

INNOVATIVE METHODS TO PRESERVE QUALITY AND SHELF LIFE OF CLEAN LABEL TORTILLAS

Presenter: Joy Zhong, Ph.D.
R&D Director
Kemin Food Technologies – North America

OVERVIEW

- Introduction
- Microbes of concern
- Preservatives
- Mode of action
- Efficacy studies



COMPANY OVERVIEW

- Founded in 1961 by R.W. and Mary Nelson
- Family owned and operated
- More than 500 patents and applications
- Annual revenue exceeds USD \$1 Billion
- Transforming a billion lives every day



1961

Our



We strive to sustainably transform the quality of life every day for 80 percent of the world with our products and services.

Consumer Purchasing Trends



Three main trends impact consumer behaviors;

1. Cost-conscious choices
2. Holistic wellness
3. Values-based purchasing

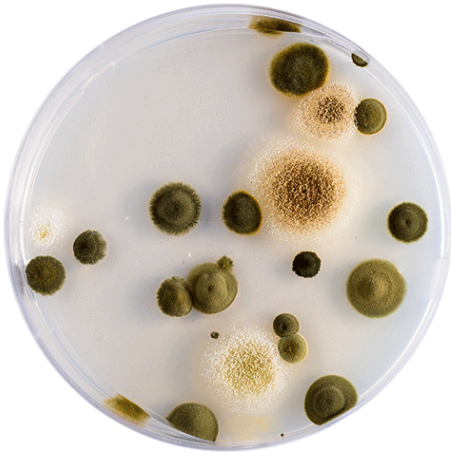
MICROBIAL SPOILAGE

MICROBIAL SPOILAGE

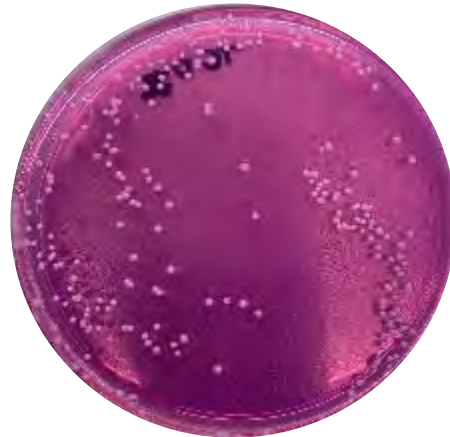
- Baked goods are generally at a lower risk of causing food poisoning as compared to other food.
- However, spoilage in baked goods is a major concern.
- Estimated bakery product loss is 5% in US & 1 - 5% in Europe
- Spoilage incurs huge economic losses
- It also affects the entire food product chain

MICROBES OF CONCERN

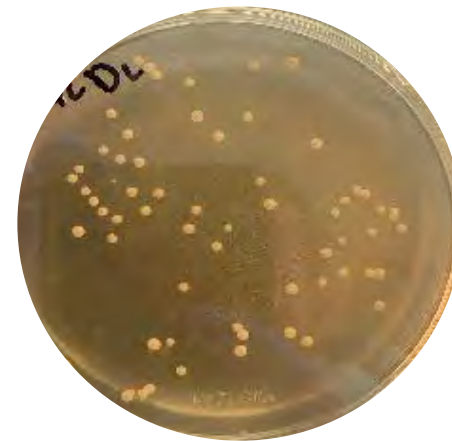
TYPES OF MICROBES



Mold



Yeast

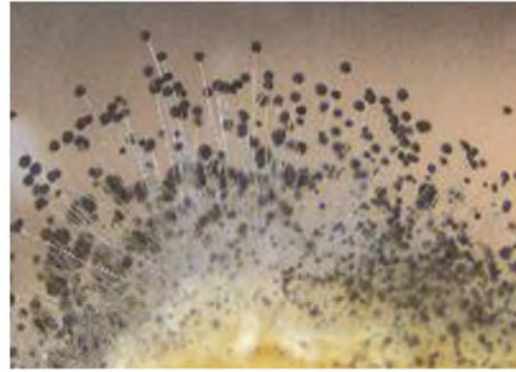


Bacteria

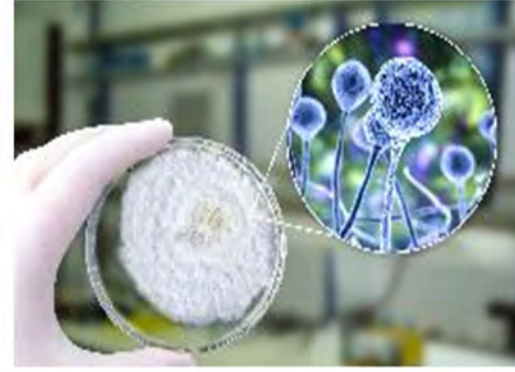
COMMON MICROBES IN TORTILLA



Aspergillus



Rhizopus



Mucor



Neospora



Geotrichum

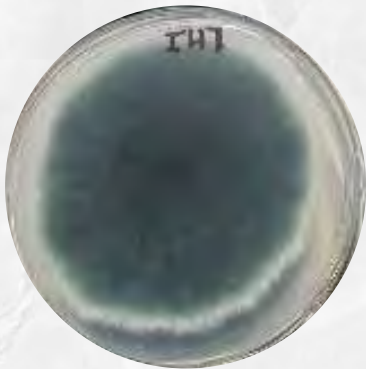


Bacillus

RESISTANT MOLDS

PENICILLIUM:

- A few species are preservative resistant - e.g., *Penicillium roqueforti*, *P. paneum*, *P. carneum*.
- *P. roqueforti* a sorbate resistant mold, produces 1,3 pentadiene - kerosene smell
- *P. roqueforti* can grow under refrigerated temperature, also called "cold weather mold"



MONASCUS:

- Heat resistant mold
- *Monascus spp*: e.g., *Monascus ruber*, *M. pilosus*
- Survive kill steps e.g., pasteurization, baking
- Also called "summer month mold" or ascospores



FACTORS THAT INFLUENCE SPOILAGE

Intrinsic Factors

- Moisture content
- Water activity
- pH
- Nutrients

Extrinsic Factors

- Raw materials
- Processing conditions
- Cleaning/sanitation
- Packaging/storage

INTRINSIC FACTORS - MOISTURE & WATER ACTIVITY (a_w)

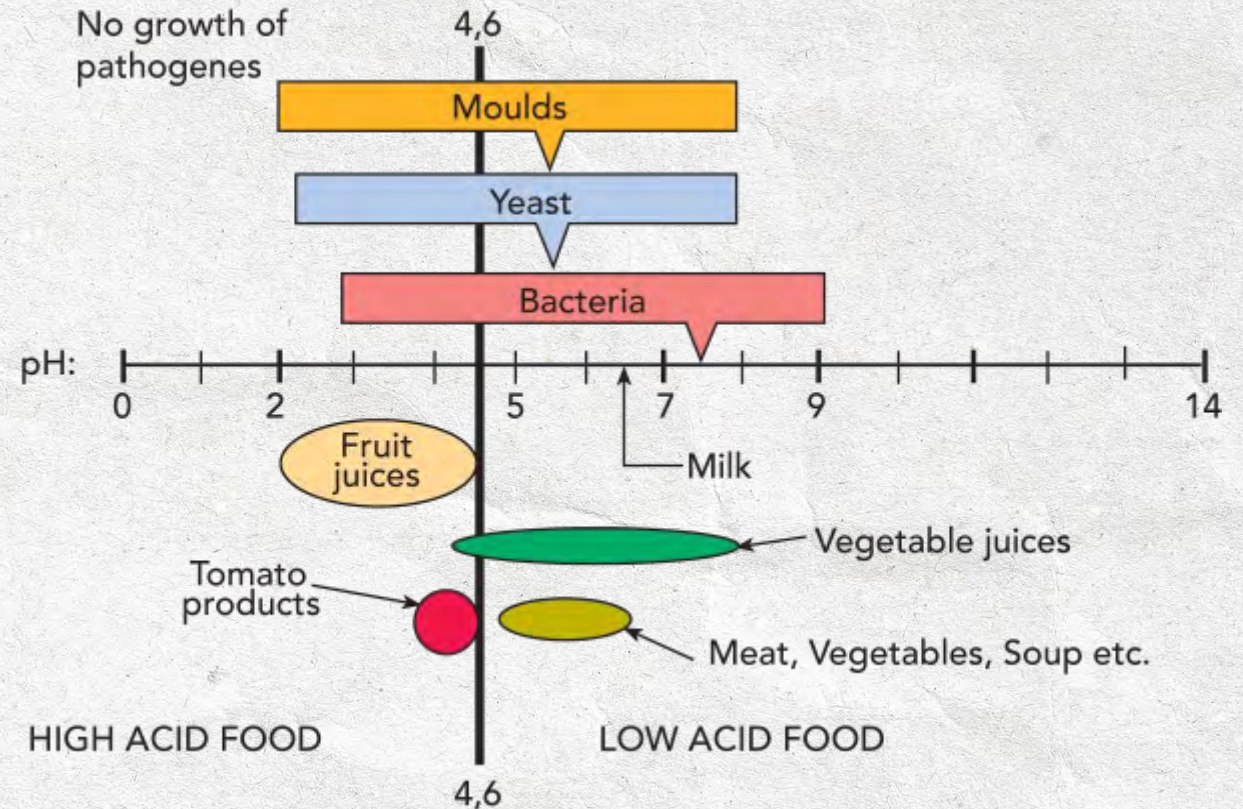
- **Moisture** = total moisture
- a_w = Free water available to microbes
- **Tortilla moisture** = 35 – 50% and a_w of 0.8 to 0.97
- Lowering a_w can hinder microbial growth
- **Solutes** – salt/sugar can reduce a_w
- Could impact sensory and texture

MICROBES	SPECIES	MINIMUM a_w
Most spoilage bacteria		0.90-0.91
Bacteria	Bacillus cereus	0.92-0.95
Bacteria	Clostridium botulinum	0.90-0.98
Most molds		0.80-0.98
Mold	Aspergillus spp	0.68-0.90
Mold	Aspergillus flavus	0.78-0.90
Mold	Aspergillus niger	0.80-0.84
Mold	Fusarium spp	0.82-0.92
Mold	Mucor spp	0.80-0.93
Mold	Penicillium spp	0.78-0.93
Xerophilic molds		0.65
Spoilage yeasts		0.88
Yeast	Saccharomyces baillii	0.80
Yeast	Saccharomyces cerevisiae	0.90-0.94
Yeast	Saccharomyces rouxii	0.62
Osmophilic yeast		0.6

<https://thefooduntold.com/blog/food-science/water-activity-aw-and-food-safety>

NUTRIENTS, pH & OTHERS

- Excellent nutrition source
 - Carbs
 - Fat
 - Protein
 - Sugar
- pH = 4.8 – 12.0
- Typical storage = 70 – 90 °F
- Oxygen in package
- Storage time is favorable for the growth of mold



EXTRINSIC FACTORS - RAW MATERIALS

Raw Materials – Raw Agricultural Commodity

- Potential source of mold, yeast and bacteria
- Spores of resistant molds - *P. roqueforti*, *P. paneum*, *P. polonicum*, *Monascus*.

Flour dust carrier for spores on equipment surface and/or processing area

- Water quality
- Wooden pallets and cardboard boxes - spores to the packaging area



EXTRINSIC FACTORS - PROCESS

Baking – Temp, Time

- HRM spores (ascospores) can survive baking
- Ascospores contaminate food equipment surfaces

Cooling/Temperature Gradient

- Water condensation
- Surfaces, walls, ceiling, overhead piping
- *Penicillium roqueforti* can grow in colder climates

Recontamination Post Baking



EXTRINSIC FACTORS - PACKAGING & STORAGE

Packaging Materials

- Vacuum packaging, MAP

Storage Conditions

- Refrigeration, frozen, ambient



EXTRINSIC FACTORS - ENVIRONMENT & CLEANING

Air Quality

- Create positive air pressure in plant
- Removal external contamination
- Filtration of incoming air – HEPA filter

Cleaning and Sanitation of Equipment

Personal Hygiene

- Wearing Gloves



PRESERVATION HURDLE TECHNOLOGY

Hurdle Technology: Multiple Barriers

- Water activity (a_w)
- Thermal kill step-Baking
- Formulation-Preservatives/pH
- Innovative Packaging/MAP, Vacuum, O_2 Scavengers
- Storage temperature (Refrigerated/Frozen)



SYNTHETIC PRESERVATIVES

Antimicrobials are extensively used to inhibit microbial spoilage in tortillas

- Propionic acid is commonly used mold inhibitor
- Sorbic acid and benzoic acid are used as helper molecules

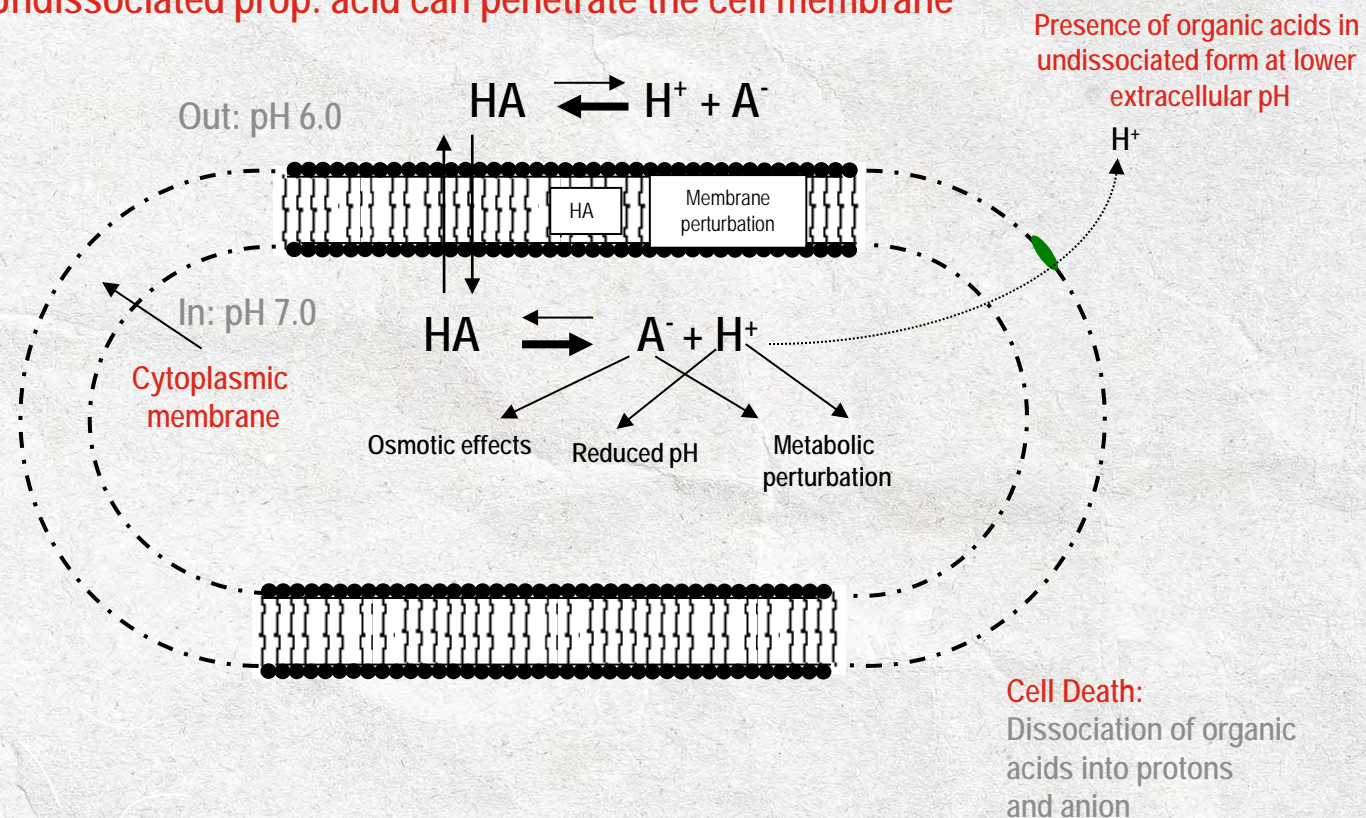


Antimicrobials	SPOILAGE MICROORGANISM		
	Mold	Yeast	Bacteria
Propionic acid	X		X
Sorbic acid	X	X	X
Acetic acid	X		X
Benzoic acid	X	X	X
Parabens	X		X

PRESERVATIVES MODE OF ACTION

- **Propionic acid**
 - Undissociated acid theory/acid stress
- **Sorbic acid**
 - Partly due to undissociated acid
 - Loss of lipid membrane integrity
 - Inhibition of enzymes required for transportation
- **Benzoic acid**
 - Alter membrane fluidity - disruption of membrane trafficking and dynamics

Undissociated prop. acid can penetrate the cell membrane



Adapted from Hirshfield *et. al*, 2003
Busta *et. al.*, 1986

pH Role in Preservation

Undissociated Propionic Acid (%)	pH
99	2.87
95	3.59
90	3.92
80	4.27
70	4.50
60	4.69
50 (pK _a)	4.87
40	5.05
30	5.24
20	5.47
10	5.82
1	6.87

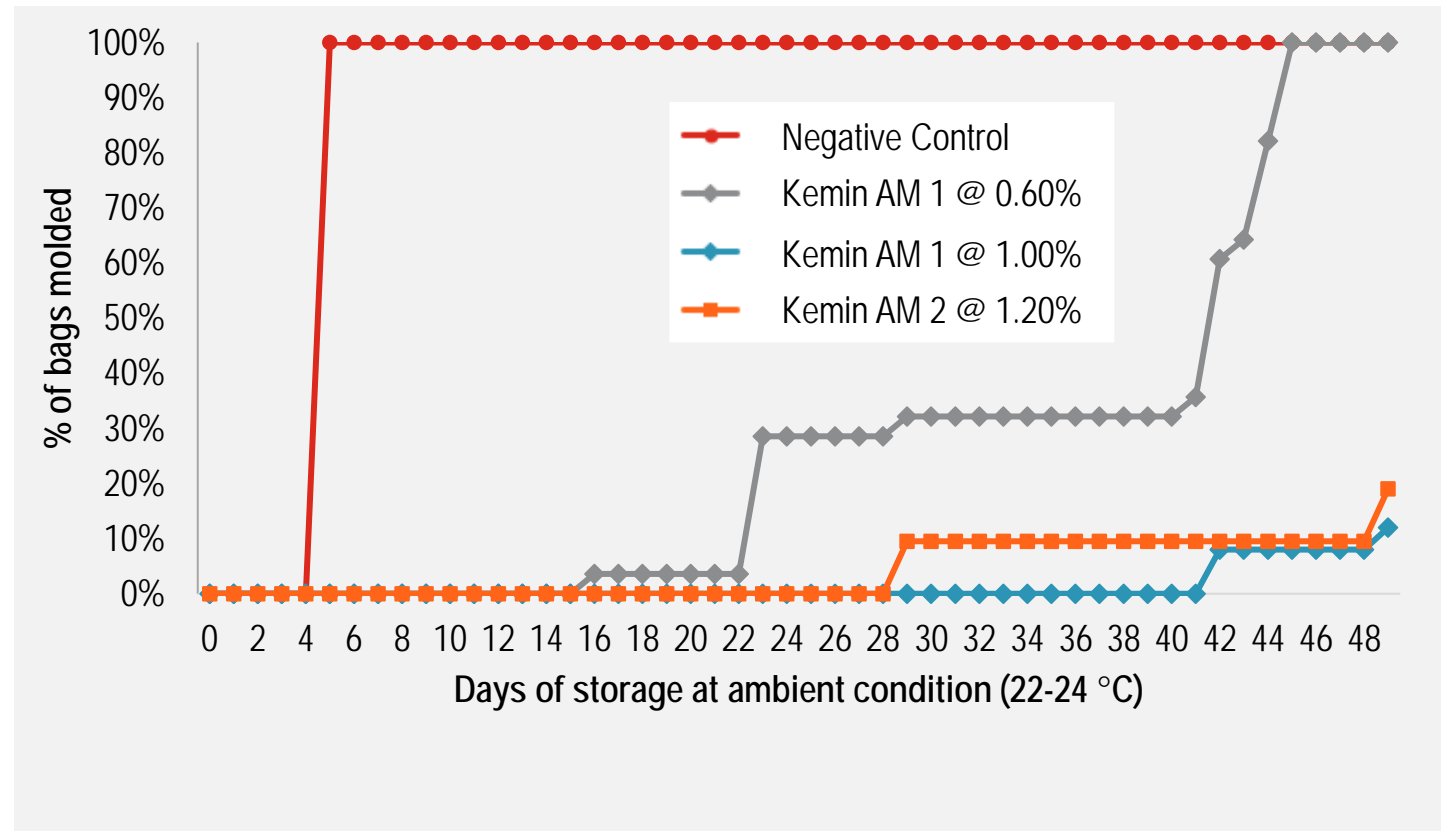
Acidulants

- Lower the pH of finished product
- Improve the efficiency of preservatives
- Disadvantage: affect the after taste of product

pK_a = pH when concentration of acid is equal to its conjugate base i.e., acid is 50% dissociated

EFFICACY OF SYNTHETIC ANTIMICROBIALS

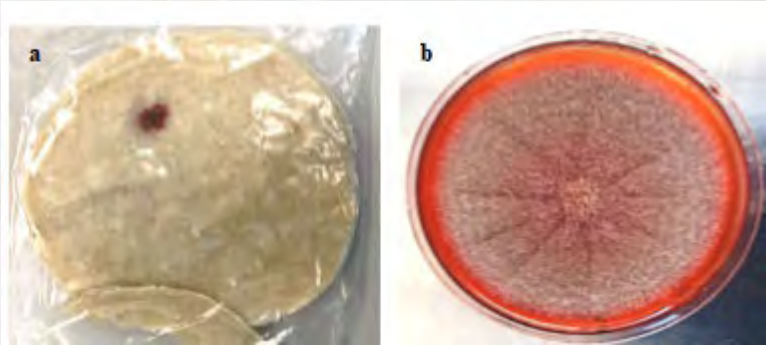
- Combination of organic acid and other antimicrobials extend the shelf life
- Efficacy is based on concentration and ingredients used
- Based on the helper molecules used the efficacy can vary.



Negative control – no antimicrobial, Kemin AM 1 – synthetic liquid antimicrobial, Kemin AM 2 – synthetic liquid antimicrobial.

EFFICACY AGAINST RED MOLD (MONASCUS)

- Monascus are highly resistant to some of the organic acids
- Helper molecules (organic acids, peptides, etc.) can enhance efficacy against these red molds
- Formulation containing propionic acid along with specific helper molecules can control red mold growth during the summer months



Treatment groups	Dosages tested %	Growth at 45 days incubation		
		<i>M. ruber</i>	<i>M. pilosus</i>	<i>M. purpureus</i>
KEMIN AM FORMULA 1	1.00, 1.25, 1.50	No	No	No
SHIELD SPB LIQUID	1.00, 1.25, 1.50	No	No	No
KEMIN AM FORMULA 2	1.00, 1.25, 1.50	Growth at 1%	Growth at 1%	Growth at 1%
KEMIN AM FORMULA 3	1.00, 1.25, 1.50	Yes	Yes	Yes

CLEAN LABEL ANTIMICROBIALS

CLEAN LABEL ANTIMICROBIALS

- Source – naturally derived
- No synthetic ingredients
- Fermented products
 - Cultured dextrose
 - Cultured wheat/whey
 - Cultured feedstock
- Essential oils
- Plant extracts – herbal, berry extracts

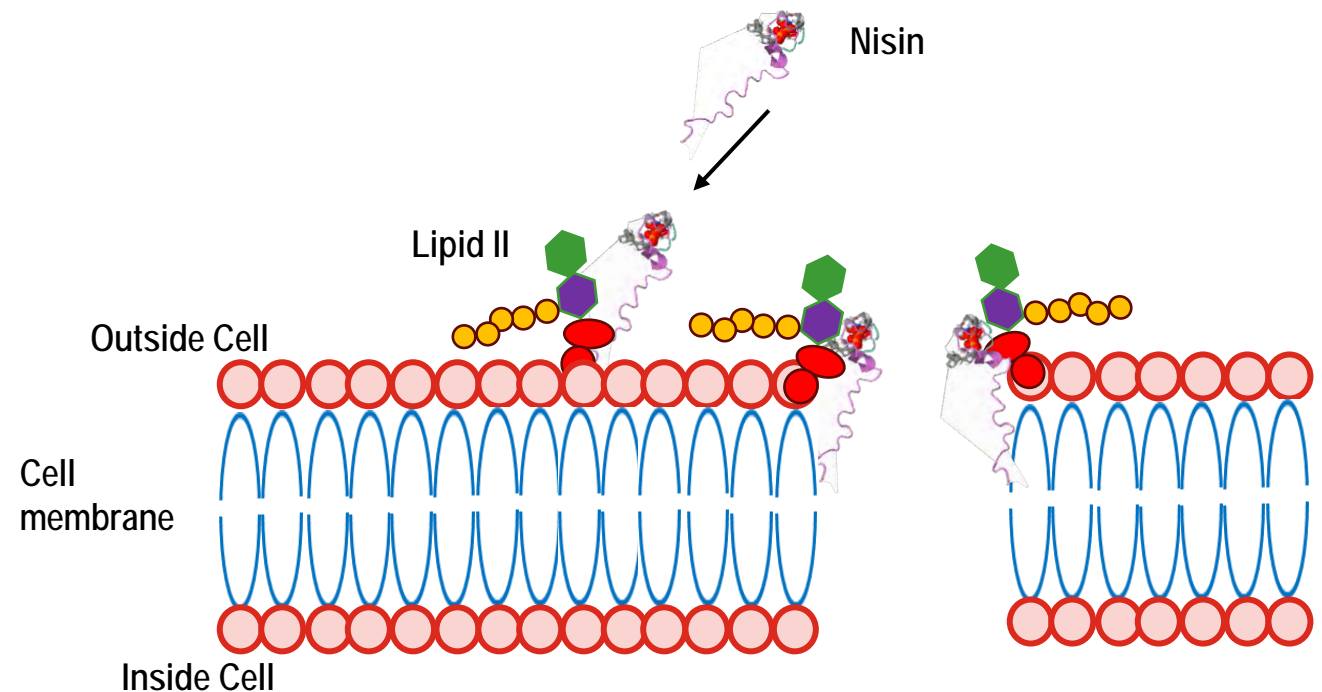
ACTIVE INGREDIENTS - FERMENTED PRODUCTS

Typical actives include

- Short chain fatty acids
- Microbial peptides - bacteriocins

Fatty acids in fermented products

- Propionic acid, acetic acid, lactic acid
- Valeric acid, butyric acid, hexanoic acid and heptanoic acid
- Mode of action of fatty acids are similar to synthetic
- Mode of action of peptides – e.g., nisin
 - Inhibition of cell wall synthesis
 - Pore formation



Adapted From

Perez *et al*, 2015. Bacteriocins from Lactic Acid Bacteria: A Review of Biosynthesis, Mode of Action, Fermentative Production, Uses, and Prospects
<https://proteopedia.org/wiki/index.php/Nisin>

PRODUCTION OF FERMENTED PRODUCTS

- Use of microbial strains
- Fermentation of different feedstocks
- Production of organic acids and other antimicrobial compounds during the growth of the microbes
- Concentration varies
- May be dried



PRODUCTS AVAILABLE

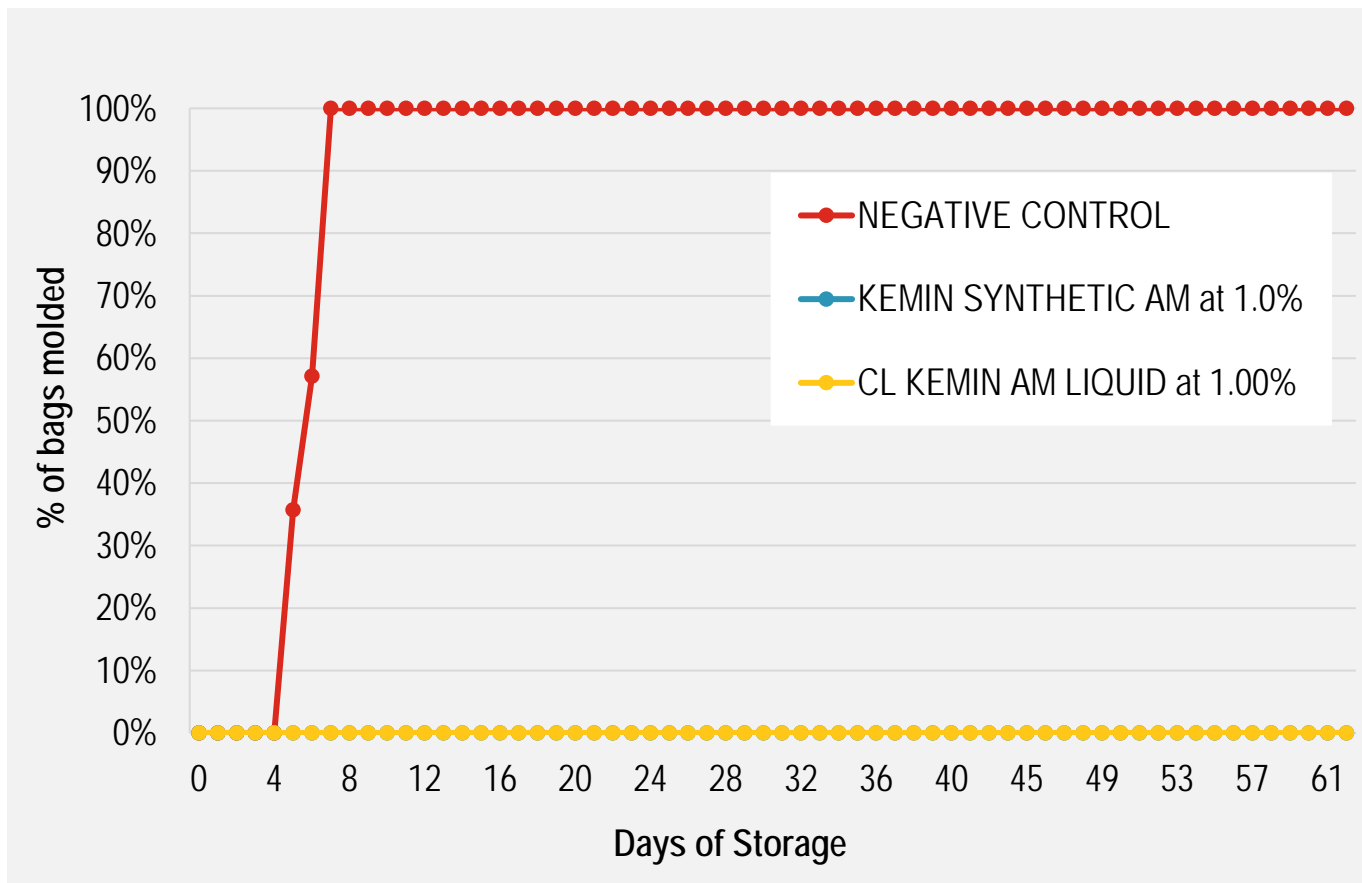
Based on the microbe used and the process, the products available in the market can vary:

- Efficacy due to the different active molecule and active level – based on process – concentration
- Sensory – based on the feedstock used and downstream process to remove impurities
- Cost/cost-in-use – vary based on manufacturing cost as well as the active concentration (dosage)

LIMITATIONS

- Typical level of actives is low – ranging from 0 – 80%, higher dosage to be used based on the product
- Consistency due to the variability in the fermentation process – if the actives are not standardized
- Impact on color due to the fermented product
- Other sugars and ingredients present in the dried – causing sensory impact
- Increase cost-in-use over synthetics
- May have an impact on the texture of the finished product

EFFICACY IN CORN TORTILLAS



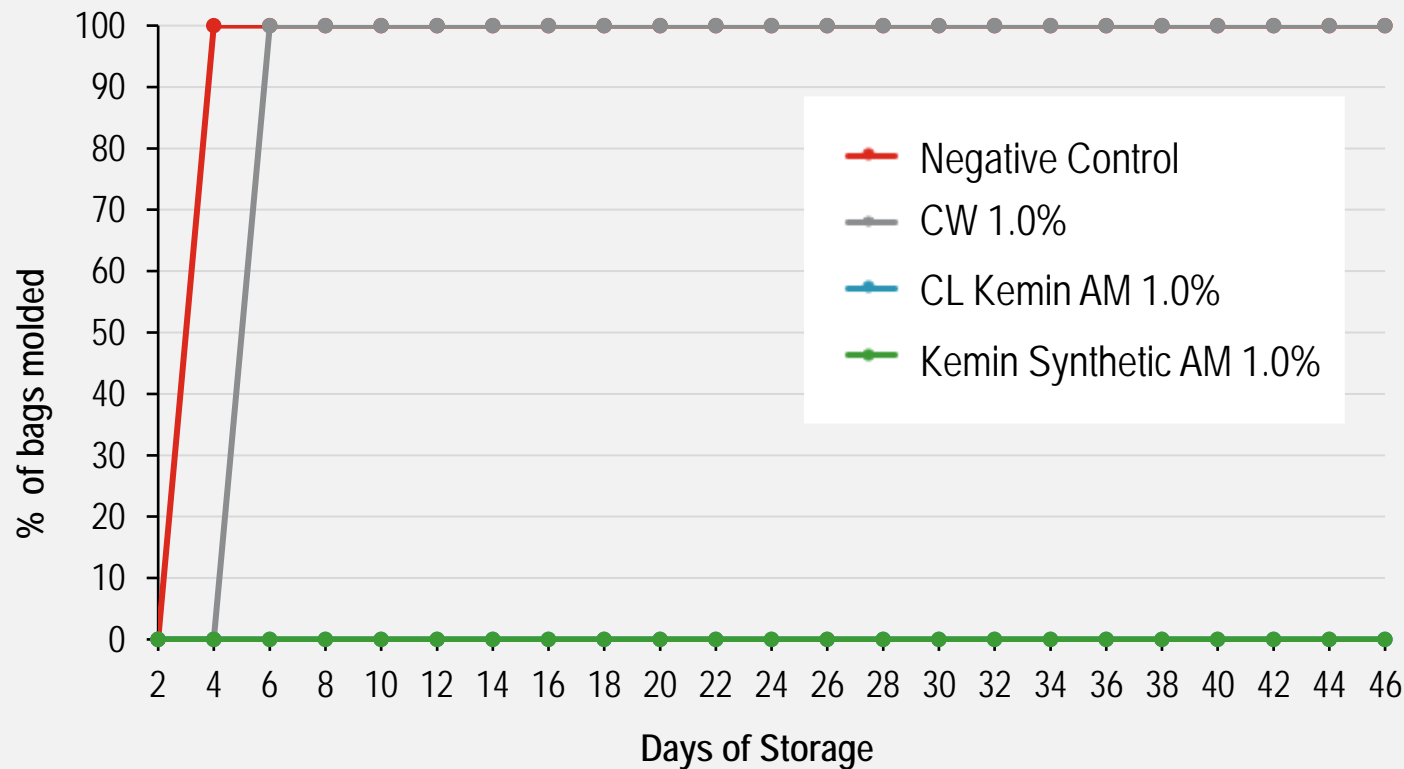
Negative control – no antimicrobial, Kemin synthetic AM – contains propionic acid and benzoic acid, CL Kemin AM liquid – cultured dextrose-based product

Sensory Results

Groups	Hedonic score
Kemin synthetic AM at 1.0%	7.17 ± 0.75
CL Kemin AM Liquid at 1.0%	6.50 ± 0.54

Cultured dextrose-based product was similar to synthetic product at equal concentration

EFFICACY IN CORN TORTILLA



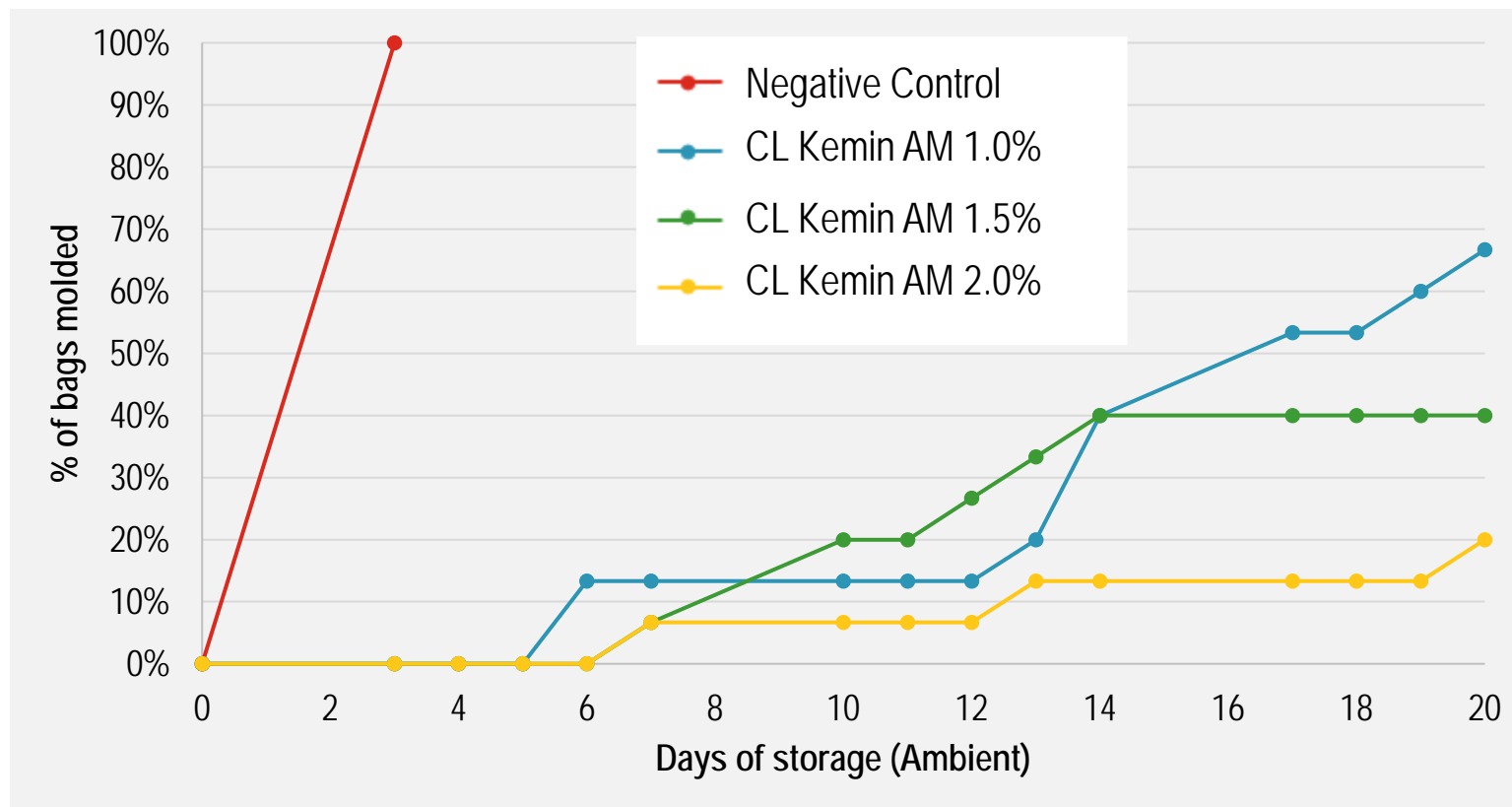
Cultured wheat provided 2 more days of shelf life than negative control.

Cultured dextrose-based product was similar to synthetic product at equal concentration.



Negative control – no antimicrobial, CW – cultured wheat, CL Kemin AM liquid – cultured dextrose-based product, Kemin synthetic AM – contains prop & benzoic acid

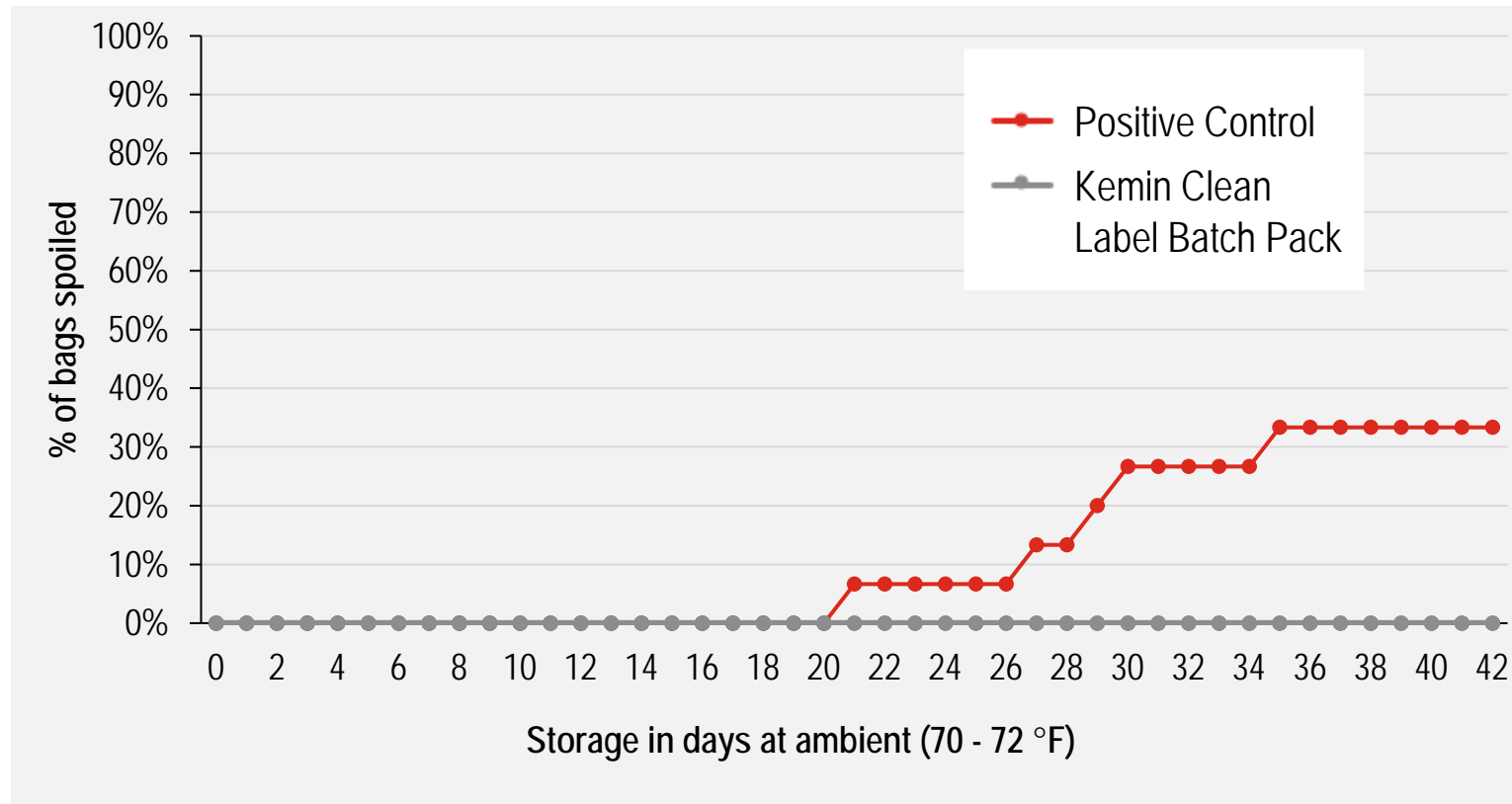
EFFICACY IN FLOUR TORTILLA



Negative control – no antimicrobial, CL Kemin AM – Clean label potassium sorbate replacement

Clean label mold inhibitor improved shelf life from 3 days to 7 days.

CLEAN LABEL BATCH PACKS - FLOUR TORTILLA



Positive control – Clean label batch pack

Clean label batch pack extended the shelf life from 21 days to more than 42 days.

ANTIOMICROBIALS FROM PLANT EXTRACTS

Blend of **buffered vinegar** and **rowan berry extract** for mold inhibition.

Typical Applications

- Baked Goods & Tortillas
- Dressings & Sauces
- Fillings



EFFICACY IN FLOUR TORTILLA



Untreated



Synthetic AM

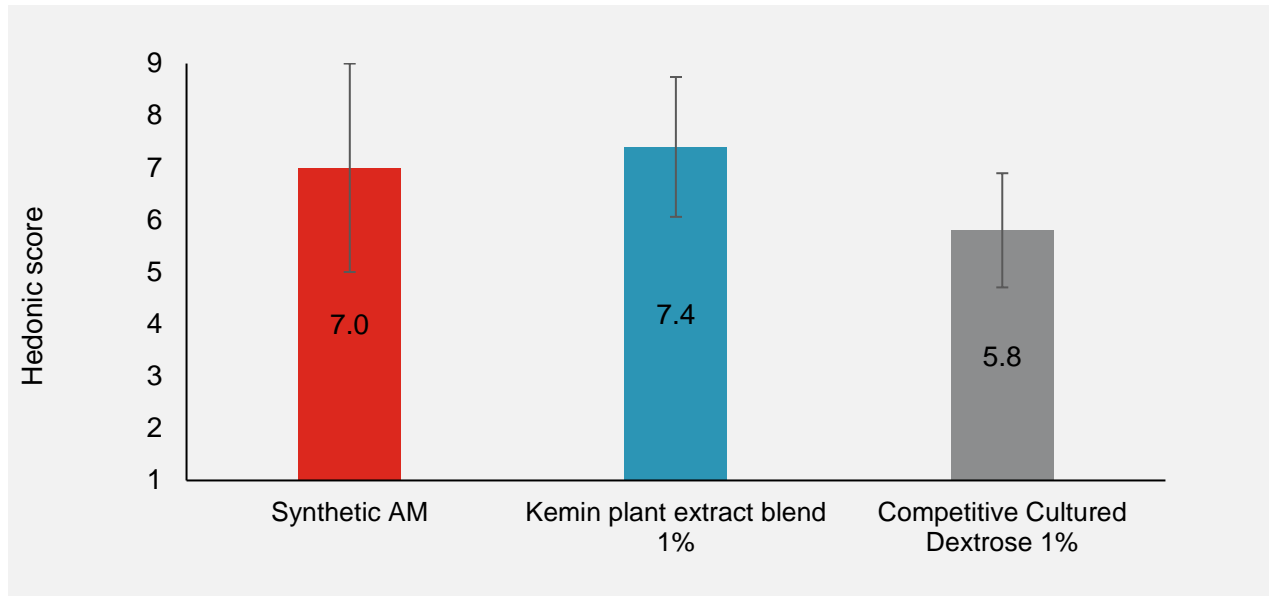


Competitive cultured
dextrose (1%)



Kemin plant extract
blend (1%)

	Untreated	Kemin plant extract blend	Competitive cultured dextrose
Days to mold	4	8	7



Sensory scores of flour tortillas at week 1 (source: TD-24-10132)

CONCLUSIONS

- Clean label antimicrobials available – fermented products/plant extracts
- The available products vary hugely on efficacy, cost and sensory impact
- Appropriate selection and testing is required to choose the most suitable product for your matrix

KEMIN ANTIMICROBIALS & MOLD INHIBITORS

- **SHIELD®**
 - Blend of synthetic propionic acid and sorbic acid that help improve shelf life of corn tortillas.
- **SHIELD® PURE**
 - A clean label cultured dextrose that can replace calcium propionate one to one in corn and flour tortillas.
- **SHIELD® V**
 - A blend of buffered vinegar and botanical extract helps to prevent mold in flour tortillas.

THANK YOU

- **Gokila Thangavel Ph.D. | Senior Scientist | Kemin Food Technologies**