ΧΟΡΙΝΟ
- ΣΤΡΟΓΓΥΛΟ

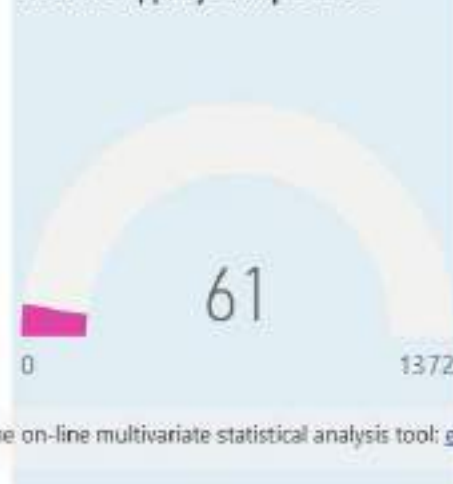
Percent of samples positive for Listeria and Salmonella



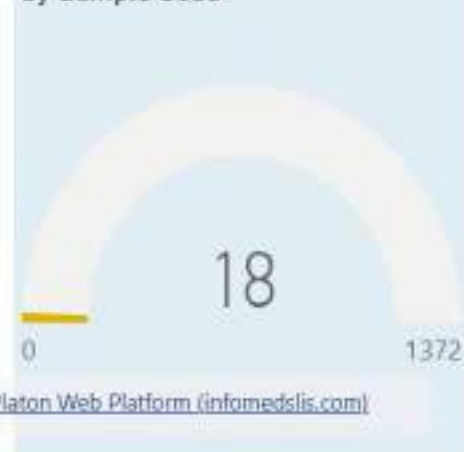
Salmonella spp. by Sample Code



Listeria spp. by Sample Code



Listeria monocytogenes (Detection) by Sample Code



Press this link to be transferred to the on-line multivariate statistical analysis tool: [ePlaton Web Platform \(infomedslis.com\)](http://ePlaton.Web.Platform.infomedslis.com)



Hygiene-pH-aw

LAB Enterobacteria

Nitrosamines

PAHs

Multispectral



Pivot Table

Custom

A typical example of multivariate machine-learning processing

This is the place for biomarkers?

1	A	B	C	J	K	L	M	N	O	P	Q	R	S	T	U	V	
2	Listeria monocytogenes isolate I	Persistent	Biofilm Formation BHC	MIC BAC	MBC BAC	FoxX	Listeria monocytogenes mp	lin	nor	testM	lmo0919 fam	Col(MG828)	Col(MP18)	Col156	ColRNAI	IncB/O/K/Z	rep25_2_MG40
3	FSR2.3_AUA_A1.29_Lmono	Persistent	0.355	3.7	5.9	1	1	1	1	0	1	0	0	0	0	0	
4	FSR2.3_AUA_A1.14_Lmono	Persistent	0.309	3.4	3.4	1	1	1	1	1	1	0	0	0	0	0	
5	FSR2.3_AUA_A1.46_Lmono	Persistent	0.138	4.8	6.5	1	1	1	1	0	1	0	0	0	0	0	
6	FSR2.3_AUA_A1.9_Lmono	Non-Persistent	0.408	3.8	3.5	1	1	1	1	0	1	0	0	0	0	0	
7	FSR2.3_AUA_A2.2_Lmono	Persistent	0.374	3.4	3.7	1	1	1	1	0	1	0	0	0	0	0	
8	FSR2.3_AUA_A2.27_Lmono	Persistent	0.375	4.0	4.6	1	1	1	1	0	1	0	0	0	0	0	
9	FSR2.3_AUA_A3.10_Lmono	Non-Persistent	0.241	4.0	5.6	1	1	1	1	0	1	0	0	0	0	0	
10	FSR2.3_AUA_A3.32_Lmono	Persistent	0.145	3.3	3.3	1	1	1	1	0	1	0	0	0	0	0	
11	FSR2.3_AUA_A3.7_Lmono	Persistent	0.142	3.5	4.2	1	1	1	1	0	1	0	0	0	0	0	
12	FSR2.3_AUA_A4.38_Lmono	Persistent	0.143	3.4	3.3	1	1	1	1	0	1	0	0	0	0	0	
13	FSR2.3_AUA_B4.15_Lmono	Non-Persistent	0.318	6.3	7.1	1	1	1	1	0	1	0	0	0	0	0	
14	FSR2.3_AUA_B4.23_Lmono	Non-Persistent	0.338	6.3	7.6	1	1	1	1	0	1	0	0	0	0	0	
15	FFoQSI_B.1.3_Lmono	Persistent	0.14	3.5	4	1	1	1	1	0	1	0	0	0	0	0	
16	FFoQSI_B.1.7_Lmono	Persistent	0.14	4	4	1	1	1	1	0	1	0	0	0	0	0	
17	FFoQSI_B.2.38_Lmono	Persistent	0.14	4	4	1	1	1	1	0	1	0	0	0	0	0	
18	FFoQSI_B.2.45_Lmono	Persistent	0.14	4	4	1	1	1	1	0	1	0	0	0	0	0	
19	FFoQSI_C.1.3_Lmono	Persistent	0.14	4	4	1	1	1	1	0	1	0	0	0	0	0	
20	FFoQSI_C.1.36_Lmono	Persistent	0.14	3.5	4	1	1	1	1	0	1	0	0	0	0	1	
21	FFoQSI_C.1.50_Lmono	Persistent	0.14	4.8	5	1	1	1	1	0	1	0	0	0	0	0	
22	FSR2.3_AUA_A4.27_Lmono	Non-Persistent	0.387	3.6	4.8	1	1	1	1	0	1	0	0	0	0	0	
23	FSR2.3_AUA_A4.47_Lmono	Persistent	0.140	3.8	3.8	1	1	1	1	0	1	0	0	0	0	0	
24	FSR2.3_AUA_B1.13_Lmono	Non-Persistent	0.224	3.2	3.7	1	1	1	1	0	1	0	0	0	0	0	
25	FSR2.3_AUA_B3.26_Lmono	Non-Persistent	0.346	3.7	4.2	1	1	1	1	1	1	0	0	0	0	0	
26	FSR2.3_AUA_B4.12_Lmono	Non-Persistent	0.342	3.3	4.3	1	1	1	1	0	1	0	0	0	0	0	
27	FSR2.3_UBU_A2.50_Lmono	Persistent	0.4	3.5	4.2	1	1	1	1	0	1	0	0	0	0	0	
28	FSR2.3_AUA_A3.8_Lmono	Non-Persistent	0.399	3.6	3.6	1	1	1	1	0	1	0	0	0	0	0	
29	FFoQSI_B.1.56_Lmono	Non-Persistent	0.14	3.5	4	1	1	1	1	0	1	0	0	0	0	0	
30	FSR2.3_UBU_D3.2	Non-Persistent	0.14	3.5	4.5	0	0	0	0	0	0	0	0	0	0	0	
31	FSR2.3_AUA_A1.15_Lmono	Non-Persistent	0.400	3.4	3.5	1	1	1	1	1	1	0	0	0	0	0	
32	FSR2.3_AUA_A4.15_Lmono	Persistent	0.353	3.4	5.8	1	1	1	1	0	1	0	0	0	0	0	
33	FSR2.3_AUA_B3.2_Lmono	Non-Persistent	0.401	3.7	3.8	1	1	1	1	0	1	0	0	0	0	0	
34	FSR2.3_AUA_B4.20_Lmono	Non-Persistent	0.296	3.3	3.7	1	1	1	1	0	1	0	0	0	0	0	
35	FSR2.3_AUA_B4.32_Lmono	Non-Persistent	0.249	3.3	3.6	1	1	1	1	0	1	0	0	0	0	0	
36	FSR2.3_UBU_E2.30_Lmono	Non-Persistent	0.14	3.5	4	1	1	1	1	0	1	0	0	0	0	0	
37	FSR2.3_UBU_H1.15_Lmono	Non-Persistent	0.14	4	4	0	0	0	0	0	0	0	0	0	0	0	
38	FSR2.3_UBU_D2.17_Lmono	Non-Persistent	0.14	3.8	4	0	0	0	0	0	0	0	0	0	0	0	

Multivariate Food Predictor

1. Load Training Dataset

Browse... No file selected

Download an XL Template
Specify the number of variables

19

- Dependent variable type
- Continuous (ANN, RFR, Gradient Boost, SVM, PLSR)
 - Categorical (RFR, Gradient Boost, SVM, PLS-DA)

- Actions for fitting (pre-processing)
- Log10 transformation of dependent variable
 - Autoscaling of independent variables (vertical normalization)
 - Mean centering (vertical normalization)
 - Savitzky-Golay smoothing (horizontal normalization)
 - SNV transition to independent variables (horizontal normalization)

- Choose derivative
- 1st derivative
 - 2nd derivative

- Choose polynomial order
- 2nd order
 - 3rd order

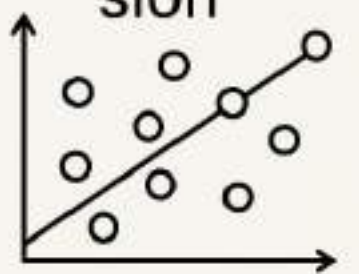
Weights for Savitzky-Golay smoothing
9

Actions for PCA

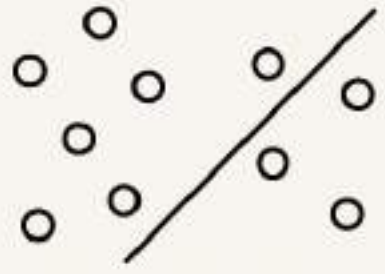
- About
- Training Graphs
- PCA
- Testing (validation) with independent data
- PLSR summary
- PLS-DA training
- Logistic regression

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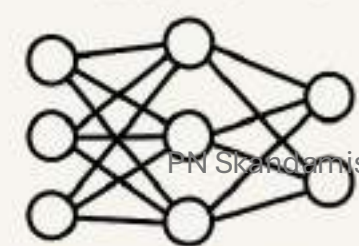
Partial Least Squares Regression



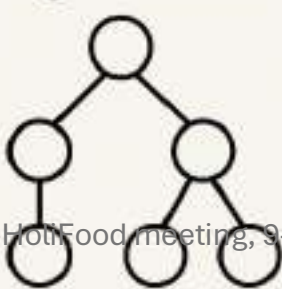
Support Vector Machines



Artificial Neural Networks



Random Forest Regression



Machine learning algorithm

- Artificial Neural Networks (1 hidden layer)
- k-Nearest Neighbor (KNN)
- Random Forest Regression (Ens. bagging)
- Adaboost (Ensemble boosting)
- Supporting Vector Machines (Linear)
- Supporting Vector Machines (Non Linear)
- Partial Least Square Regression
- Least Angle Regression (LARS)

Cross-Validation for PLSR

- k-fold CV (default for all)
- Leave-One-Out CV (only for PLSR)

Cross-Validation for RFR

- k-fold CV (default for all)
- Bootstrap (only for RFR)

Bootstrapping for CI estimation of PLSR coefficients

2. Load Testing dataset

Browse... No file selected

Use the same XL format as for training, but without the need to provide numbers for the Y variable

Predicting (assigning a P value) of persistence based on genotypic data of *L. monocytogenes*



Confusion matrix results for persistence classification from Multivariate Food Predictor, based on presence absence of (virulence) genes. A total of 316 isolates were included in the dataset, 220 of which were used for training the model, and 96 for testing of model accuracy.

Challenge 3: Ride on the future (follow the “tsunami”)

AI-in food safety

Harness the technology to move forward – revisit classical approaches

Harnessing the capacity of machine learning algorithms – going beyond (or ignoring) causality



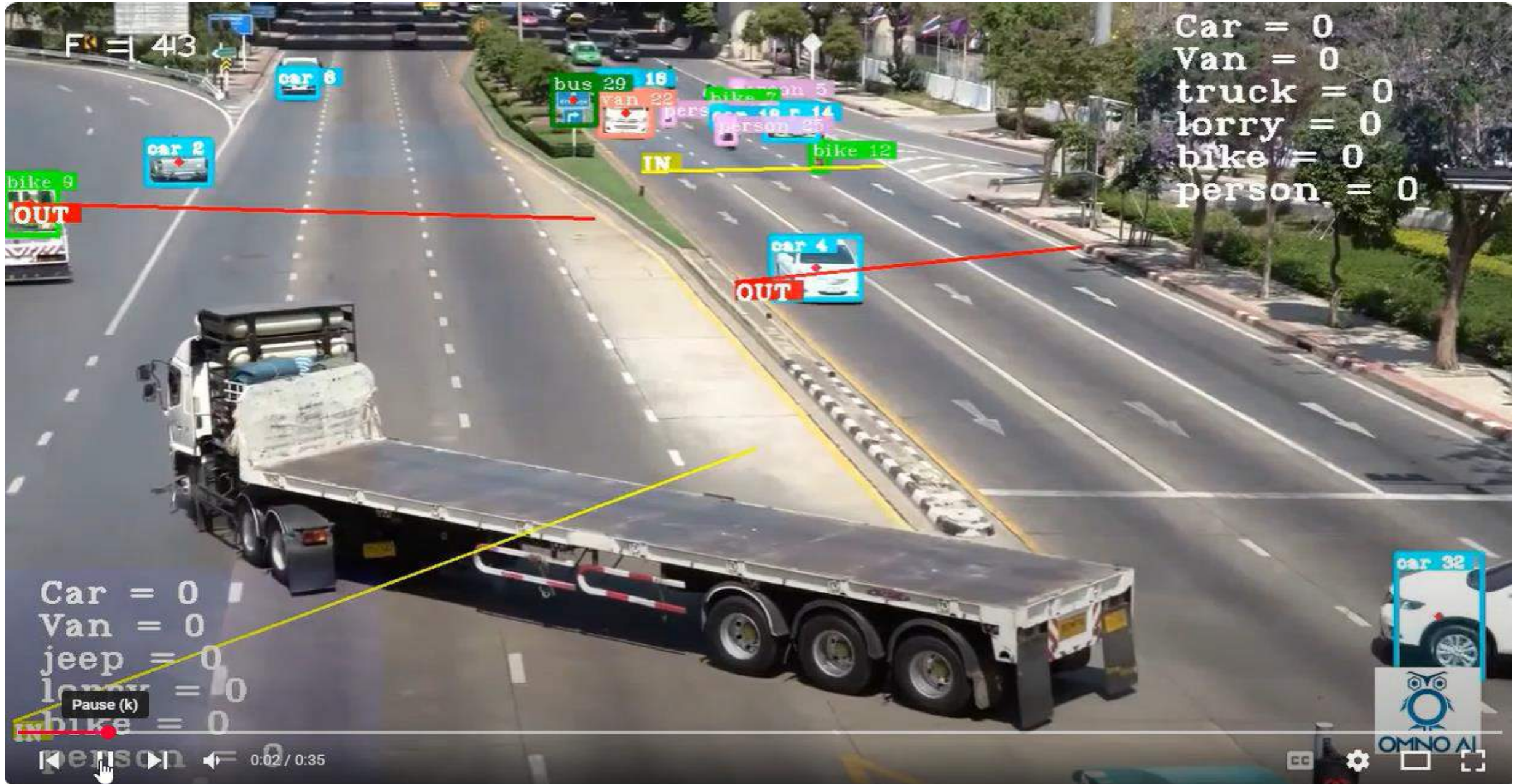
FOOD MICROBIOLOGY



Accelerating the Detection of Bacteria in Food Using Artificial Intelligence and Optical Imaging

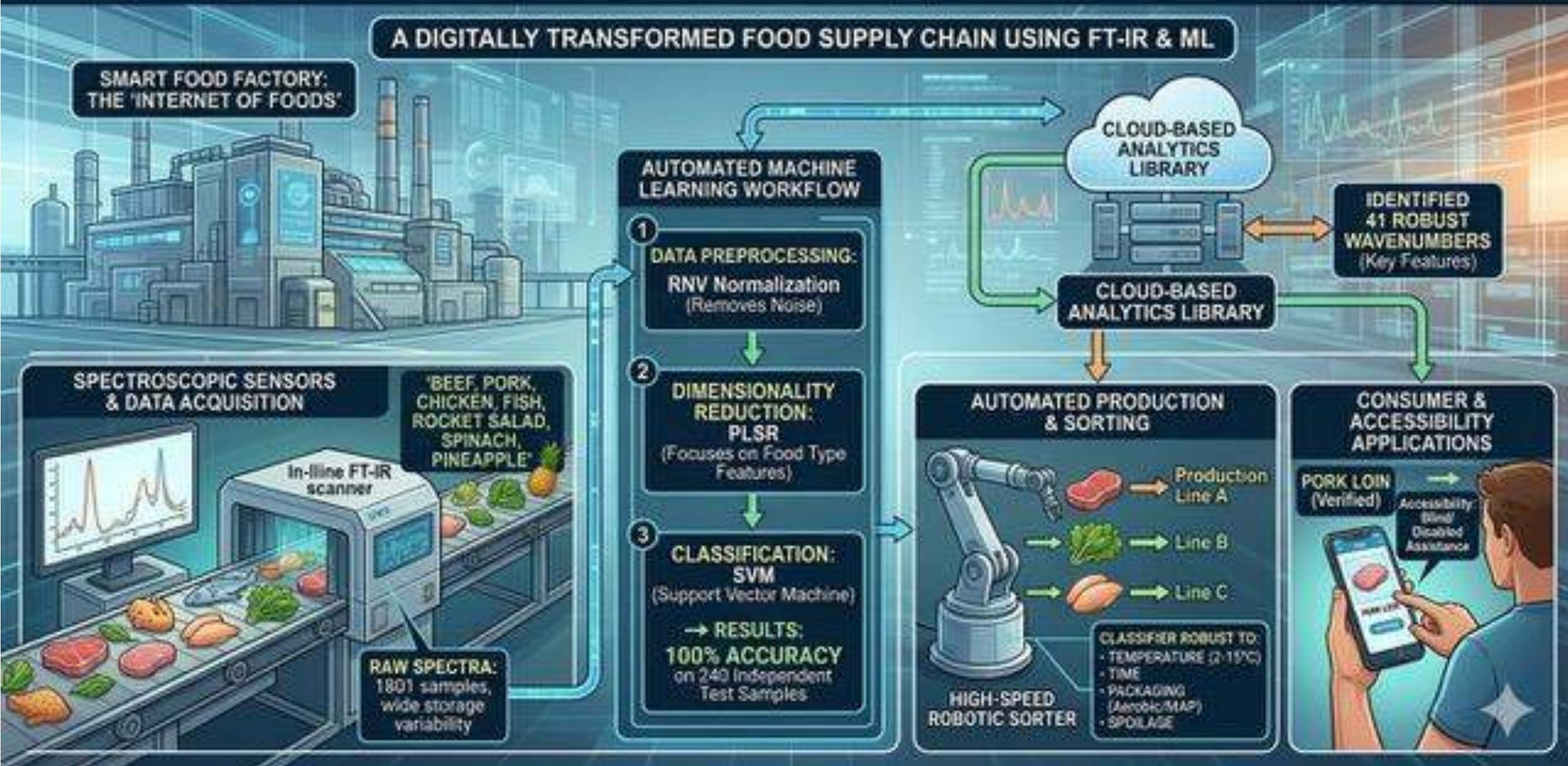
 Luyao Ma,^a  Jiyeon Yi,^b Nicharee Wisuthiphaet,^a Mason Earles,^{b,c}  Nitin Nitin^{a,b}

YOLO: Object recognition for traffic regulation



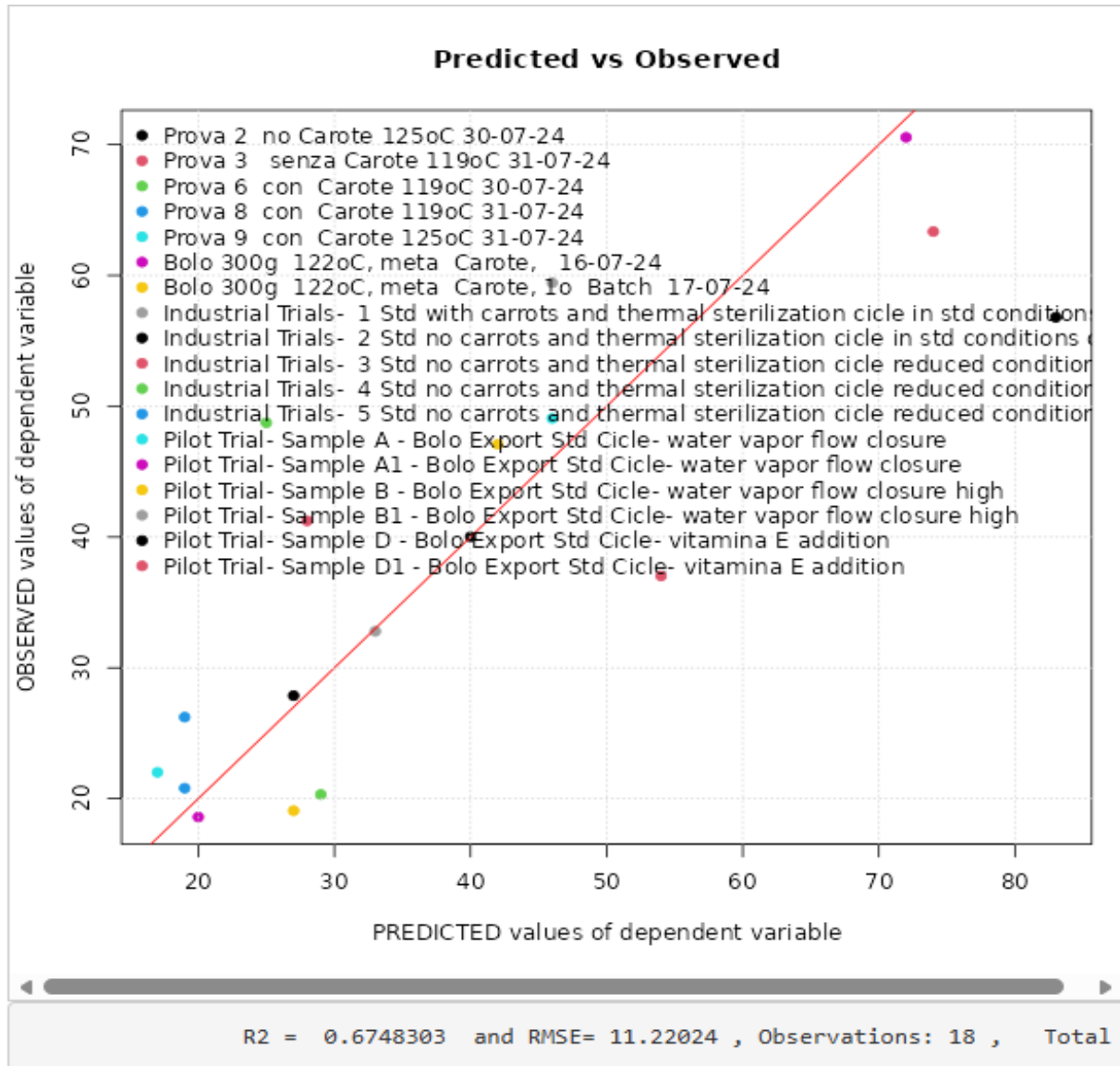
Future Food Industry: Automated Spectroscopic Classification

Based on Tsakanikas et al. (2020)



ML prediction of Furan levels in sauces based on multispectral data

Independent validation: very good!



Nr	Sample	Obs_Value	Prediction
1	Prova 4 senza Carote 125oC 31-07-25	46.00	33.84
2	Prova 5 300g senza Carote 119oC 01-08-24	24.00	18.86
3	Industrial Trials- 6 Std no carrots and thermal sterilization cycle in std conditions cesta 5 autoclave 3	24.00	29.50
4	Pilot Trial- Sample C - Bolo Export Std Cycle- thermal stressed	64.00	68.89
5	Pilot Trial- Sample C1 - Bolo Export Std Cycle- thermal stressed	77.00	43.62

TO CONCLUDE

Handbooks/textbooks, review articles,
Expert knowledge exchange, certification

Laboratory analysis, technical
papers

Scientific publications, data bases,
statistical process control

Spatio-temporal illustration, interactive
tools, cloud-based applications, (AI),
ChatGPT

Information .

Data !

Knowledge !!

Wisdom
!!!(?)

...Reality is what we observe, what we measure and what we can tolerate...

...Is it the same as truth??

Classical predictive modelling

AI-assisted quantitative assessments



...to maintain balance, transparency needs to also enable fit-for-purpose efficiency

*“Challenges in the
holistic risk
assessment of the
21st century”*

Panagiotis N. Skandamis

Professor

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