



Application of Natural Antimicrobials in Food: Food Industry User Perspective

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FAPC/IFT-OK Research Symposium - Keynote Presentation

Food & Agricultural Products Center

Oklahoma State University, Stillwater, OK 74078

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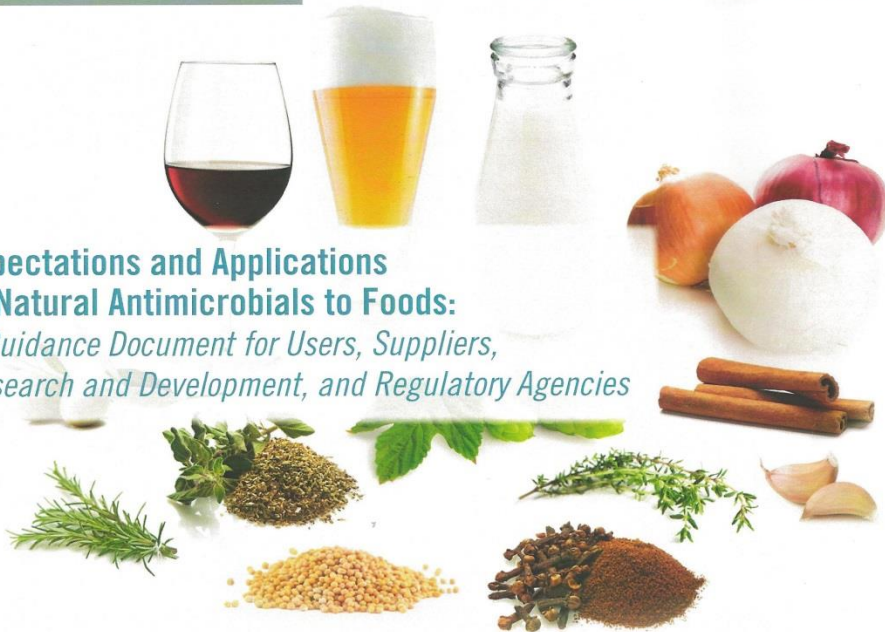
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Food you love





**Expectations and Applications
 of Natural Antimicrobials to Foods:**
*A Guidance Document for Users, Suppliers,
 Research and Development, and Regulatory Agencies*

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ABSTRACT

Efficient use of natural antimicrobials in food is predicated on the proper implementation of hurdle technology. These substances are meant to increase the robustness of existing food safety or quality assurance programs, not to correct or mask poor practices. The objective of this paper is to outline the important aspects of application of natural antimicrobials to foods, including selection of antimicrobial, determination of target microorganisms, efficacy testing against target microorganisms in vitro and in foods, and issues that must be addressed in the commercial application of the antimicrobial. Because natural antimicrobials are secondary hurdles, expectations of them must be realistic, and considerations should include other aspects, such as effect on sensory and quality attributes of the food, cost (and cost-in-use) of the antimicrobial, and regulatory and labeling considerations, in addition to efficacy against target microorganisms in the food matrix. The "idea-to-launch" business framework and governance is recommended for pairing of a potential antimicrobial with a complex food matrix, along with clearly defined objectives, inputs, outputs, and technical success criteria and business decision criteria. To help quantify the benefits of hurdles, including antimicrobials, we propose use of the "Food Protection Objective" (FPO), which is defined as the acceptable level of microbiological quality and/or safety at the moment of consumption or at the end of shelf life of a food.

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Food Industry User Perspective

Jairus David
 Larry Steenson
 & Michael Davidson

Food Protection Trends
 Vol. 33, No. 4, July-August
 2013, IAFP

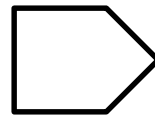


What are Antimicrobials?

❑ Naturally Occurring Ingredient or Extract that

→ Slows the Growth

→ Kills



Pathogens & spoilage organisms

Lactate/diacetate is an example of antimicrobial used in meats to control *Listeria monocytogenes*

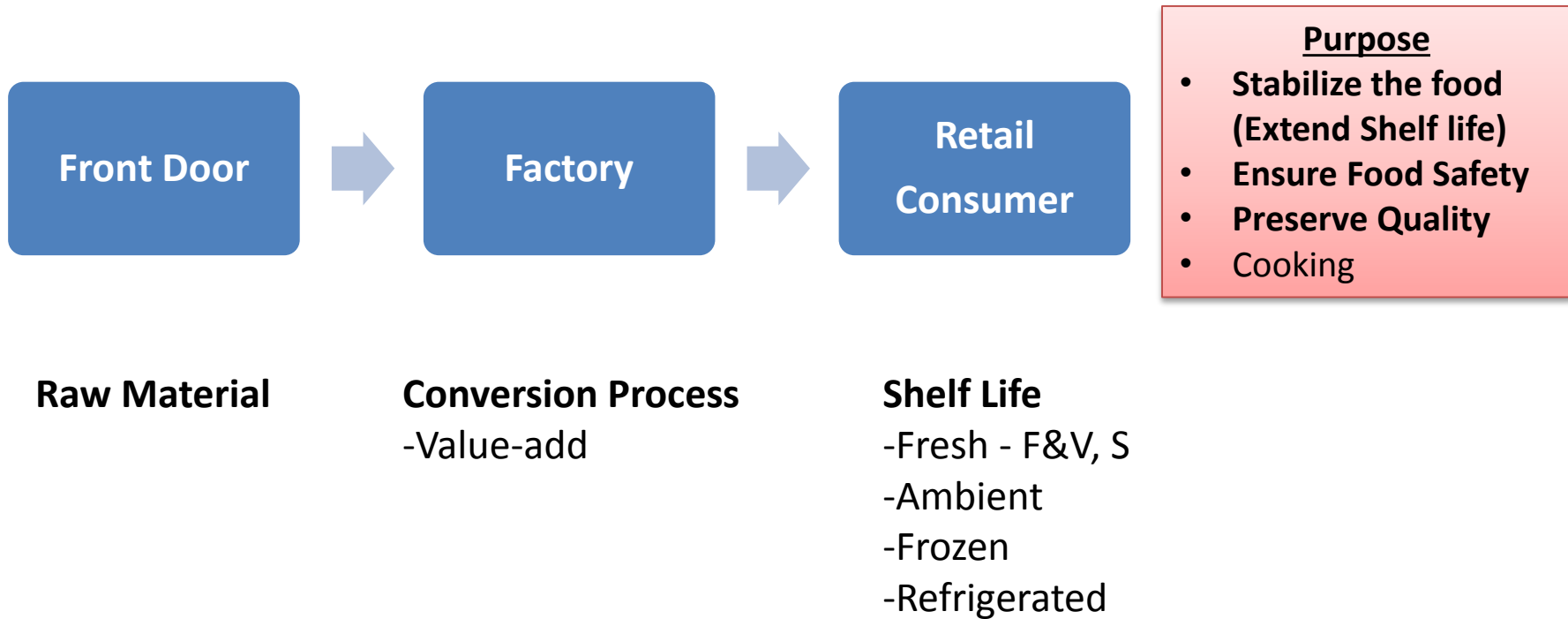
❑ Business Opportunity to use natural antimicrobials to

- Confirm & Extend Shelf Life
- Reduce Pathogen Risk
- Meet Consumer Demand for Minimally Processed RTE High Quality Foods
- Replace Synthetic Preservatives with Natural Clean Label Antimicrobials

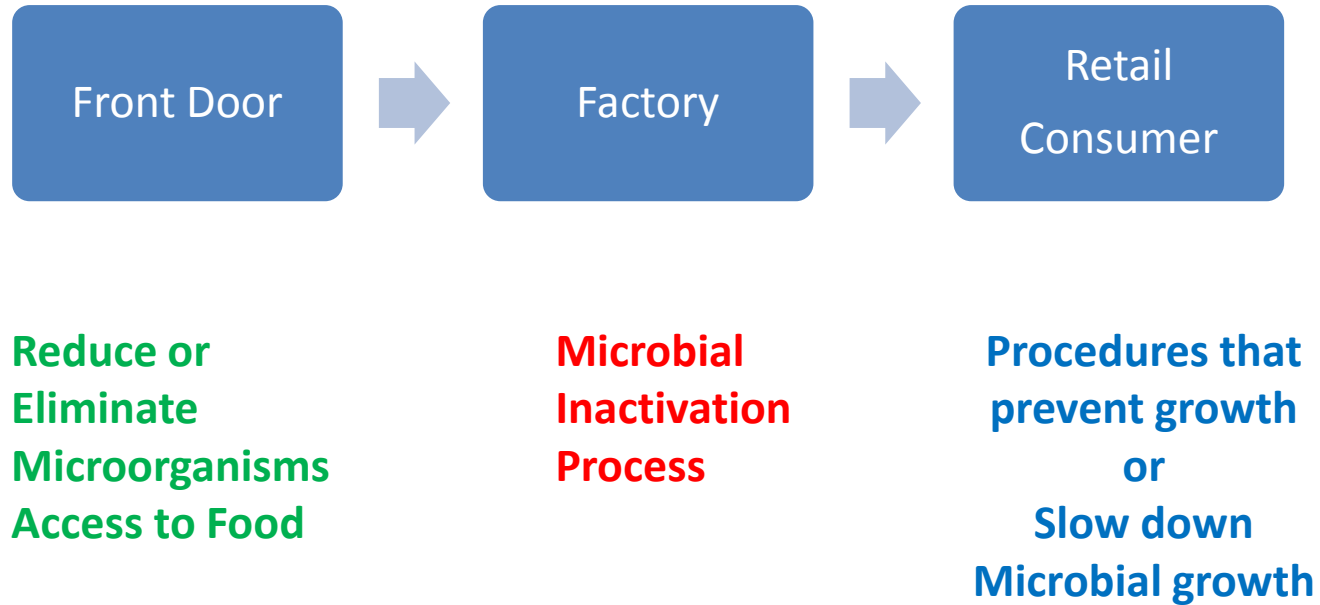
Manufacturing Process



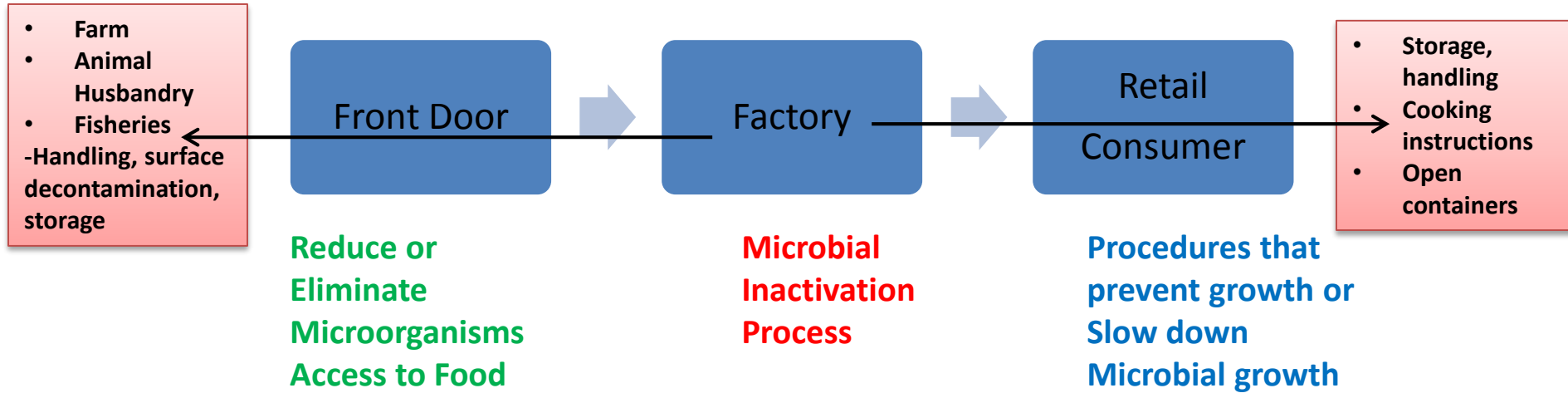
Food Preservation - PURPOSE



Food Preservation - DIMENSIONS



Food Preservation - WHY?



• 48 million cases of illness
• 128,000 hospitalizations
• 3000 deaths
CDC, 2011

• 31% of the available food supply at the retail and consumer levels was NOT consumed
• = \$162 billion. (Buzby et al, 2014, USDA)

Food Preservation - Details

- Farm
- Animal Husbandry
- Fisheries
- Handling, surface decontamination, storage

Materials



Factory



Retail Consumer

- Storage, handling
- Cooking instructions
- Open containers

- **Quality is Conserved**
- **Safety is Not Compromised**

incoming Ingredient Specifications

- RAC
- Meat
- Powder, Liquid, Solid

DRYING

IRRADIATION

HEAT PASTEURIZATION & CHILLED

FROZEN

Antimicrobials

LOW pH

LOWER aW

-Curing

-Drying

TANAKA MODEL

FERMENTATION

MAP

ADDITION OF ANTIMICROBIALS

ING TEMPERATURE

prevent

slow down

growth

ermination

NING

T-FILL-HOLD/CO

SEPTIC

LASH-18

HPP

Food Preservation Continuum – The Context for Use of Antimicrobials



Reduce or Eliminate Microorganisms Access to Food

Bio-Burden

Incoming Ingredient Specifications

- RAC
- Meat
- Powder, Liquid, Solid

DRYING
IRRADIATION
HEAT PASTEURIZATION & CHILLED
FROZEN

Microbial Inactivation Process

-Thermal
Sterilization
Pasteurization

- Non-thermal
- Combination

CANNING
HOT-FILL-HOLD/COOL
ASEPTIC
FLASH-18

Procedures that prevent growth or slow down Microbial growth

AMBIENT SHELF STABLE

LOWERING TEMPERATURE

- Frozen
- Refrigeration
- LOWERING pH
- LOWERING aW

- Curing
- Drying
- TANAKA MODEL
- FERMENTATION
- MAP

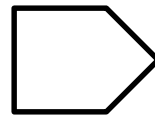
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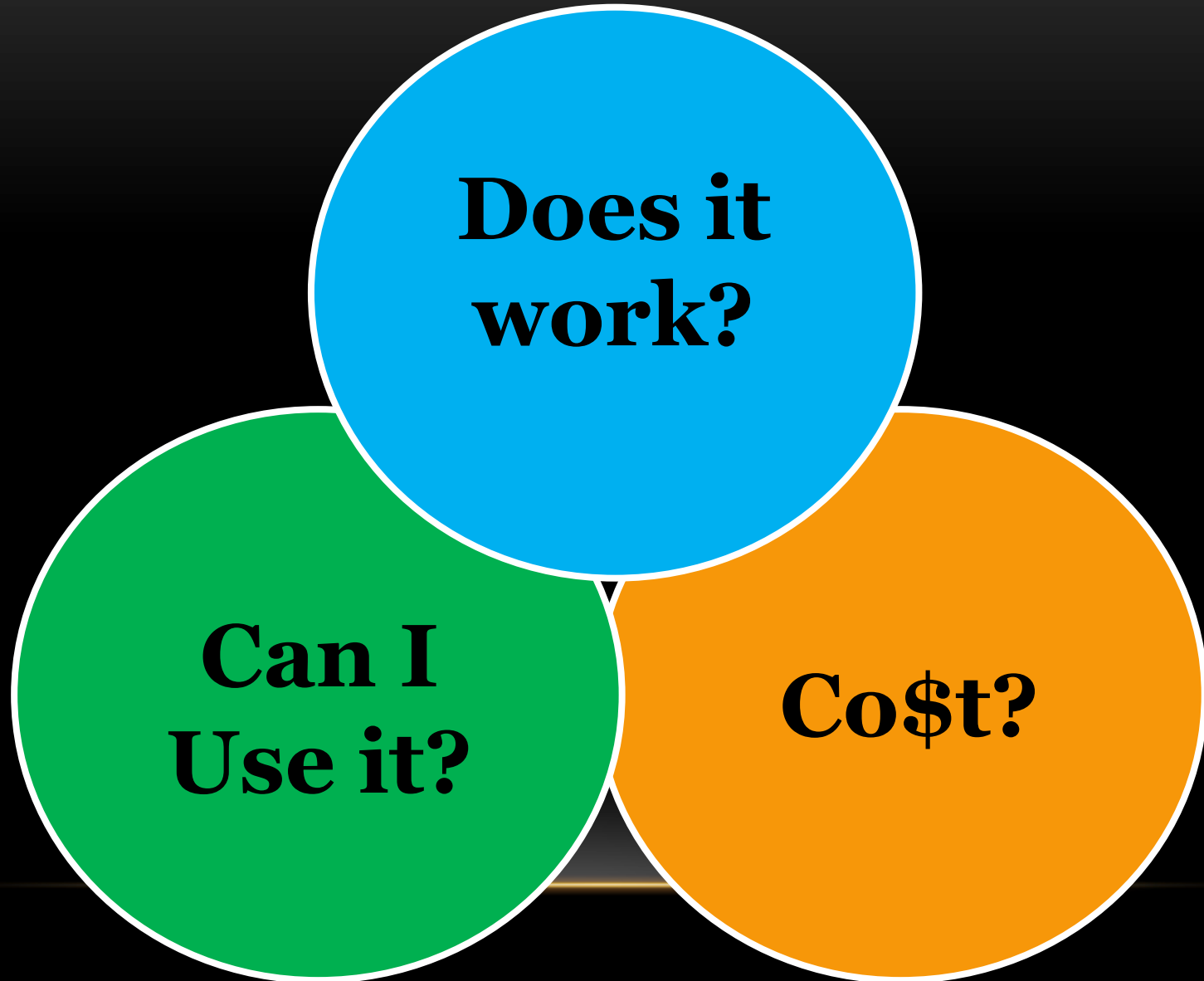
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Questions We Must Answer



Natural Antimicrobials

**Does it
work?**

- **Efficacy in Food**
- **Sensory Impact**
- **Regulatory limit**

≠

Can I Use it?

- **Clean Label**
- **GRAS/Tox Data**
- **Use-Patents**

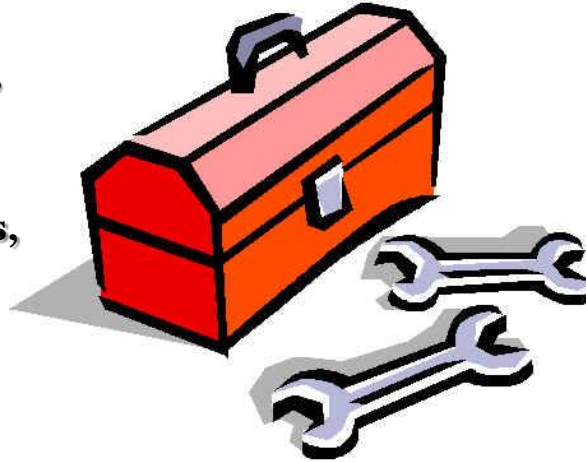
Cost-in-Use?

- **Upcharge/Case**
- **Capital Cost**

There is No Silver Bullet

FOOD

- ❑ **Savory-Sweet- Neutral Foods**
- ❑ pH & pKa Classification: <5.0, 5.8-6.2, >7
- ❑ **Partition Coefficient – Formulation, Fat, Proteins, Gums, CMC, TiO₂**
- ❑ aW
- ❑ Adding Natural Antimicrobial does not make the entire product natural

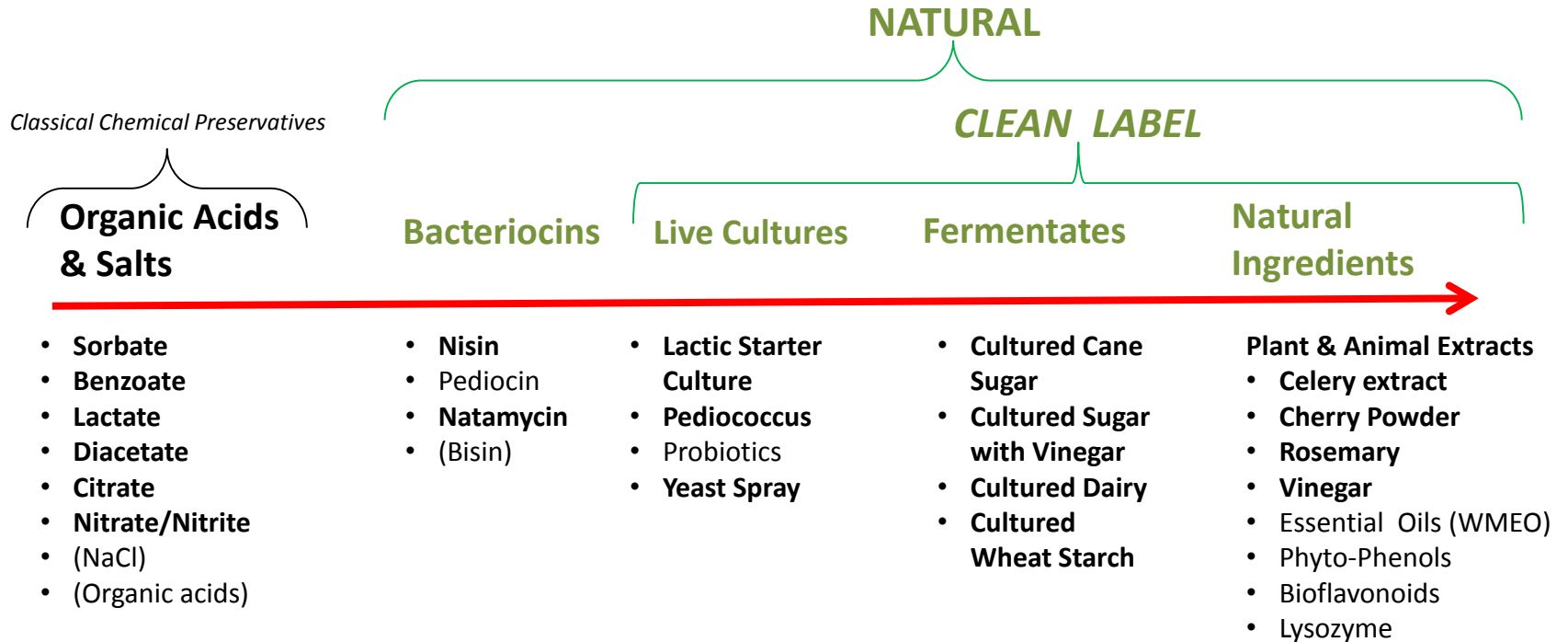


- 1. Natural ingredients**
Celery Juice Powder, Cherry Powder, Rosemary Extract, Plant Essential Oils
- 2. Fermentates**
Natural & Clean Label: Cultured Cane Sugar, Cultured Sugar with Vinegar, Cultured Wheat Starch
- 3. Bacteriocins**
Nisaplin; non yet for Gram negative bacteria
- 4. Live cultures**
Lactic acid bacteria, Pediococcus, and yeast spray

Antimicrobials

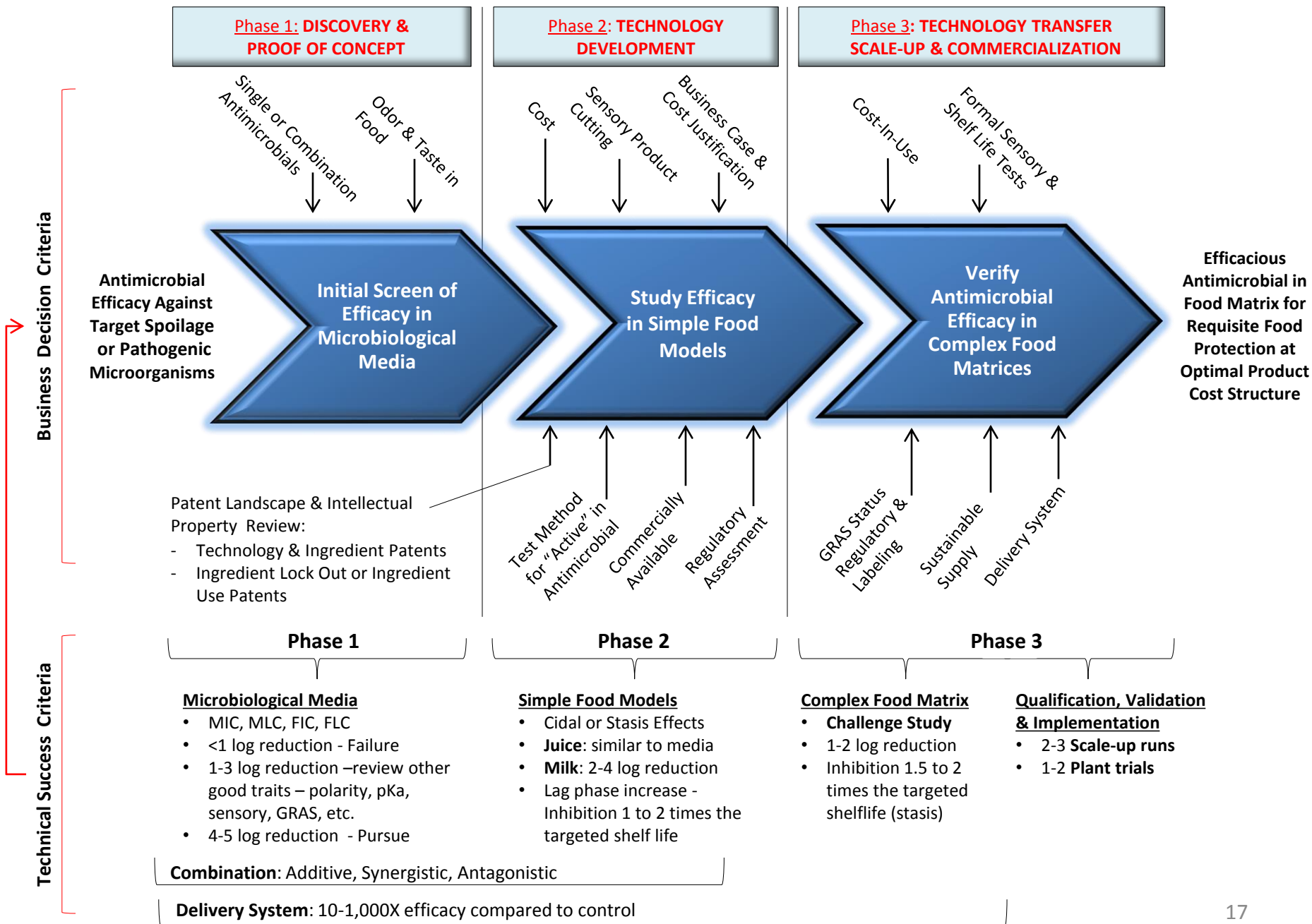
- **Narrow Spectrum**
- **Gram +ve Bacteria**
- Vegetative vs. Spores
- **Gram –ve**
- **Yeast & Mold**
- **Acid & Preservative (Sorbate) Resistant Yeasts**

Antimicrobial Toolbox



The “Hate List”

STAGE GATE for Achieving Due Diligence



Process for Achieving Due Diligence – PHASE 1



Phase 1: DISCOVERY & PROOF OF CONCEPT

Single or Combination Antimicrobials
 ↓
 Odor & Taste in Food (*Prospective*)

Antimicrobial Efficacy Against Target Spoilage or Pathogenic Microorganisms

Initial Screen of Efficacy in Microbiological Media

Efficacious Antimicrobial in Food Matrix for Requisite Food Protection at Optimal Product Cost Structure

Business Decision Criteria

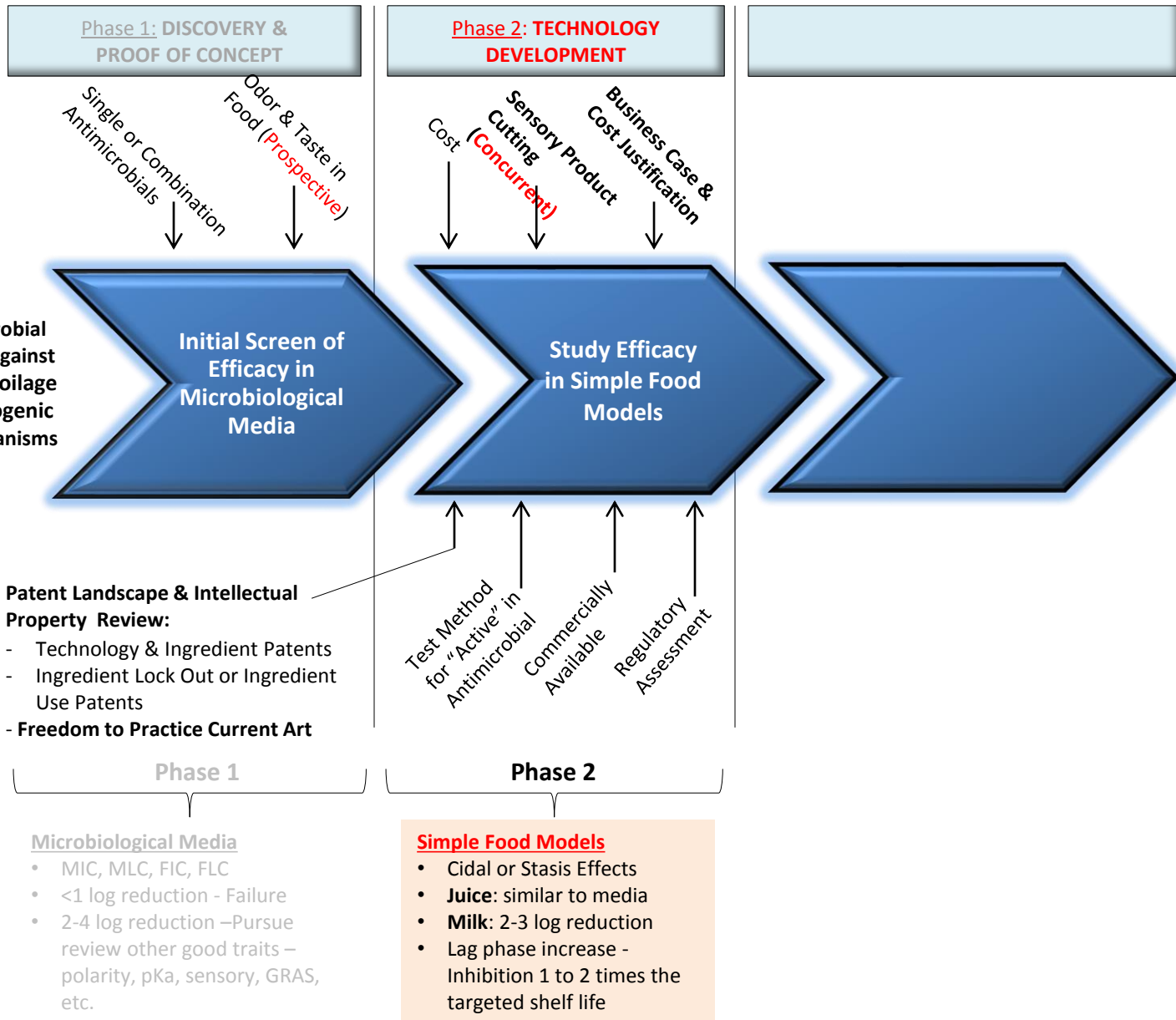
Technical Success Criteria

Phase 1

Microbiological Media

- MIC, MLC, FIC, FLC
- <1 log reduction - Failure
- **2-4 log reduction** –Pursue review other good traits – polarity, pKa, sensory, GRAS, etc.

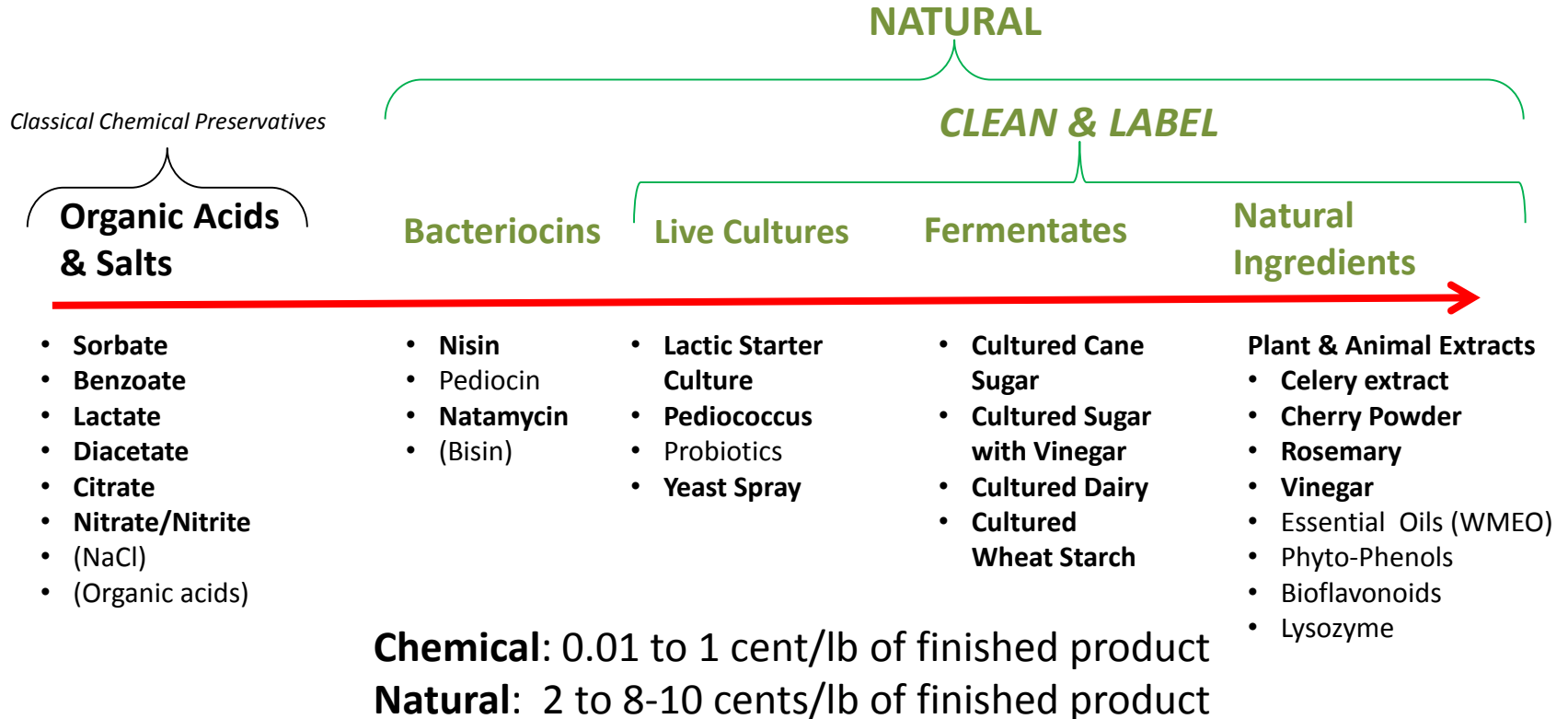
Process for Achieving Due Diligence – PHASE 2



Future Prospects

- 1.** Stronger Partnership: University-Vendor-User
- 2.** Bacteriocins for Gram negative Bacteria
- 3.** Effective preservative in the pH range of 5-7
- 4.** Standardization of Efficacy Determination
- 5.** Delivery system
- 6.** Influence on Natural Microflora

Antimicrobial Toolbox – PRICE LIST



Q: If an ingredient is \$10.00/lb. If your use level is 1% in finished product.

A: Then the cost-in-use will be 10 cent/lb of finished product

Example 1: Cost-in-Use

Bottom line:

50-100x more expensive for use of Natural Antimicrobial Clean Label version compared to Chemical version, *for same structure-function*

Basis: 300-400 gram product, 60 ppm nitrite

Natural Clean Label Antimicrobial

Cost per pound of Cultured Celery Juice Powder: **\$ 25.63**

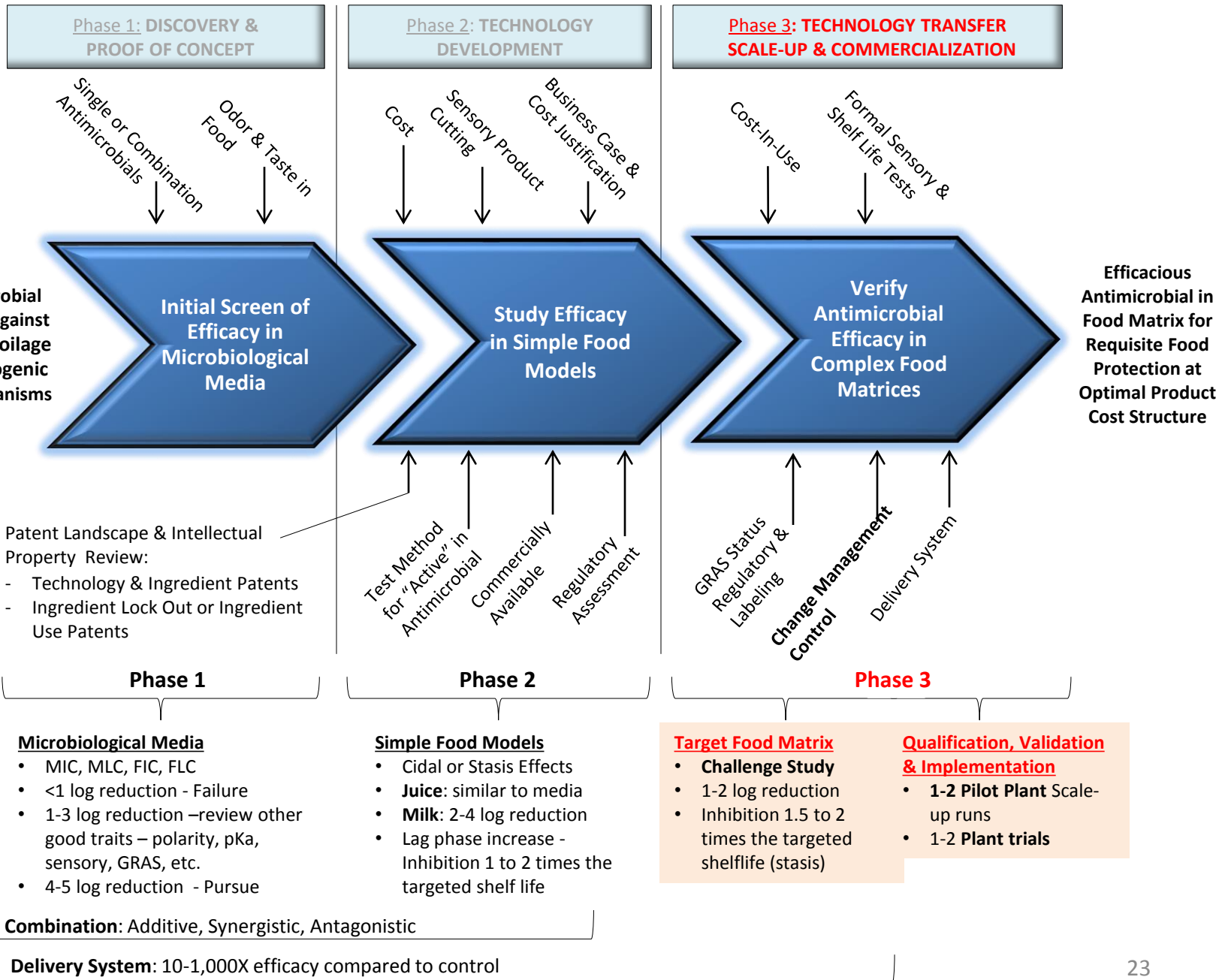
Cost-in-use would be approx 1.2 g to deliver 60ppm nitrite. This would result in approx **6-7 cents** in cost per 300-400 g

Approved Chemical Antimicrobial

Cost per pound for nitrite is **\$0.60**

Cost-in-use approx 0.43 g to deliver 60 ppm nitrite. This would result in approx **\$0.00057 (0.06 cent)** in cost per 400 g

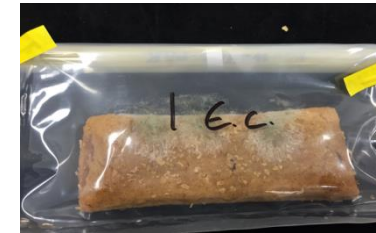
Process for Achieving Due Diligence – PHASE 3



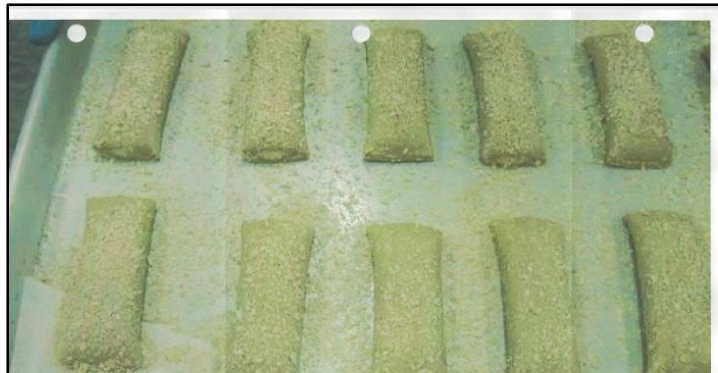
Example 2: Unintended Consequence



Eurotium chevalieri on CY20S agar plate



E. Chevalieri growth on a model cereal bar



CONTROL



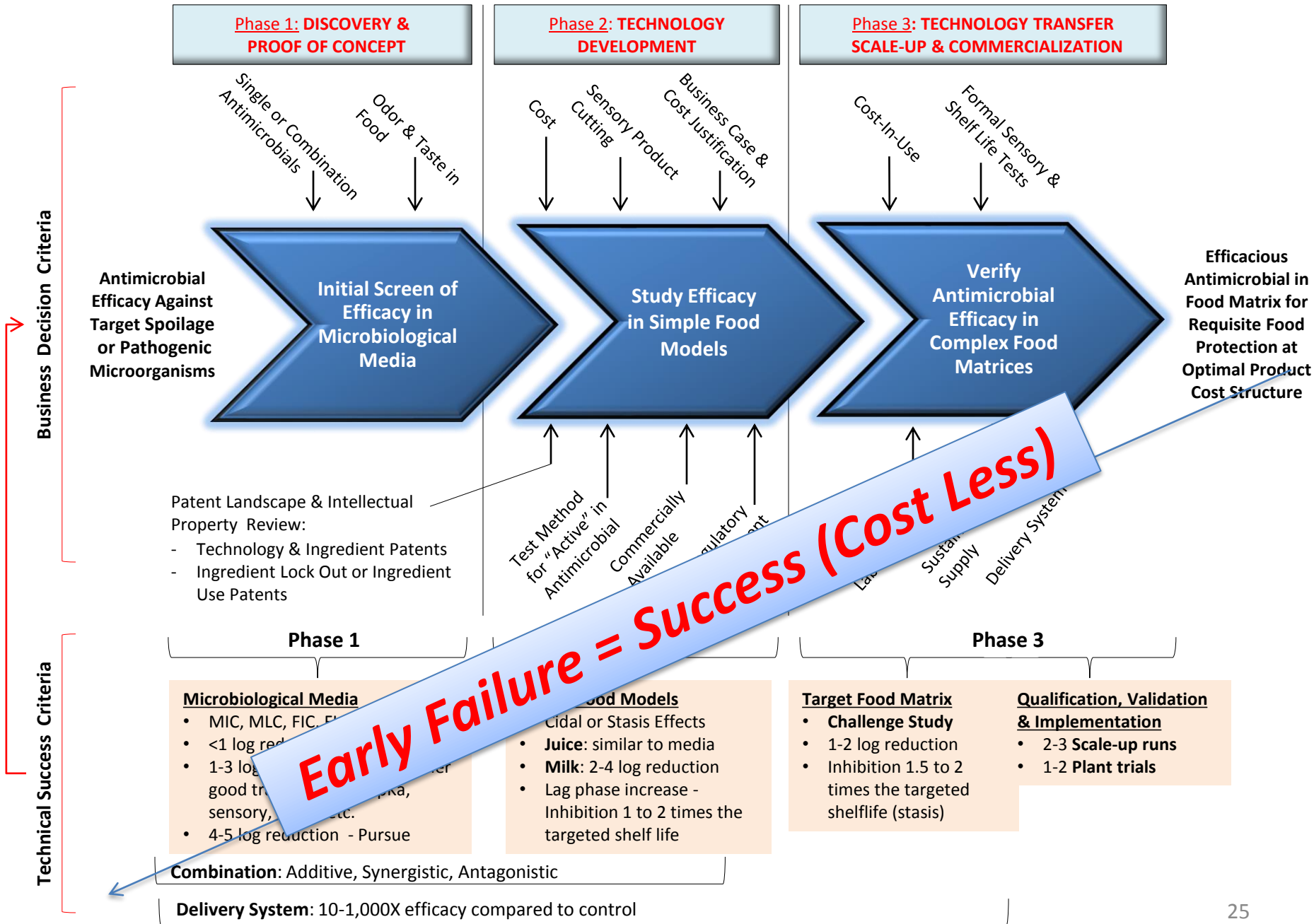
Syneresis of fruit core



Bake Test

Added antimicrobial should work with the current process parameters & formulation

STAGE GATE for Achieving Due Diligence



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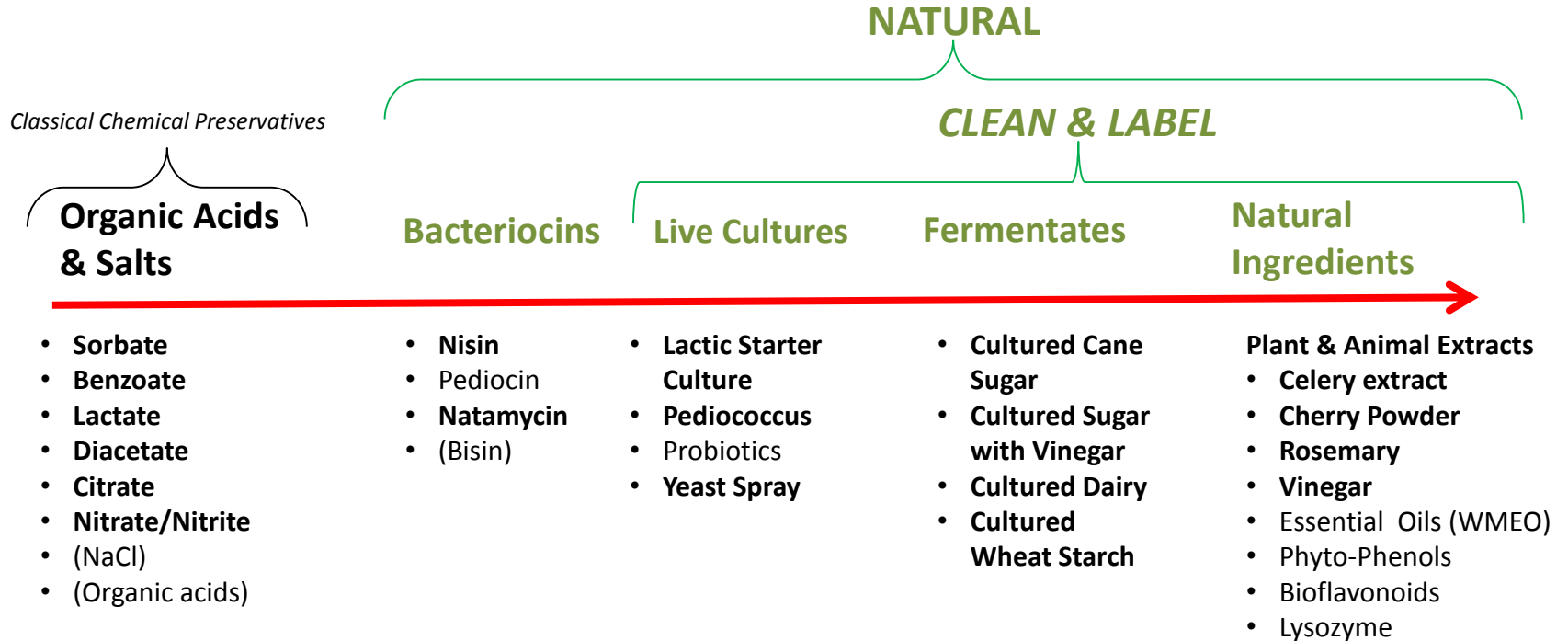
LOWERING aW

- Curing
- Drying

TANAKA MODEL
FERMENTATION
MAP

**ADDITION OF
ANTIMICROBIALS**

Antimicrobial Toolbox



KEY TAKEAWAYS

- ❑ **Consumer want natural or naturally derived antimicrobials vs chemical preservatives**
- ❑ **Food Industry Answer: Antimicrobial Tool Box for Pairing with Foods**
- ❑ **Steps for Adding Natural Antimicrobials to Foods – NOT SIMPLE**
- ❑ **Call for Continued Partnerships – University-Vendor- Food Industry**



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Appendix

Case Study: Model Fresh Sausage



Microbial Successions Are Associated with Changes in Chemical Profiles of a Model Refrigerated Fresh Pork Sausage during an 80-Day Shelf Life Study

Andrew K. Benson,^a Jairus R. D. David,^b Stefanie Evans Gilbreth,^b Gordon Smith,^b Joseph Nietfeldt,^a Ryan Legge,^a Jaehyoung Kim,^a Rohita Sinha,^a Christopher E. Duncan,^b Junjie Ma,^a Indarpal Singh^b

Department of Food Science and Technology, University of Nebraska, Lincoln, Nebraska, USA^a; ConAgra Foods, Inc., Omaha, Nebraska, USA^b

Applied and Environmental Microbiology p. 5178–5194

September 2014 Volume 80 Number 17

Fresh sausage product
Spice blend
Antimicrobials

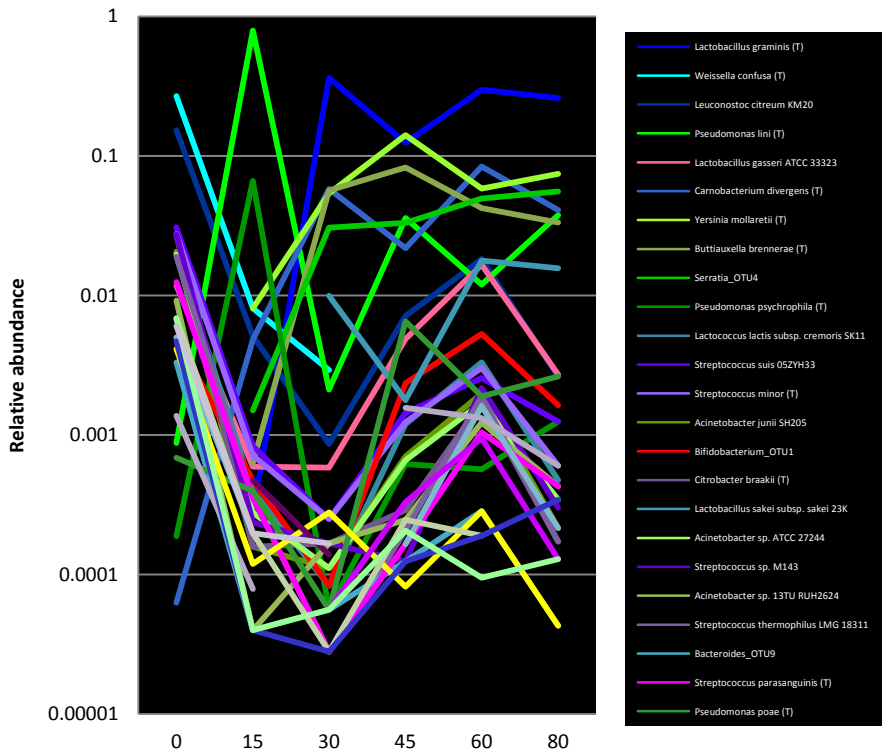


1. Traditional microbiological methods
2. Sniff testing –Meat Science
3. Microbiome analysis (16S)
4. Chemistry (eNose array)

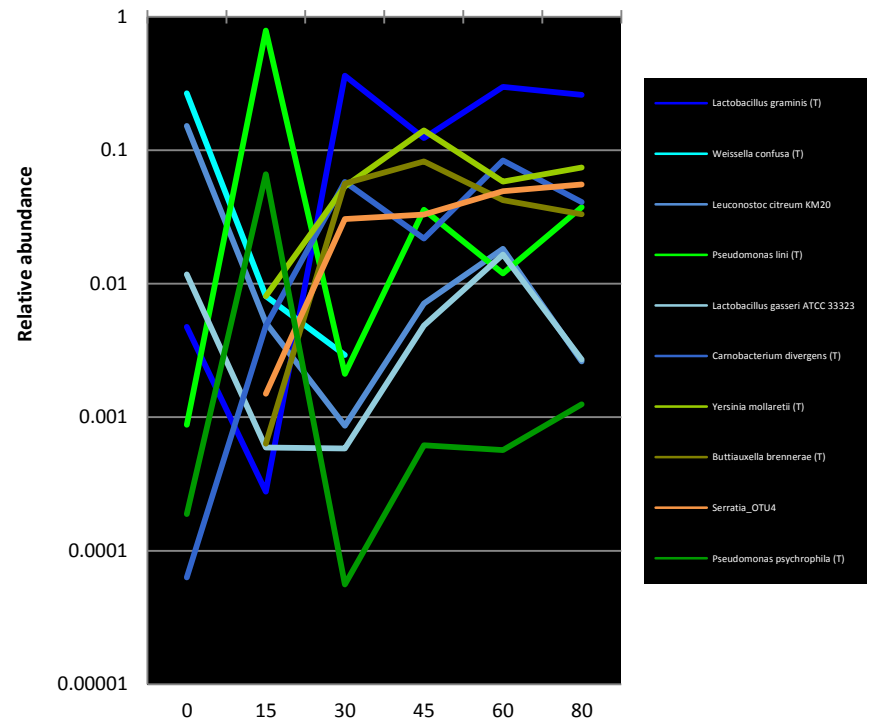
1. Big Disconnect between plate data and sensory spoilage
2. Ecological successions detected by NGS
 - a. Three major successions
 - b. Not observable by traditional micro
3. Shelf-life extenders eliminated successions, favored growth of **single species**
4. Source-tracking of **species** to spice blends, and not meat
5. Correlation analysis from eNose array identified 3 candidate taxa

Data Reduction: Focus on Behavior of The Most Abundant Taxa

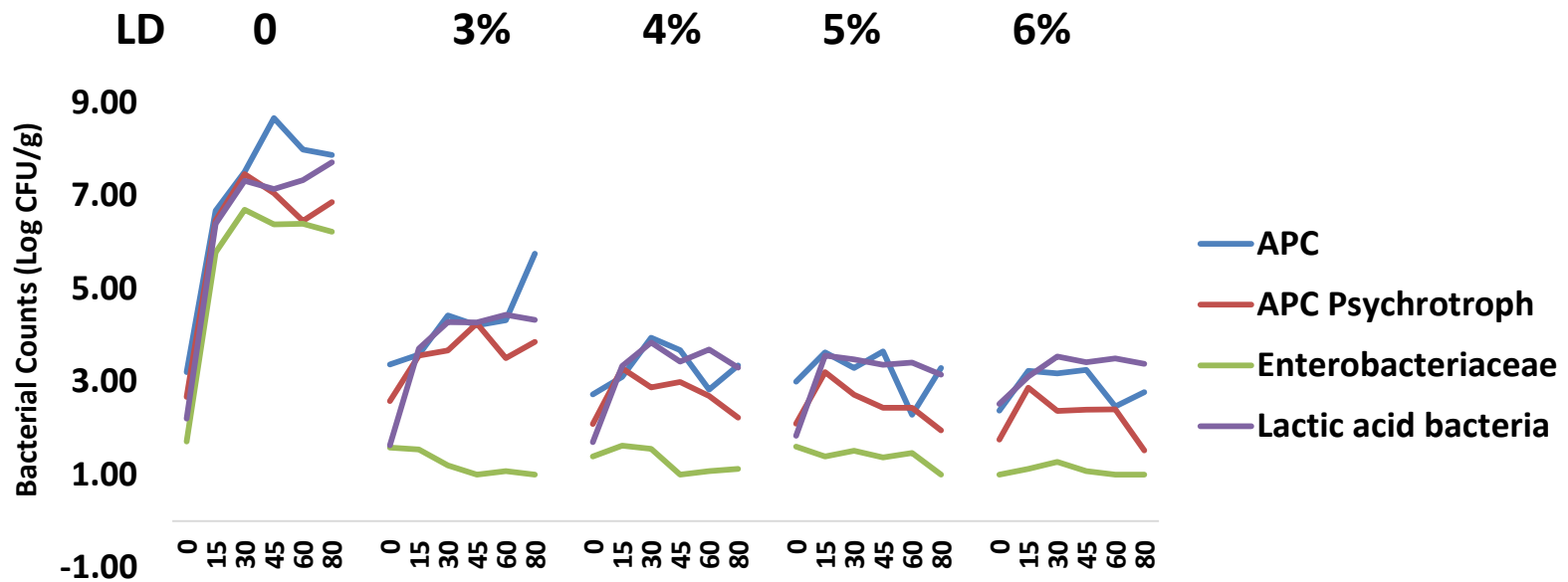
Most abundant



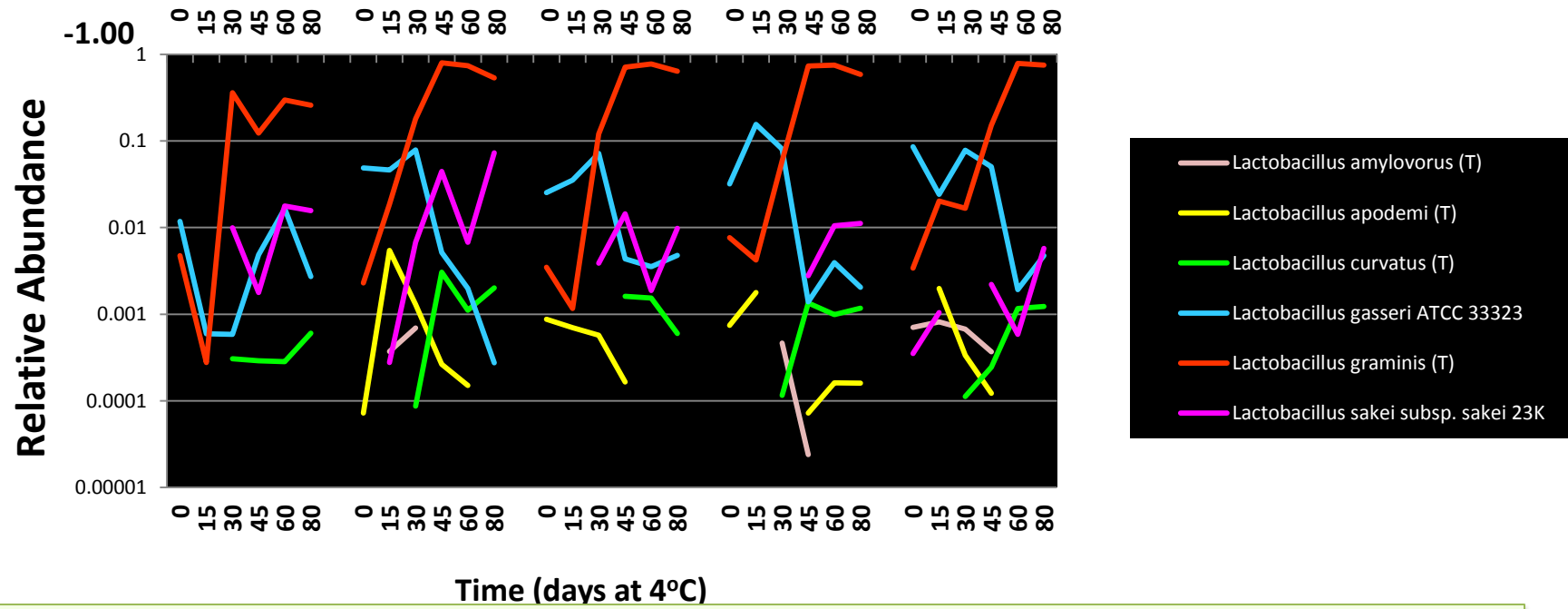
Overall most abundant



3 Major Successions



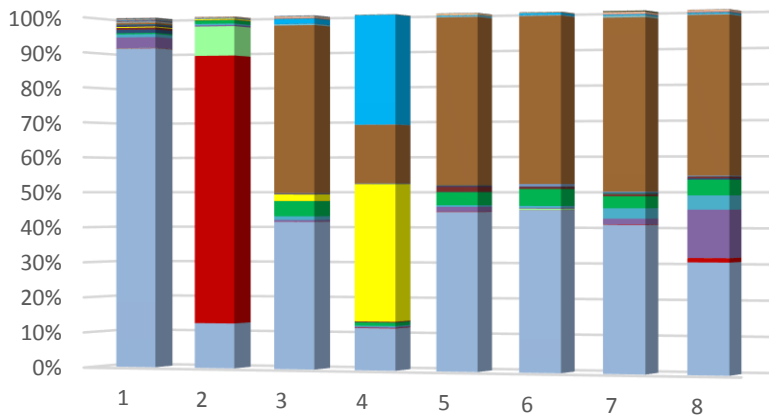
- APC
- APC Psychrotroph
- Enterobacteriaceae
- Lactic acid bacteria



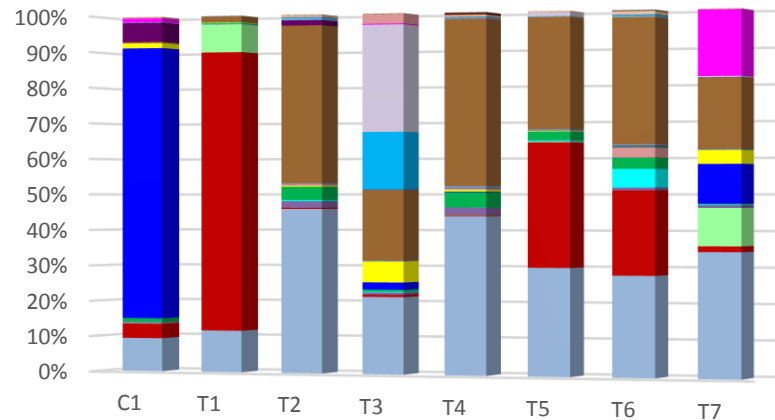
- Lactobacillus amylovorus (T)
- Lactobacillus apodemi (T)
- Lactobacillus curvatus (T)
- Lactobacillus gasseri ATCC 33323
- Lactobacillus graminis (T)
- Lactobacillus sakei subsp. sakei 23K

Antimicrobials Eliminated 3 Major Successions
Favoring a Single Species of Lactobacillus = Signature Spoilage Microorganism

Day 0



21 days at 4C



- Bifidobacterium
- Pseudomonas
- Yersinia
- Bacillus
- Rhizobium
- Shinella
- Streptococcus
- Schlegelella
- Rahnella
- Dechloromonas
- Chryseobacterium
- Cronobacter
- Veillonella
- Lactobacillus
- Acinetobacter
- Pantoea
- Citrobacter
- Fusobacterium
- Brevundimonas
- Sphingomonas
- Porphyromonas
- Hymenobacter
- Shewanella
- Xanthomonas
- Brevibacterium
- Gibbsiella
- Lactococcus
- Rhodococcus
- Pediococcus
- Psychrobacter
- Novosphingobium
- Staphylococcus
- Leuconostoc
- Methylobacterium
- Weissella
- Aeromonas
- Cohnella
- Klebsiella

- Bifidobacterium
- Pseudomonas
- Yersinia
- Bacillus
- Rhizobium
- Shinella
- Streptococcus
- Schlegelella
- Rahnella
- Dechloromonas
- Chryseobacterium
- Cronobacter
- Veillonella
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- Acinetobacter
- Pantoea
- Citrobacter
- Fusobacterium
- Brevundimonas
- Sphingomonas
- Porphyromonas
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- Xanthomonas
- Brevibacterium
- Gibbsiella
- Lactococcus
- Rhodococcus
- Pediococcus
- Psychrobacter
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- Staphylococcus
- Leuconostoc
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- Aeromonas
- Cohnella
- Klebsiella

Phase 3: C1 vs. T1, T2, T4