

Understanding Product Risk & Appropriate Intervention

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NAMI
NORTH AMERICAN
MEAT INSTITUTE

FOUNDATION FOR
MEAT & POULTRY
RESEARCH EDUCATION

Evolution of Listeria Control (Maturity Model)

Awareness	Enlightenment	Preventive	Predictive
<p><i>Listeria monocytogenes</i> (<i>Lm</i>) is a microorganism and has growth requirements like all microorganisms</p>	<ul style="list-style-type: none"> - <i>Lm</i> requires <ul style="list-style-type: none"> • temperature and pH in a certain range, • adequate moisture, • nutrients. - It is inhibited by certain chemicals. - It is susceptible to physical stressors. - Need to know where your product fits. 	<p>Employ a change and/or intervention to product environment to retard growth of <i>Lm</i>.</p>	<p>Understand</p> <ul style="list-style-type: none"> • developments in control mechanisms • your product's interaction with means of intervention and fine tune and/or update periodically.

Outbreaks of Listeriosis in the United States (2015-2022) with Known Food Vehicles

Year	Food Vehicle	State	Cases	Perinatal cases	Deaths
2015	Soft cheese	10 states	30	21	3
2015	Ice cream	AZ,KS,OK,TX	10	6	3
2016	Frozen vegetables	4 states	10	9	3
2016	Raw milk	FL & CA	2	1	1
2016	Packaged salads	9 states	19	Unknown	1
2017	Soft raw milk cheese	4 states	8	3	2
2018	Deli hams & pork	6 states	8	8	1
2019	Unidentified	13	24	Unknown	2

Outbreaks of Listeriosis in the United States Cont.

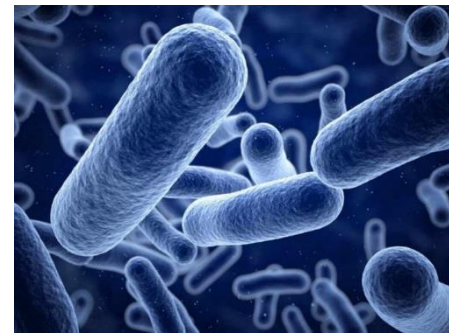
Year	Food Vehicle	State	Cases	Perinatal cases	Deaths
2019	Deli sliced meat & cheese	5 states	10	10	1
2020	Hard boiled eggs	5	8	Unknown	1
2020	Enoki mushrooms	17	36	Unknown	4
2021	Queso cheese	4	12	4	1
2021	Deli meats	4	12	Unknown	1
2021	Fully cooked chicken	2	3	Unknown	1
2022	Soft cheese	6	6	Unknown	0
2022	Ice cream	10	23	Unknown	1

Determination of Listeriosis Risk in Products

- Estimates are that undiagnosed Listeriosis or illnesses not linked to outbreaks far exceed number of identified outbreaks
- Could look at outbreak data in U.S. or worldwide
- Annual surveillance reports available online-CDC
- FDA/USDA/CDC quantitative risk assessment is a very comprehensive analysis of contributing factors



Risk Components



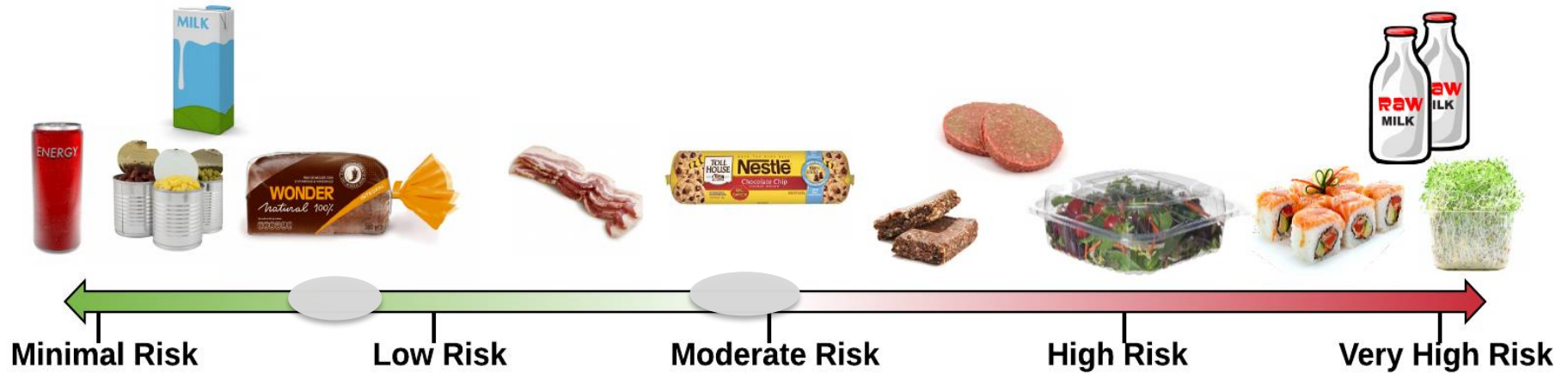
- Inherent rate of contamination
- Growth rate of *L. monocytogenes* on the product
- Typical storage length prior to consumption
- Likelihood of heat treatment prior to consumption
- Servings consumed annually
- Amount of product consumed per serving
- Likelihood product would be served to susceptible populations

Results of FDA/USDA Listeriosis Risk Assessment of RTE Foods - 2003



- **Risk Very High** – Deli meats, frankfurters not reheated
- **Risk High** – High fat and other dairy products, unpasteurized milk, pate & meat spreads, soft un-ripened cheeses, smoked seafood
- **Risk Moderate** – Deli salads, frankfurters reheated,, pasteurized fluid milk, fresh & un-ripened soft cheese
- **Risk Low** – Dry/semidry sausage, fruits & and vegetables, preserved fish, breads, pastries
- **Risk Very Low** – Cultured milk products, hard or processed cheese, ice cream and other frozen dairy

Microbiological **Safety** Risk Continuum: Producer Perspective



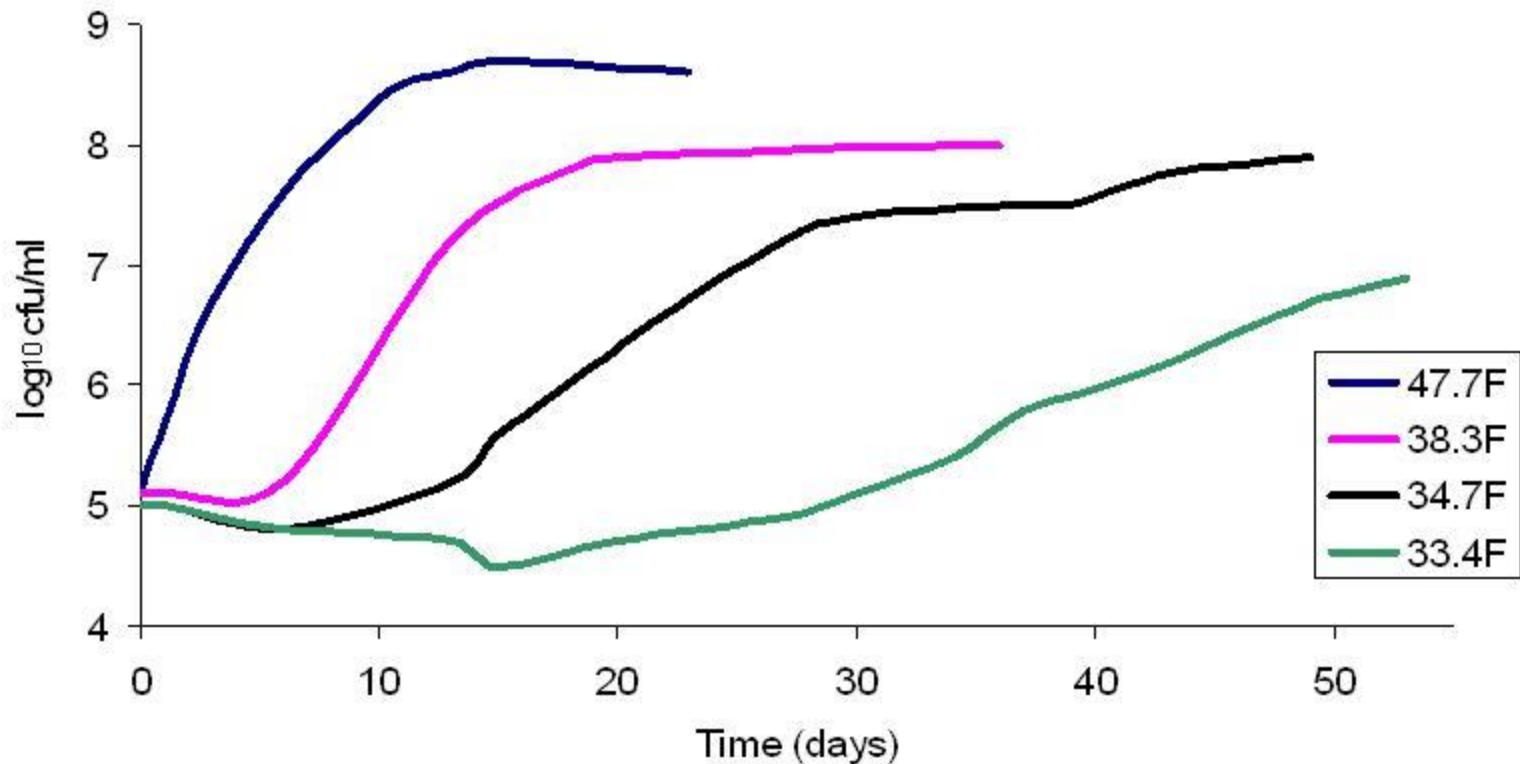
Risk Components We Can Influence

- Inherent rate of contamination
- Growth rate of *L. monocytogenes* on the product
- Typical length of storage of product prior to consumption
- Likelihood of heat treatment prior to consumption
- Servings per annum consumed
- Amount of product consumed per serving
- Likelihood product would be served to susceptible populations

Changing Risk Level of our Products

- Inherent Rate of Contamination
 - Remove *L. monocytogenes* from post-lethality / product-exposed environment
 - Provide additional lethality step after product is in package
- Growth rate of *L. monocytogenes* on product
 - Change inherent characteristics of the product, i.e., pH, a_w
 - Change storage conditions
 - Add an antimicrobial(s) to restrict growth

Changing Risk Level by Temperature



Growth of *Listeria monocytogenes* CFA 433 in chicken broth when incubated at 47.7, 38.3, 34.7, and 33.4°F. Adapted from S.J. Walker et al. (1990).

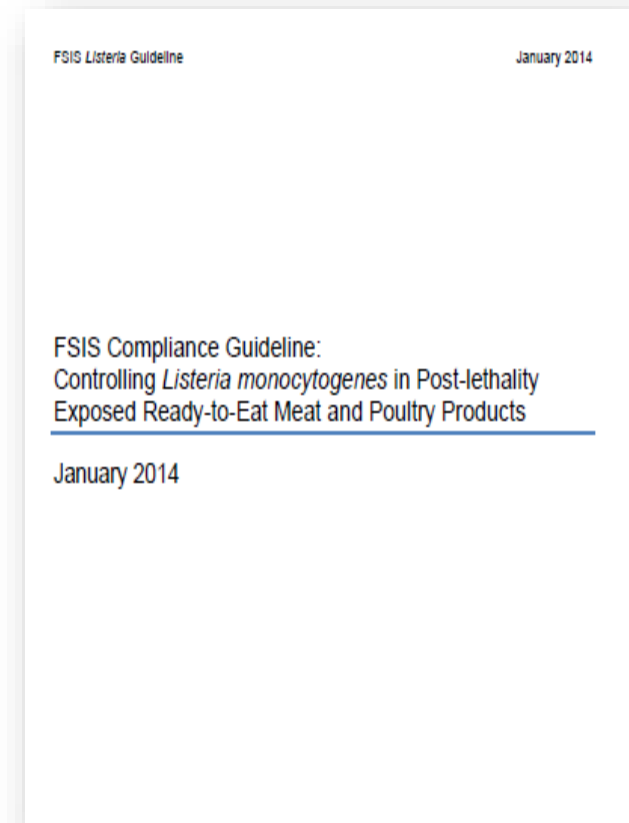
FSIS Compliance Guideline

- FSIS Compliance Guideline:
Controlling *Listeria monocytogenes* in Post-lethality Exposed Ready-to-Eat Meat and Poultry Products

- January 2014

https://www.fsis.usda.gov/sites/default/files/media_file/2021-02/Controlling-Lm-RTE-Guideline-2014.pdf

- January 2017 updated draft guidance document



FSIS Listeria Rule

- *L. monocytogenes* is a hazard that establishments producing post-lethality exposed RTE products must control through a HACCP plan or prevent through a Sanitation Standard Operating Procedure (SSOP) or a prerequisite program (9 CFR 430.4(a)).
- To maintain the sanitary conditions to meet this requirement, establishments must comply with one of three alternatives (9 CFR 430.4(b)).
- Alt. 1 & 2 are the most common
- Alt. 3 relies on sanitation alone

USDA/FSIS LISTERIA RULE

Must suppress or limit *L. monocytogenes* growth on the product

ALTERNATIVE 1

- Post lethality treatment (may be antimicrobial agent)
- AND**
- Antimicrobial process or agent

ALTERNATIVE 2

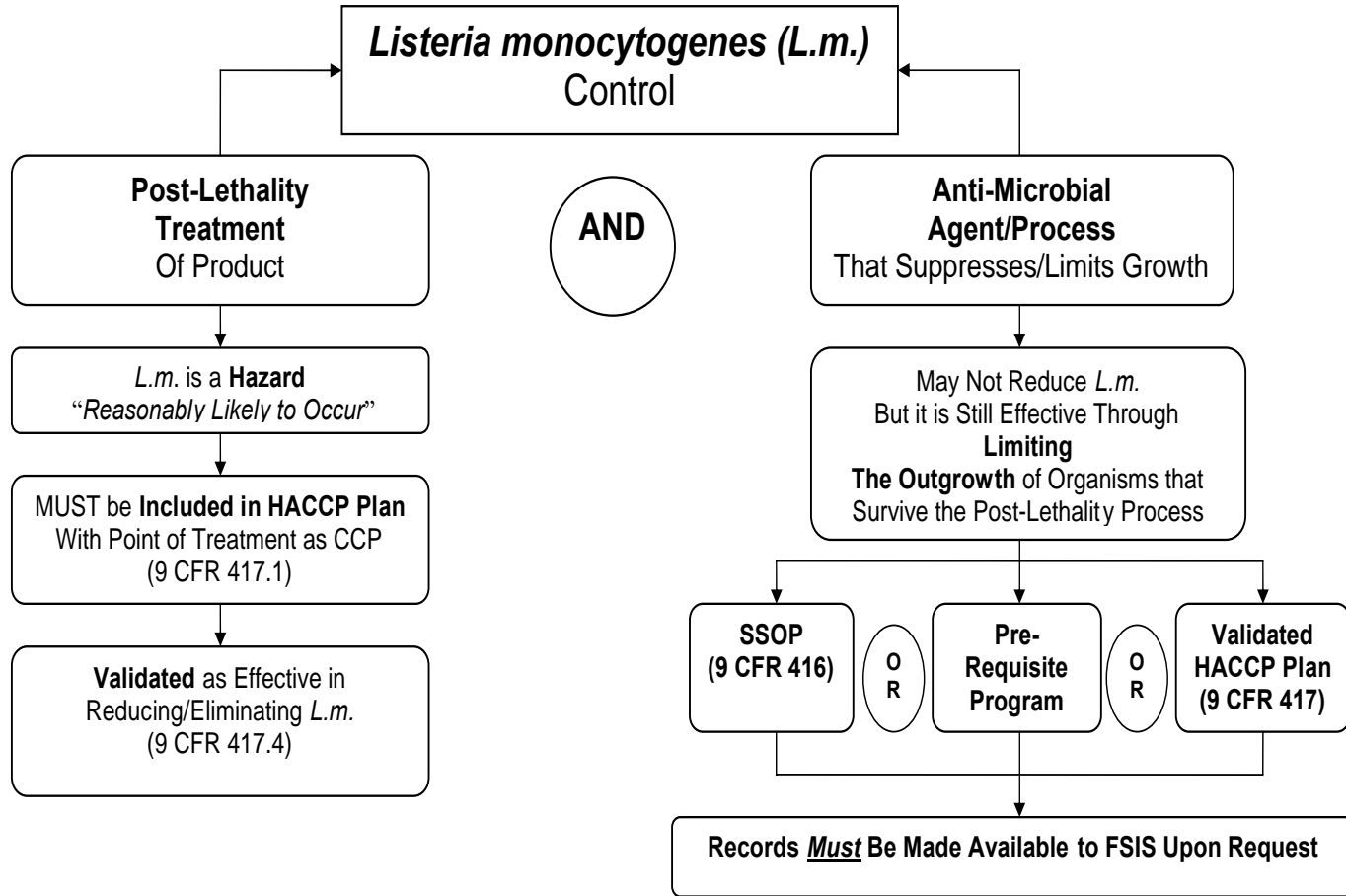
- Post lethality treatment (may be antimicrobial agent)
- OR**
- Antimicrobial process or agent

ALTERNATIVE 3

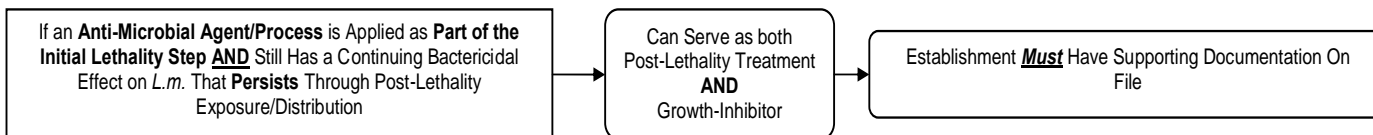
- Relies on sanitation alone to control *L. monocytogenes*
- No requirements for
 - Post lethality treatment
 - Antimicrobial process or agent

The post lethality treatment, antimicrobial agent or process must be included in either the HACCP Plan, the SSOP's, or other pre-requisite program and shown to be effective.

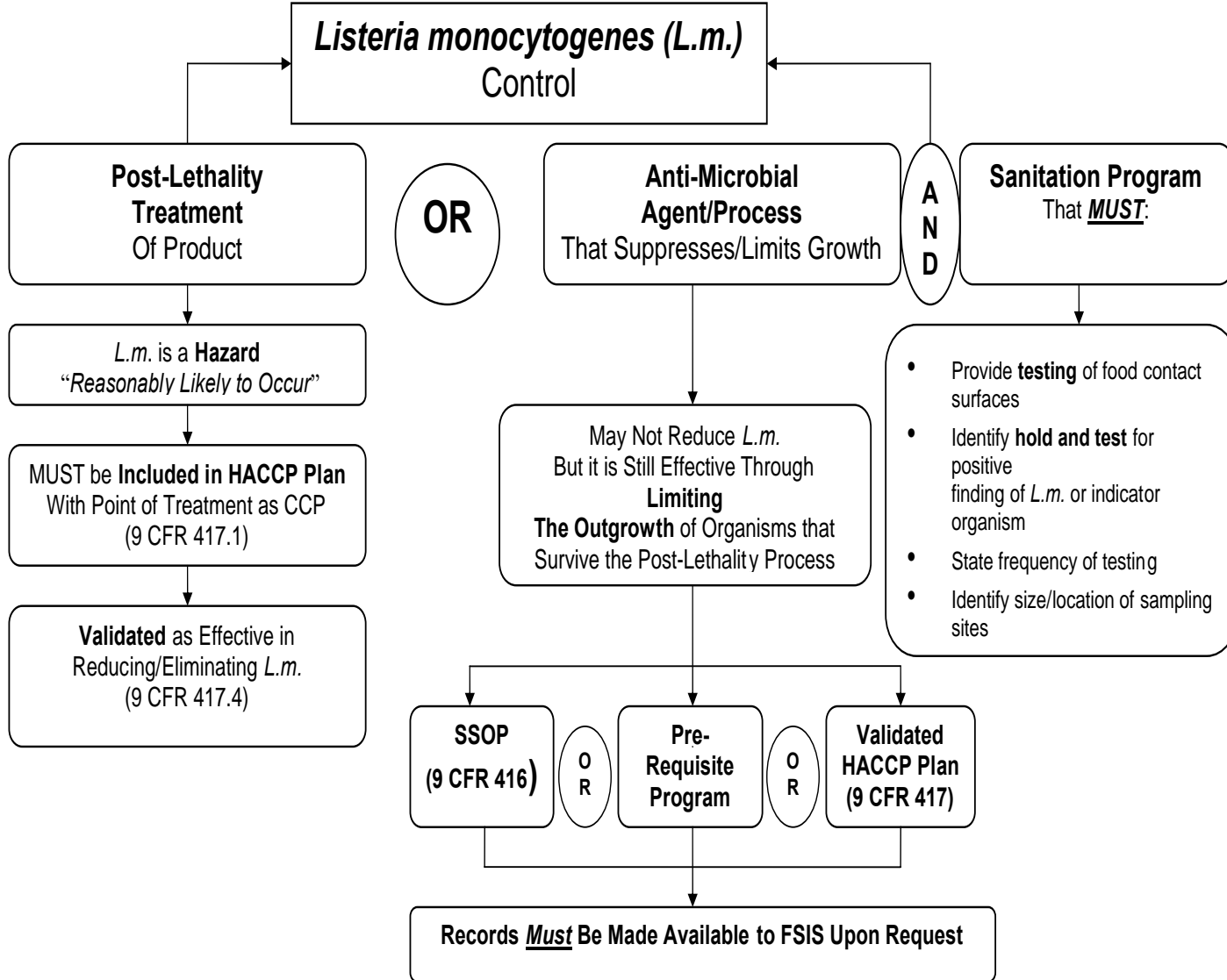
Alternative 1



Note:



Alternative 2

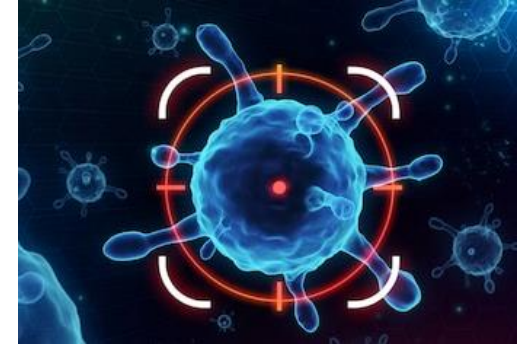


Alternative 3: Sanitation Only

- Relies on sanitation alone to control *L. monocytogenes*
- Not common or preferred
- No use of
 - Post lethality treatment
 - Antimicrobial process or agentto reduce, eliminate or control *L. monocytogenes*
- Specific requirements for
 - Non-deli, non-hot dog
 - Deli and hot dog producers

Lethality Treatments

(usually after packaging)



- Heat treatment – hot air impingement, hot water dip or spray, steam impingement
- High Pressure Processing
- Application of bactericidal agent (liquid)
- Application of bacteriophage
- Other high energy application – UV, pulsed light, pulsed electric field

“Clean Label” Food Technology

- **Post-Pasteurization** – RTE product heated in final package
- **High Pressure Processing** – RTE product in final package exposed to extreme hyperbaric conditions
 - ✓ 86,000 PSI for 2-5 minutes
- **Topical Antimicrobials** – processing aids sprayed on food surface during packaging
 - ✓ Lauric arginate, bacteriophage

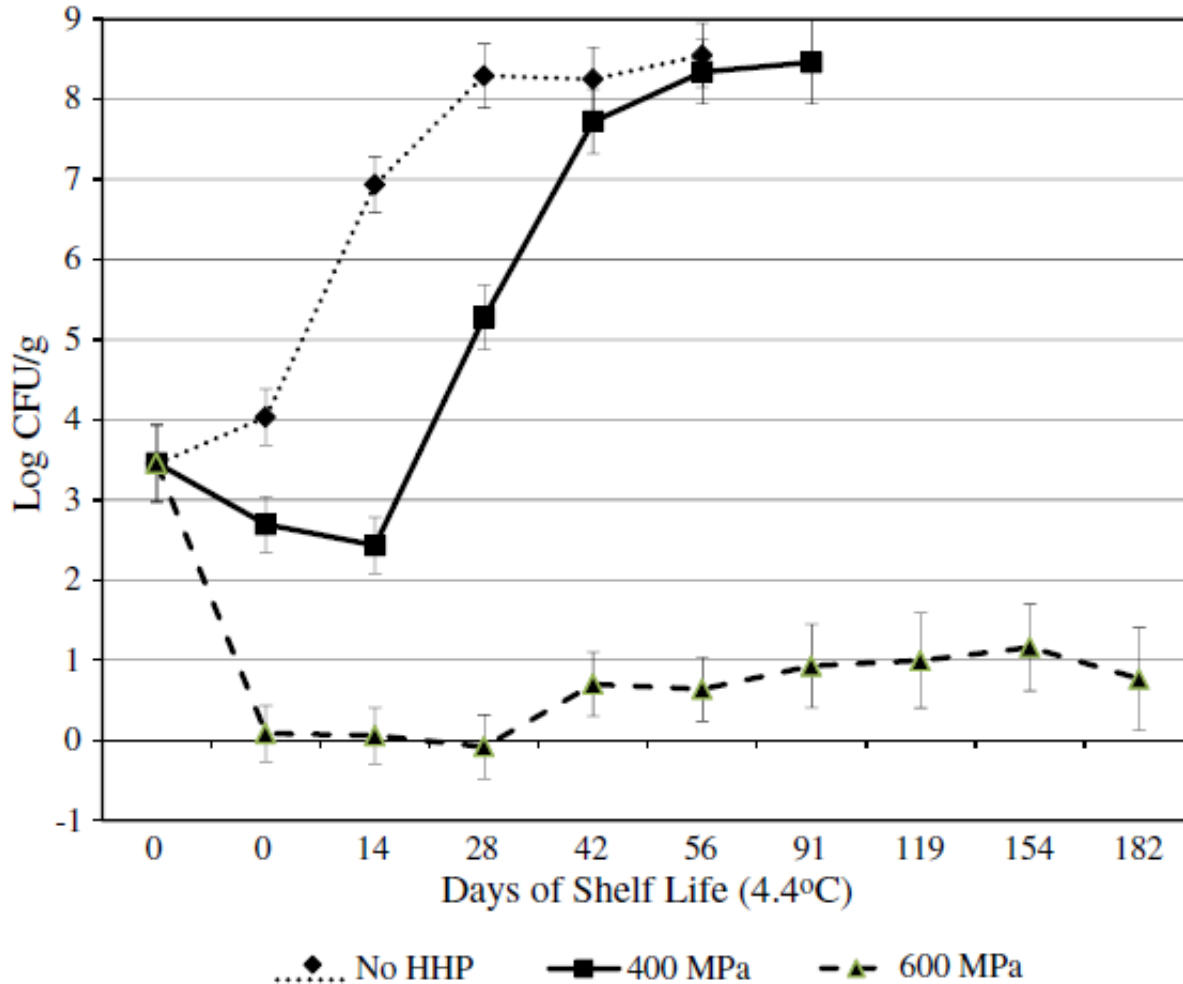


High Pressure Processing



- High pressure processing or high pressure pasteurization is very successful with most vegetative microbes.
- Many foods can be treated effectively with minimal deleterious affects.
- Cannot contain entrapped air or be in a rigid container.
- Considered clean label with few or no changes in nutritional value.
- In addition to being valid for eliminating *Listeria* and other pathogens, treatment often results in significant shelf-life extension.

Inactivation of *L. monocytogenes* on ham slices by HPP (400-600 MPa)

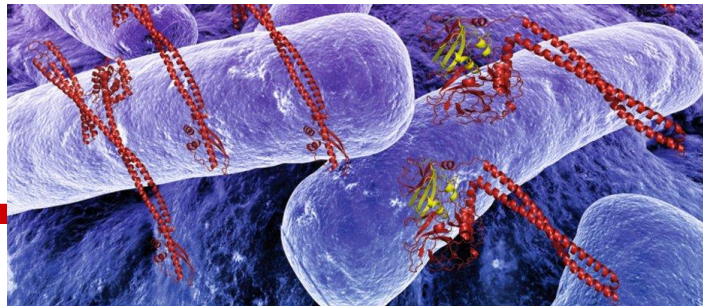


Means of *L. monocytogenes* counts by HPP treatment after inoculation of ham slices with a 5-strain cocktail of *L. monocytogenes* at 103 CFU/g followed by no HPP, 400 MPa HPP treatment for 3 min, and 600 MPa HPP treatment for 3 min (detection limit=1.0 log₁₀ CFU/g).

Myers et al. 2013. Effects of HPP and varying concentrations of sodium nitrite from traditional and vegetable-based sources on the growth of *L. monocytogenes* on RTE sliced ham.

Bacteriostatic Treatments – Surface or Internal (inhibit growth)

- Lower water activity to 0.92 or less
- Organic acids
- Natural or liquid smoke
- Spices, natural resins, oleoresins
- Preservatives
- Bacteriocins - nisin or pediocin
- Small peptides or amino-acid based surfactants
- Change storage conditions, e.g. freezing



INHIBITORS

ANTIMICROBIALS
THAT PRESERVE
PRODUCTS BY
INHIBITING
FOODBORNE
PATHOGENS
AND SPOILAGE
MICROBES

• Traditional Inhibitors

- Sodium or potassium lactate + sodium diacetate or acetate
- Sodium propionate

• “Natural” Inhibitors for Clean Label

– Vinegar

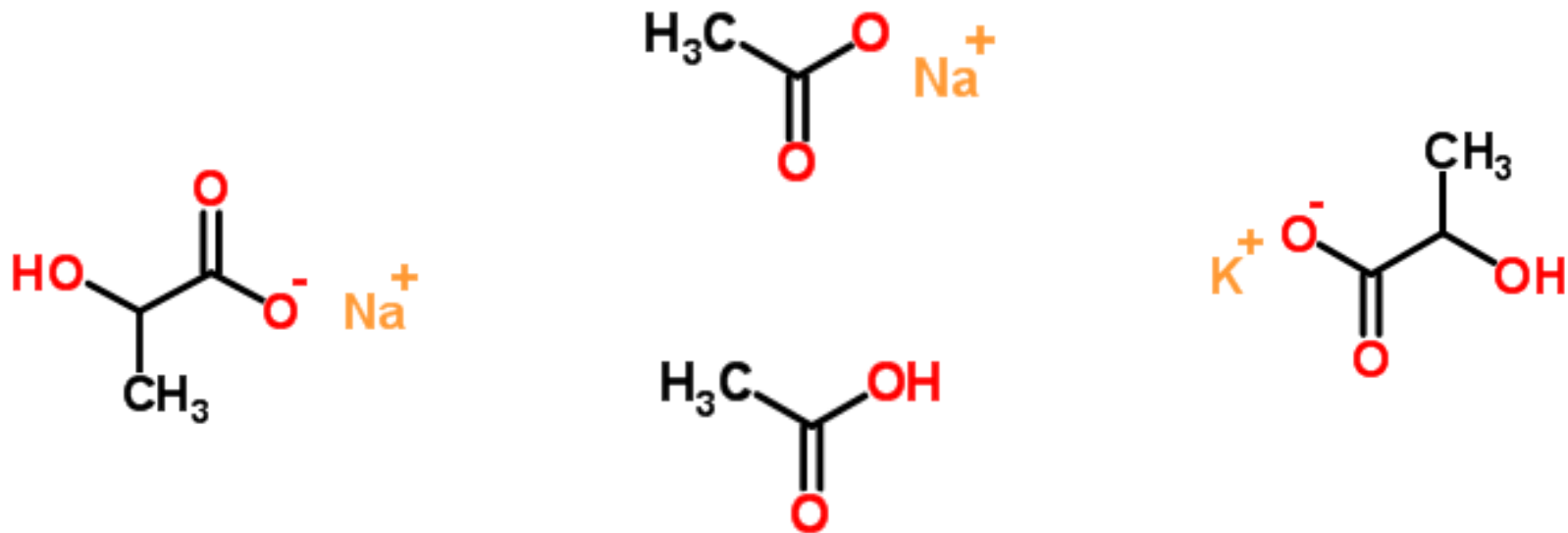


– Fruit extracts + vinegar



– Cultured sugar (bacterial fermentates)





ORGANIC SALTS

Most RTE Products



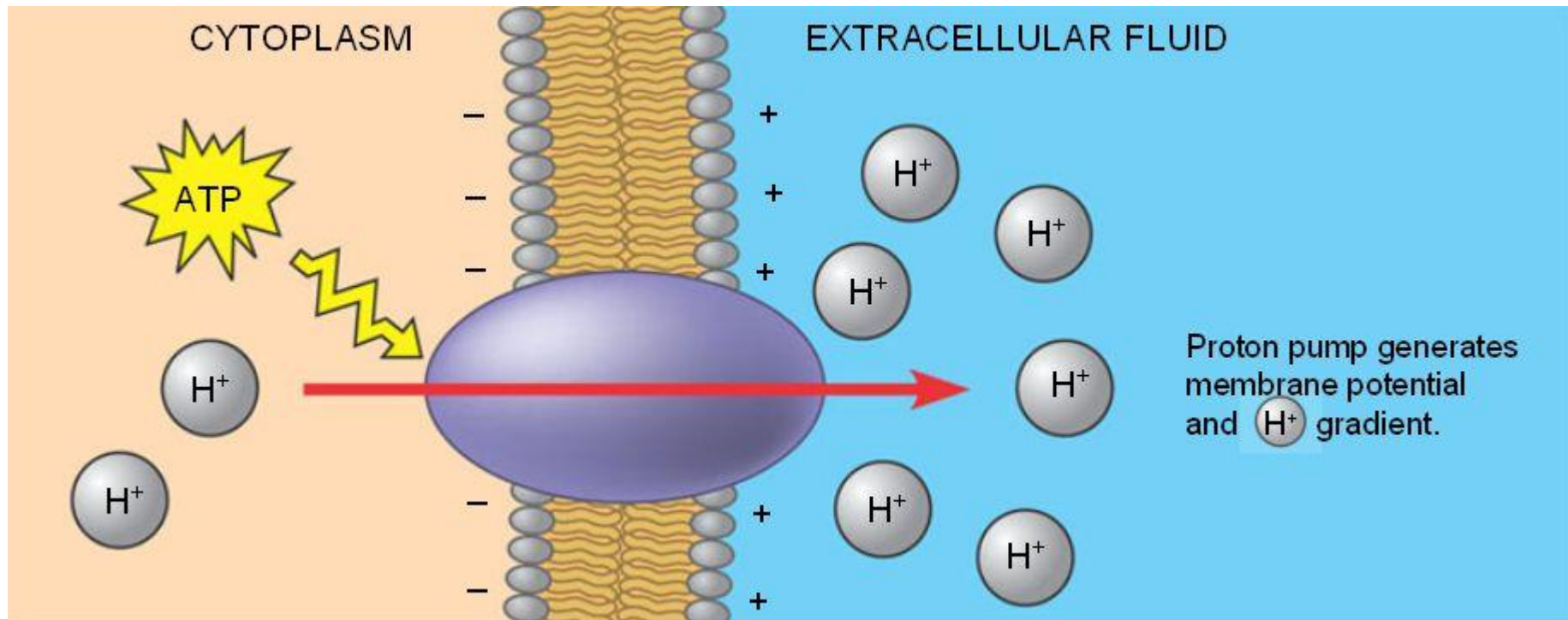
Mechanisms of Action

- Lowering of a_w (e.g. from 0.985 to 0.970)
 - ❑ Reduces amount of unbound water that is biologically available
 - Sodium acetate or sodium diacetate
 - Sodium lactate
 - Potassium sorbate or potassium benzoate
 - Sodium citrate
 - ❑ Extending the lag phase before growth



Mechanism of Action

- Weak lipophilic acids (e.g., lactic acid) pass across the cell membrane in undissociated form, dissociate within the cell and acidify the cell interior

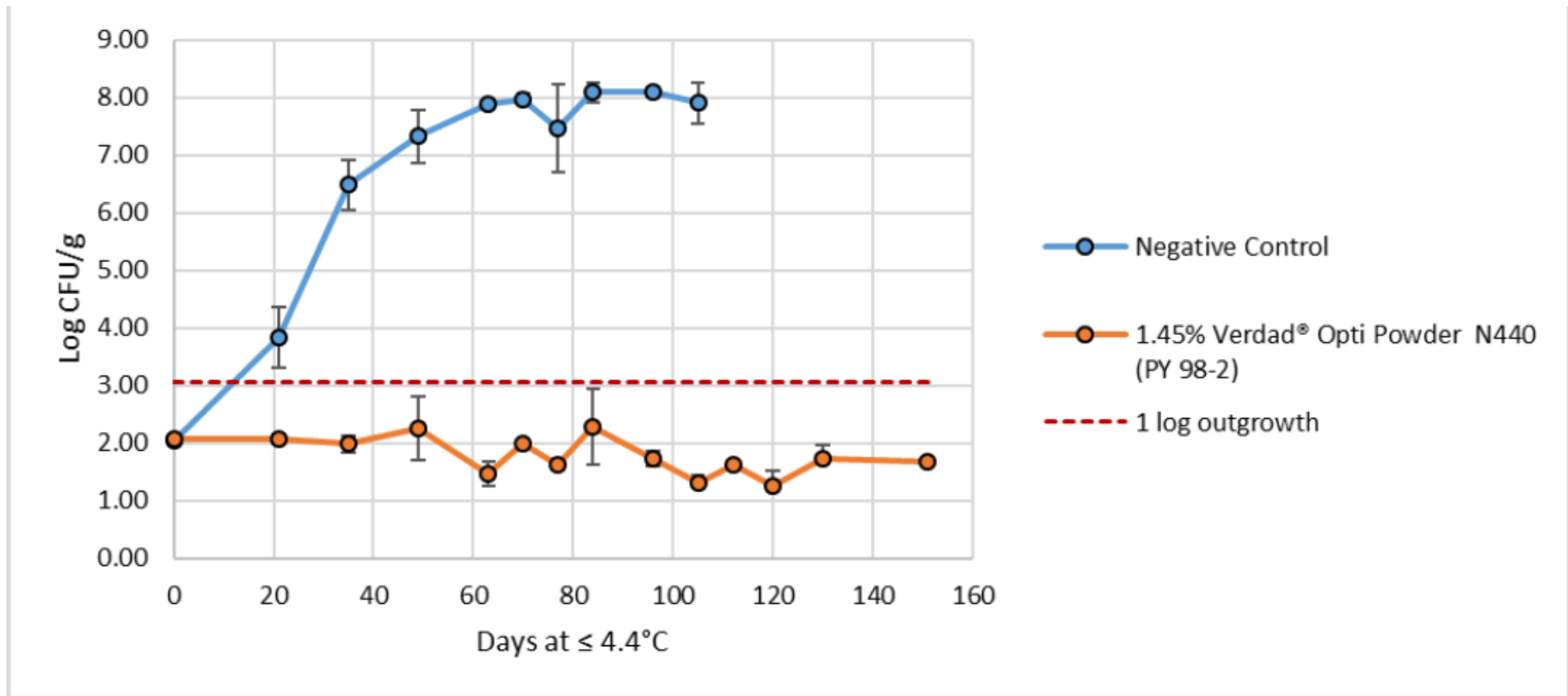


VINEGAR



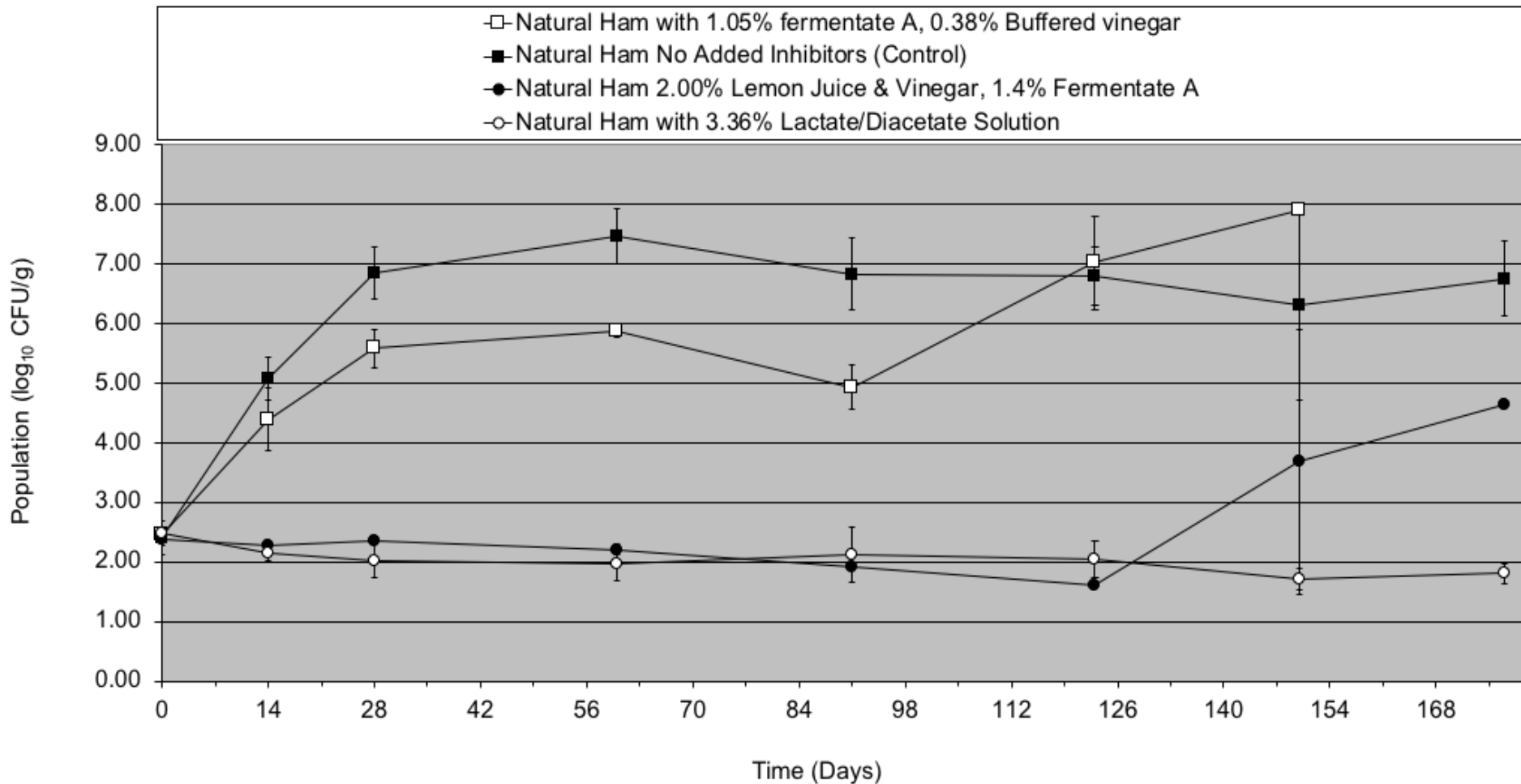
For “Clean Label” Products

Behavior of *L. monocytogenes* on Ham Slices as Affected by Vinegar Based Antimicrobial During Vacuum-Packaged Storage at 4.4°C (40°F)



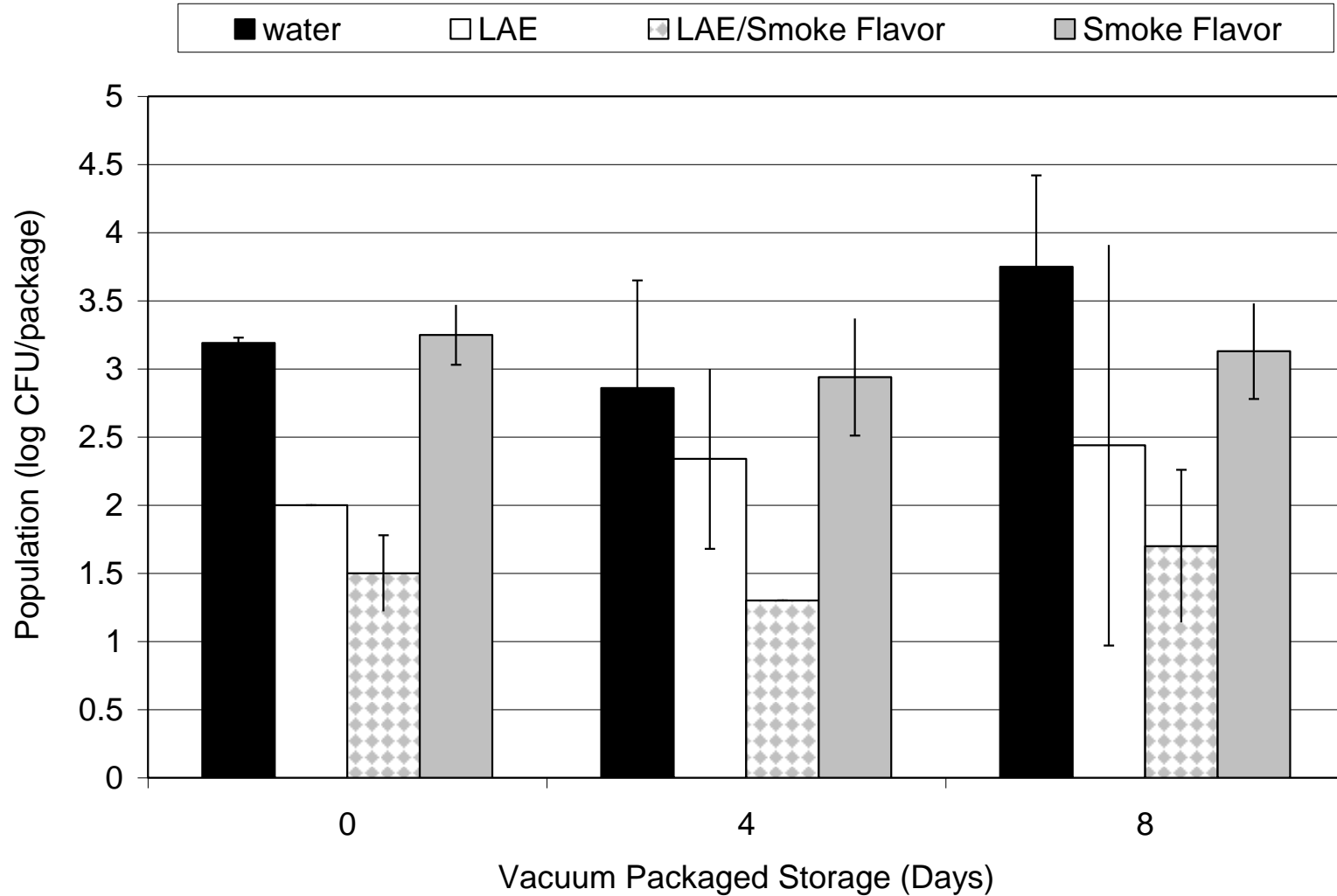
Spiral-sliced Honey Baked ham samples (50 g per package) were inoculated with a 5-strain *L. monocytogenes* cocktail obtained from Dr. Kathy Glass's lab at the University of Wisconsin-Madison at ca. 2.0 log₁₀ CFU/g. Inoculated samples were placed in bags that vacuum packed and stored at or below 4.4°C, and sampled over time by diluting with buffer, stomaching, and plating on MOX agar plates. Presumptive *L. monocytogenes* colonies are reported as CFU/g.

Behavior of *L. monocytogenes* on Natural Ham Slices as Affected by Antimicrobial Agents During Vacuum-Packaged Storage at 4.4°C (40°F)



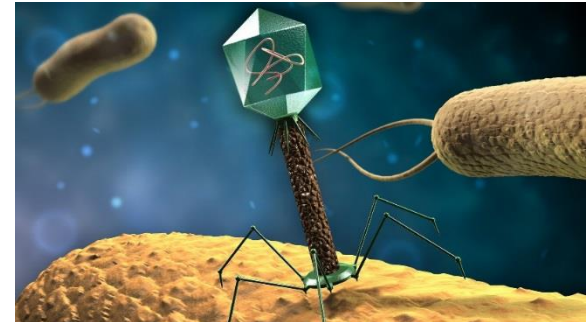
Sliced ham samples (99g) were inoculated with *L. monocytogenes* (Scott A, ATCC 19111, ATCC 19115, and two plant environmental isolates) at ca. 2.5 log CFU/g. Means and standard deviation bars represent duplicate plates from duplicate samples at each time.

Inactivation of *L. monocytogenes* on hot dogs by 2ml of 5,000 ppm Lauric arginate (LAE) Solution (~28 mg/kg of meat)



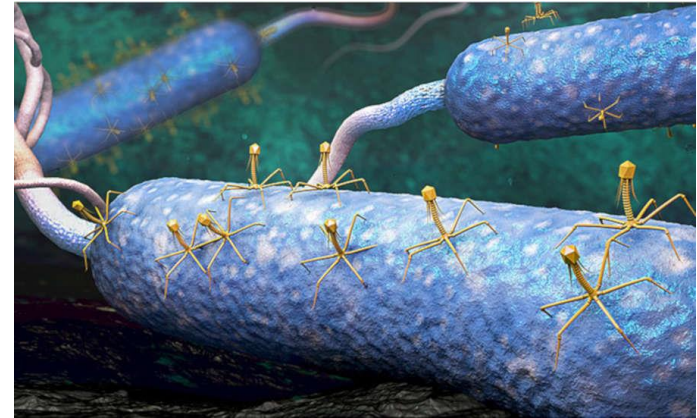
Bacteriophages

- Virus - infects and replicates within bacteria
- Natural parasites of bacteria
- Targeted control of foodborne pathogens
- Environmentally friendly
 - Composed of protein and DNA/RNA
- Unlike antibiotics, may evolve to combat bacterial resistance
- Cost effective, ubiquitous in nature and cheap to produce

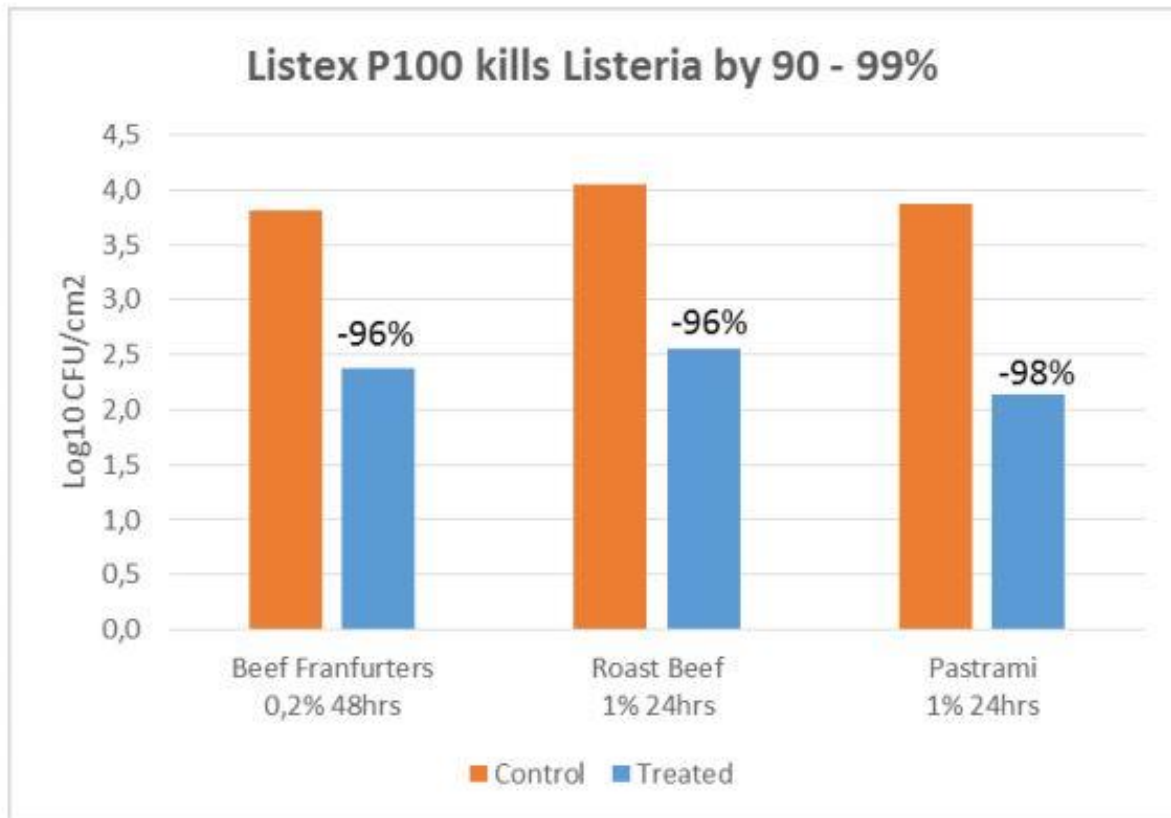


Bacteriophages in Food

- FDA/USDA GRAS status
- Very high specificity
- No organoleptic effect
- Easy application
 - Spray onto product prior to packaging
 - Spray into package
 - Spray onto slicer blade/dicer blades
 - Dipping/immersion into a phage solution
 - In combination with other antimicrobial interventions
- No labelling requirement - clean
 - Processing aid



PhageGuard-Listex



- 1 log CFU/cm² reduction within minutes
- 1.5-2 log CFU/ cm² reduction after 24hrs

Effect of PhageGuard Listex after 48hrs on Frankfurters using a 0.2% solution, and after 24hrs on Roast Beef and Pastrami using a 1% solution of Listex sprayed at 10µml/cm².

<https://phageguard.com/wp-content/uploads/2019/10/PhageGuard-Listex-Application-Data-Sheet-RTE-Meat-FINAL.pdf>

Choosing Intervention Level & Type

- Inherent product risk – Very high, high, med., low
 - Complexity of process and handling while product is exposed
 - Historical level of environmental contamination
- Product characteristics and effects of intervention(s)
- Production volumes and rates
- Price sensitivity of product
- Customer requirements
- History of recalls or outbreaks



Choosing Intervention Method

Example 1 – Sliced Luncheon Meat

- Very high-risk evaluation by FDA/USDA
- Individual slices in package with all surfaces exposed
- High production rates
- Product sensitive to purge, texture, and color changes
- Low profit margins
- Would like Alternative 1, but with current technology choose Alternative 2 with antimicrobial in formulation



Choosing Intervention Method

Example 2 – Breakfast Links

- Estimated moderate risk due to likelihood of reheating, minimal handling post lethality
- High production rates
- Multiple pieces per package
- Moderate margins
- No history of being associated to outbreaks
- Choose Alternative 2, change storage conditions (freezing)



Establishing Efficacy and Critical Limits of Treatments (Validation)

- Using intrinsic characteristics of *L. monocytogenes*
 - High acid concentration – pH < 4.4
 - Low water activity – $a_w < 0.92$
 - Temperature (control growth) – Product held < 30°F
 - Temperature (lethality treat.) – $D_{145^\circ\text{F}}$ value of 1.2 min
- Computer simulations or estimations
- Published studies
 - Lethality treatments
 - Growth preventive agents
- Supplier studies
- Internal challenge studies



Must be done before first production and whenever significant product/process changes occur

Predictive Microbial Models

ComBase

Home About Donate data FAQ Contact us

ComBase Predictor

The **ComBase Browser** enables you to search thousands of microbial growth and survival curves that have been collated in research establishments and from publications

The **ComBase Predictive Models** are a collection of software tools based on ComBase data to predict the growth or inactivation of microorganisms

Login/Register

58,500 + records | 70,000 + users

Supported by



A Web Resource for Quantitative and Predictive Food Microbiology

It includes:

- ✓ A systematically formatted database of quantified microbial responses to the food environment with more than 60,000 records
- ✓ ComBase Predictor and Food Models – to predict the growth and inactivation of microorganisms in food

It can be used for:

Information on the status of food safety research



New updates

Click here to see the new updates to data and features

New online models

- Lactic acid bacteria in cooked sausage emulsions
The model predicts the growth of *lactic acid bacteria* in cooked meat emulsions vacuum-packaged in low oxygen atmospheres.

Predictive Microbial Models

[Static | Dynamic]

Listeria monocytogenes/innocua

Init. level: 3
Phys.state: 2.1e-2
Temp (°C): 10
pH: 6.50
Aw | NaCl (%): 0.997

0 7
0 1
1 40
4.4 7.5
0.934 1

Max.rate (log.conc/h) 0.053
MPD (log CFU/g) 8.52
Dbl. time(Hours) 5.678
Lag time (Hours) 31.96

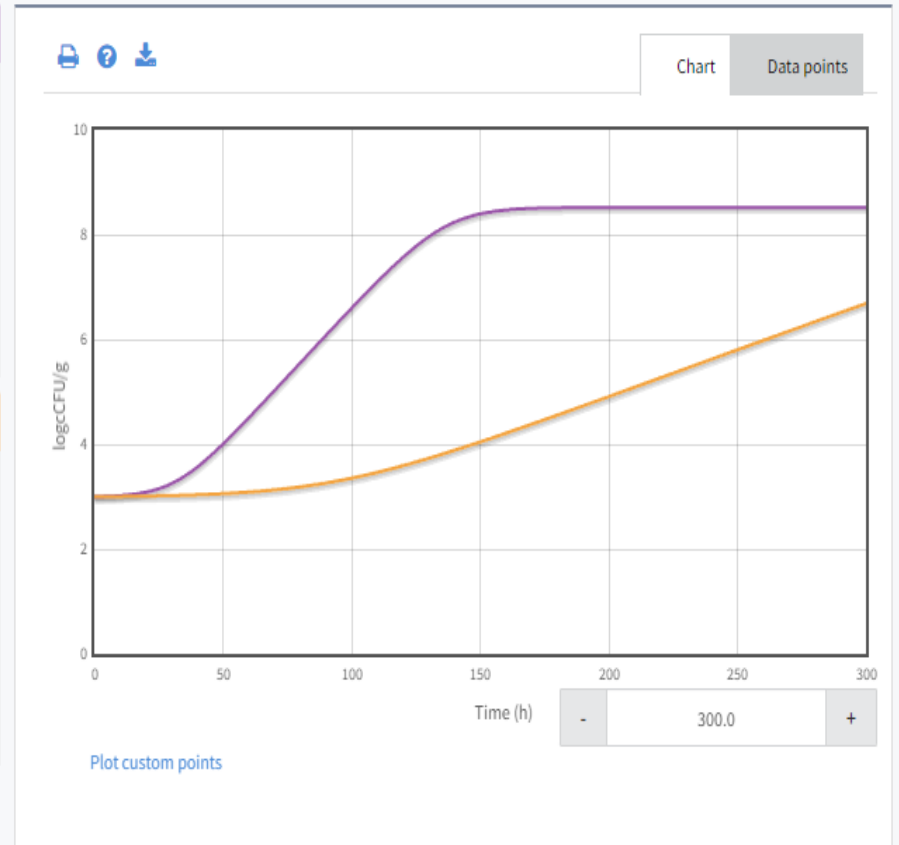
Listeria monocytogenes/innocua

Init. level: 3
Phys.state: 2.1e-2
Temp (°C): 4.4
pH: 6.50
Aw | NaCl (%): 0.997

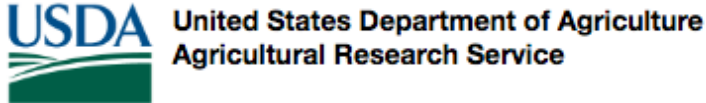
0 7
0 1
1 40
4.4 7.5
0.934 1

Max.rate (log.conc/h) 0.018
MPD (log CFU/g) 8.52
Dbl. time(Hours) 16.743
Lag time (Hours) 94.1

[Add prediction]



Predictive Microbial Models



Pathogen Modeling Program (PMP) Online

[PMP Home](#)

[PMP Online](#)

[About PMP](#)

[Tutorial](#)

[Frequently Asked Questions](#)

[Reference Material](#)

[Project Scientists](#)

You are here: [PMP Home](#) / PMP Online

SELECT A PATHOGEN MODEL ▶

The models are based on extensive experimental data of microbial behavior in liquid microbiological media and food.

There can be no guarantee that predicted values will match those that would occur in any specific food system. Before the models could be used in such a manner, the user would have to validate the models for each specific food of interest.

OK

ARS.USDA.gov

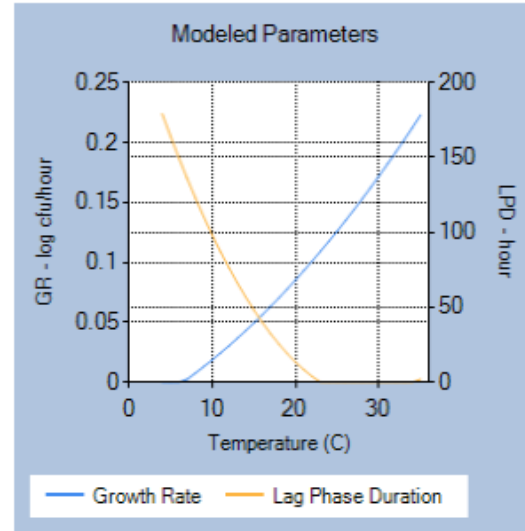
Predictive Microbial Models

Growth of *Listeria monocytogenes* in Ground Ham Containing Sodium Lactate and Sodium Diacetate

Input Conditions

Sodium Lactate
 Range: 1.0% - 4.2%

Sodium Diacetate
 Range: 0.05% - 0.2%



MODELED PARAMETERS

Temp (C)	GR (log cfu/h)	LPD (h)
4.0	0.000	179.1
5.0	0.000	164.1
6.0	0.000	149.7
7.0	0.003	135.9
8.0	0.008	122.8
9.0	0.013	110.3

Corbion[®] *Listeria* Control Model



Corbion[®] *Listeria* Control Model

Food characteristics

Enter the characteristics of your finished cured meat product as specifically as possible. If you are unsure of a food parameter, please use the default value. You may also select a Corbion ingredient and enter an addition level.

Moisture %
 pH
 NaCl %
 Sodium nitrite ppm (on total formulation)

Storage conditions

Temperature °F

Corbion Solution

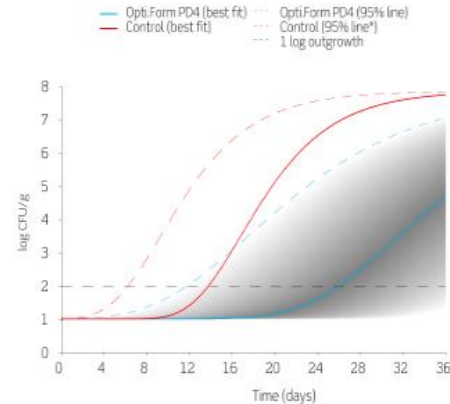
%

Microorganism data

Initial level log CFU/g
 Maximum allowed level log CFU/g

Reset to defaults

Listeria growth in chicken



About this graph

Time to 1 log outgrowth in days

	Control	With Opti.Form PD4
Best fit	14	26
95% line*	6	12

* The lines are based on specifically designed and validated *Listeria* challenge studies. According to these studies, 95% of growth is expected to be slower than the 95% line.

Calculate

Choosing a Validation Method



- Agency Ranking
 - Post Lethality: Internal challenge study > published challenge study > modeling program
 - Antimicrobial Process/Agent: Intrinsic *Listeria* characteristics = internal challenge shelf life study = modeling program specific to agent used > published study
- Customer Requirements
- Time Required for Results
- Characteristics of Product In Question
- Accuracy, Predictive Strength, and Repeatability of Method
- Cost: external challenge studies \geq internal challenge studies > supplier studies > published studies \geq computer modeling > intrinsic characteristics

Verification Activities



- Traditional HACCP Verification

- Calibration of process monitoring instruments
- Direct observations of monitoring activities and corrective actions
- Review of records associated with HACCP plan such as CCPs, calibration activities, etc.

- Periodic Product Testing

Note: If antimicrobial agent or process is used to qualify for Alternative 2 status, an environmental testing program must be in place (USDA).

Maturity Model

Stage 2 Awareness	Stage 3 Enlightenment	Stage 4 Preventive	Stage 5 Predictive
<p>Environmental sampling infrequent and after sanitizing.</p> <p><i>Listeria</i> positives result in extraordinary cleaning.</p> <p>Different attire for raw versus packaging employees.</p> <p>Follow equip. Manufacturers guide for sanitation.</p> <p>Understand growth factors for <i>Listeria</i>.</p>	<p>Environmental sampling weekly and during operations.</p> <p><i>Listeria</i> positives - examine records, do investigative sampling.</p> <p>Physical separation of raw and RTE welfare and production areas.</p> <p>Understand materials and design of equipment and select.</p> <p>Where possible reformulate for intrinsic factors.</p>	<p>Environmental sampling weekly and rotated throughout.</p> <p><i>Listeria</i> positives - permanent change to cleaning methods, frequency, or equip. Separation of personnel, areas, support staff, equip.</p> <p>Work with equip. suppliers in basic design and materials.</p> <p>Employ supplier's recommendations for use of antimicrobial.</p>	<p>Environmental sampling as before plus after disassembly. <i>Listeria</i> positives – total evaluation of circumstances.</p> <p>Understand movement of all personnel/equipment in RTE and control. Help design equipment and visit during construction.</p> <p>Work with supplier for tailored, experimentally supported antimicrobial.</p>



**LISTERIA
WINS GOLD!**

Event: Cellular Division

*** **

600 bacteria to win

LISTERIA: 602

E. COLI: 157

SALMONELLA: 83



BACTERIA REFRIGERATOR OLYMPICS 2018

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FOOD SAFETY REMINDER: *Listeria* bacteria can multiply even in refrigerated foods. Mark open and leftover food in the refrigerator with a use-by date no later than seven days after it was opened or prepared. Discard food that has passed its use-by date.



DON'T LET



WIN !