Uses of Liquid Preservatives for Clean Label Tortillas

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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

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Our VISION

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

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Consumer Purchasing Trends

Three main trends impact consumer behaviors;

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

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MICROBIAL SPOILAGE

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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

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MICROBES OF CONCERN

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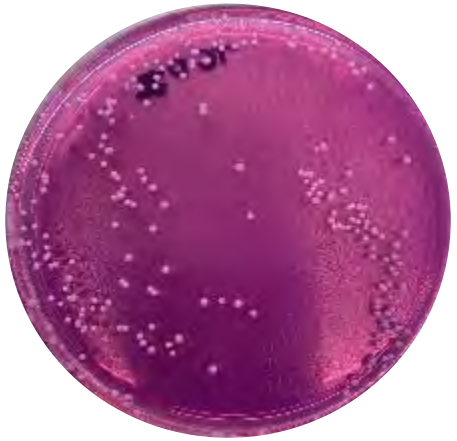
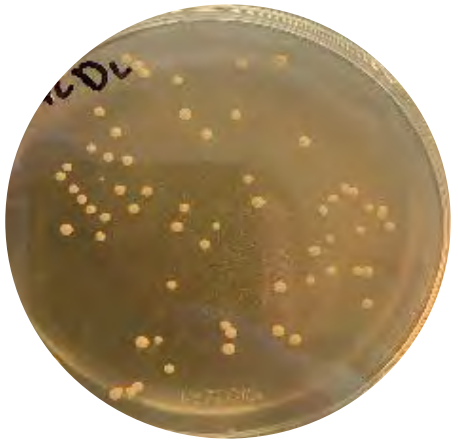
TYPES OF MICROBES

Mold



Yeast



Bacteria

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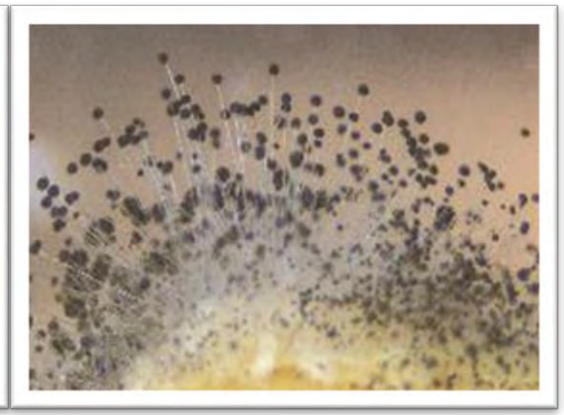
COMMON MICROBES IN TORTILLA

Aspergillus



Rhizopus

Mucor

Neospora

Geotrichum

Bacillus

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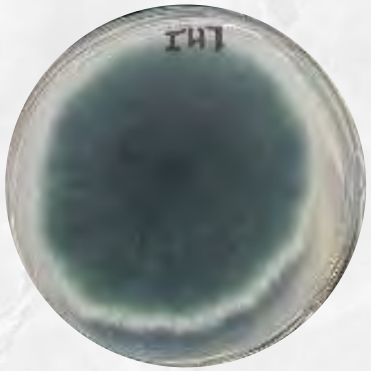
RESISTANT MOLDS

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

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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 bailii	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

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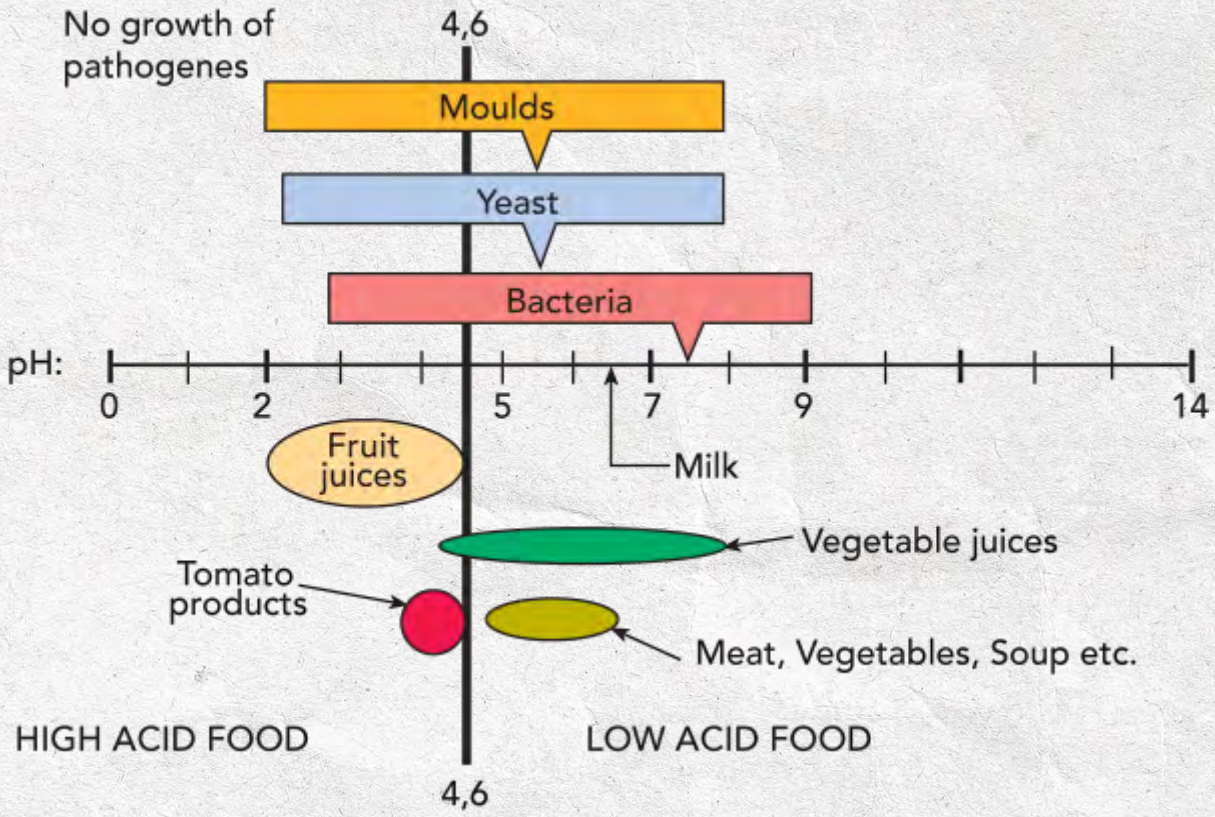
NUTRIENTS, pH & OTHERS

- 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



https://dairyprocessinghandbook.tetrapak.com/chapter/microbiology

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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

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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

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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)

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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

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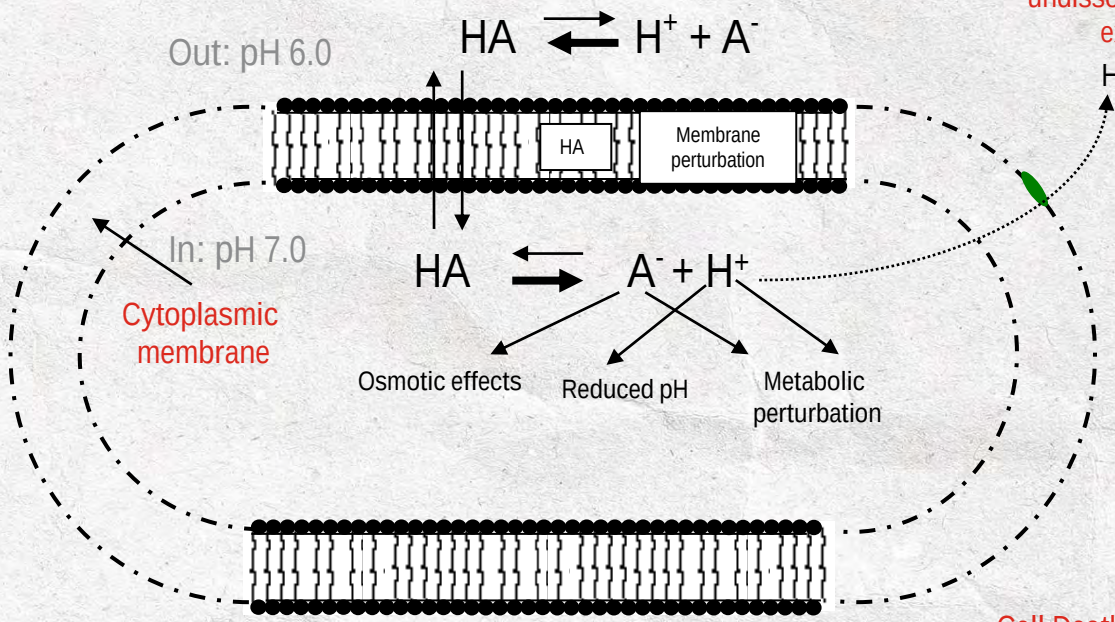
PRESERVATIVES MODE OF ACTION

- 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

Presence of organic acids in undissociated form at lower extracellular pH



Cell Death:
Dissociation of organic acids into protons and anion

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

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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

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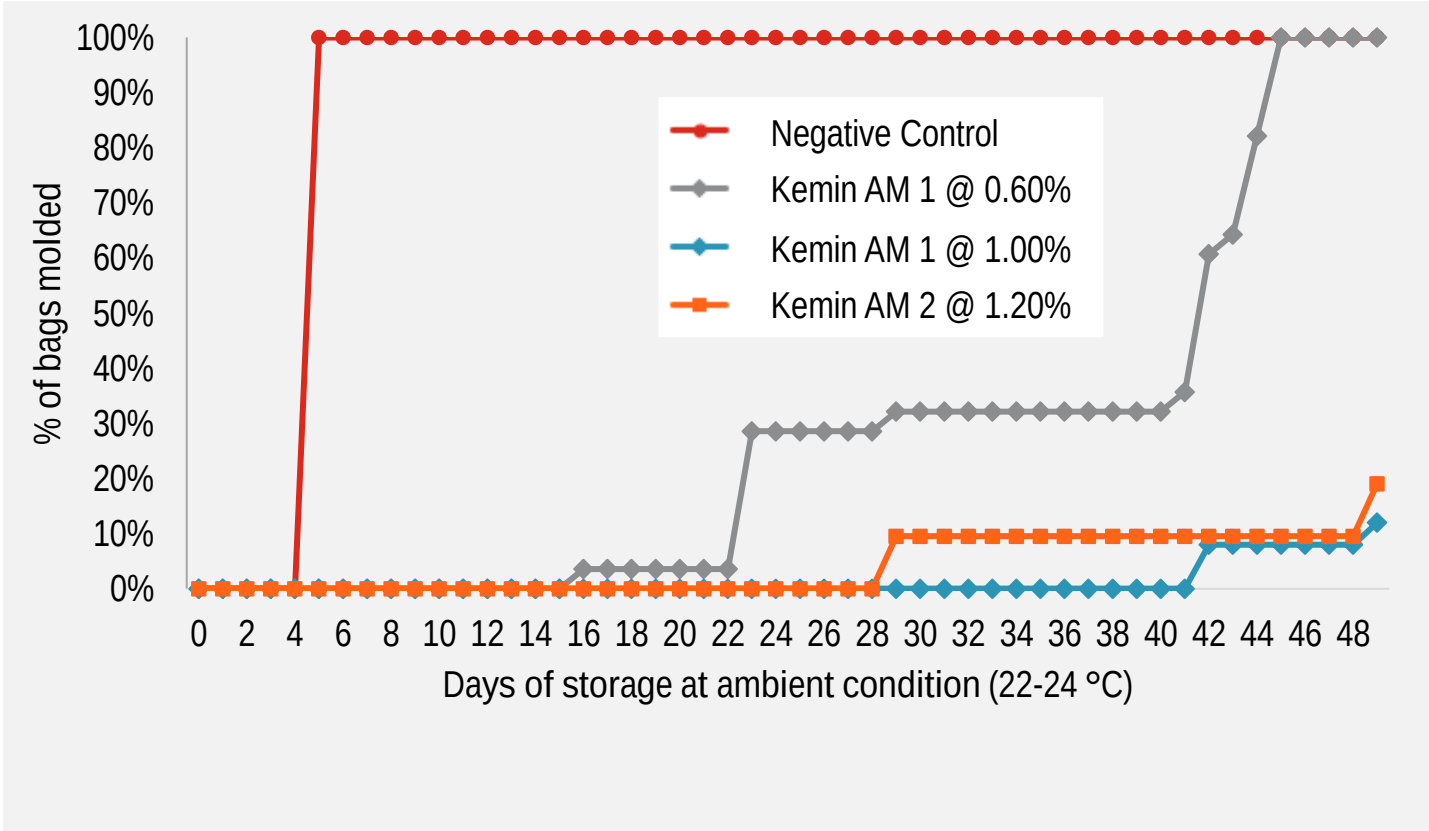
EFFICACY OF SYNTHETIC ANTIMICROBIALS

- 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.

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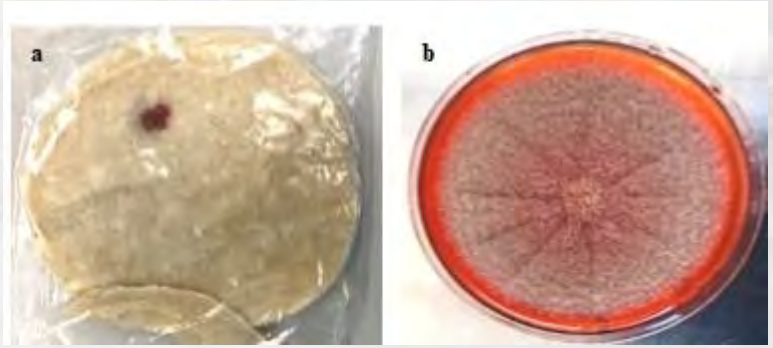
EFFICACY AGAINST RED MOLD (MONASCUS)

- 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		
		M. ruber	*M. pilosus*	*M. purpureus*
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

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CLEAN LABEL ANTIMICROBIALS

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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

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ACTIVE INGREDIENTS - FERMENTED PRODUCTS

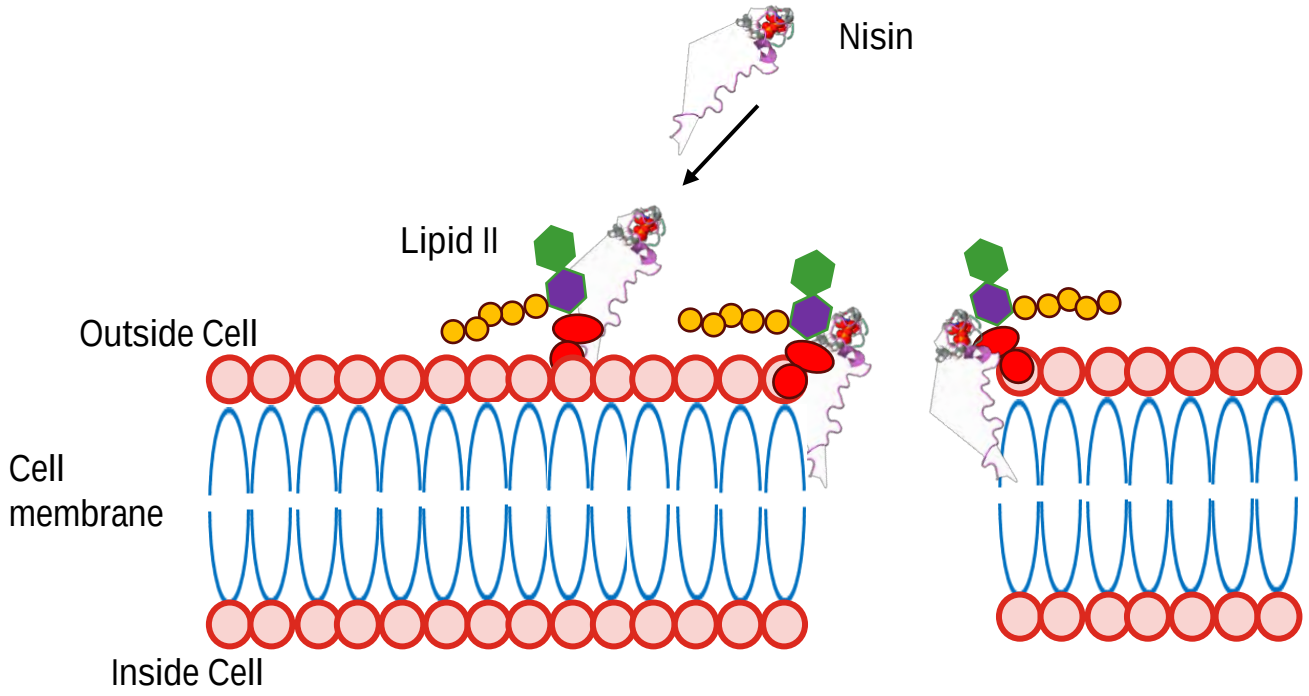
Typical actives include

- 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

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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)

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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

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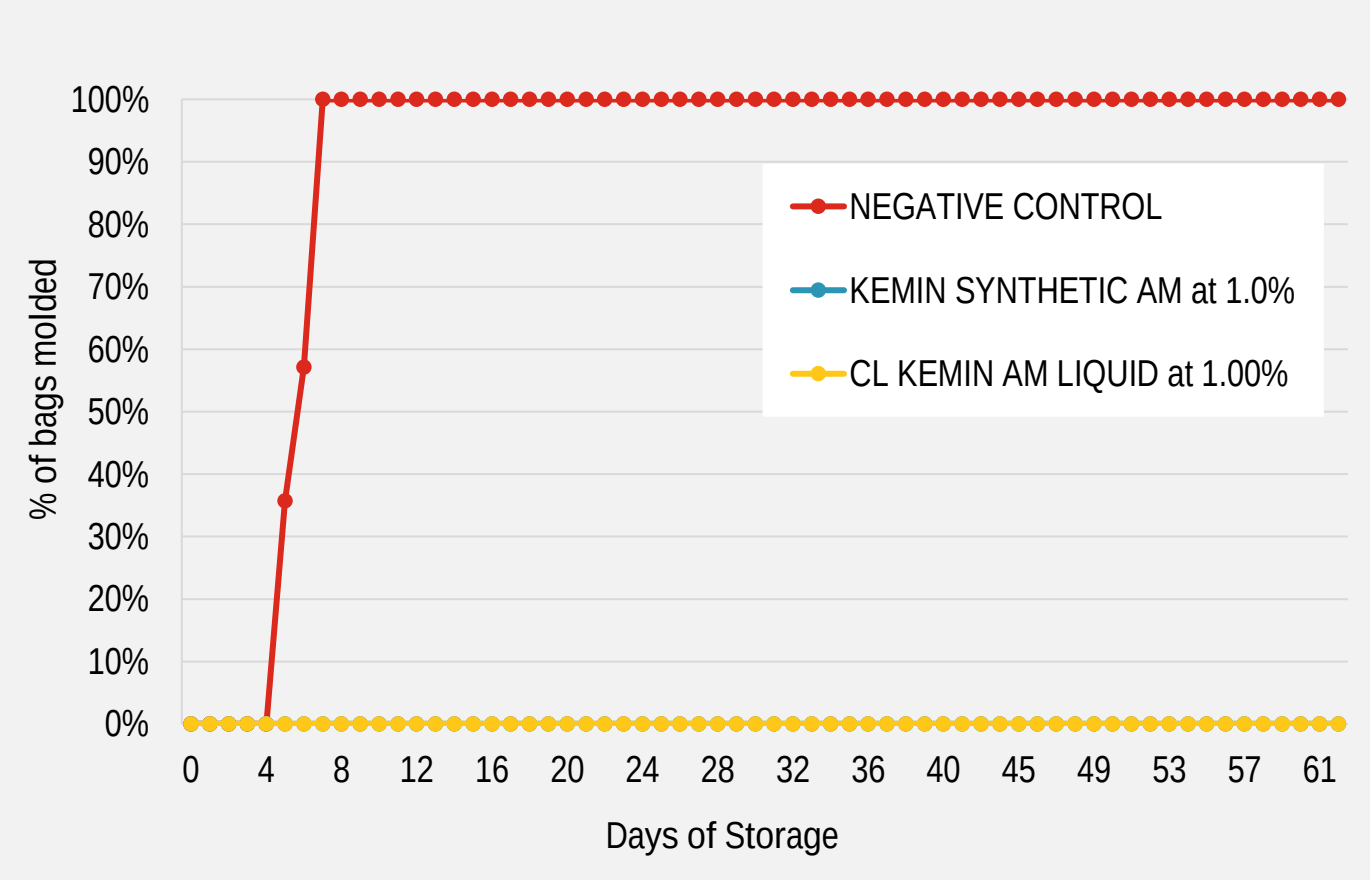
EFFICACY IN CORN TORTILLAS



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

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

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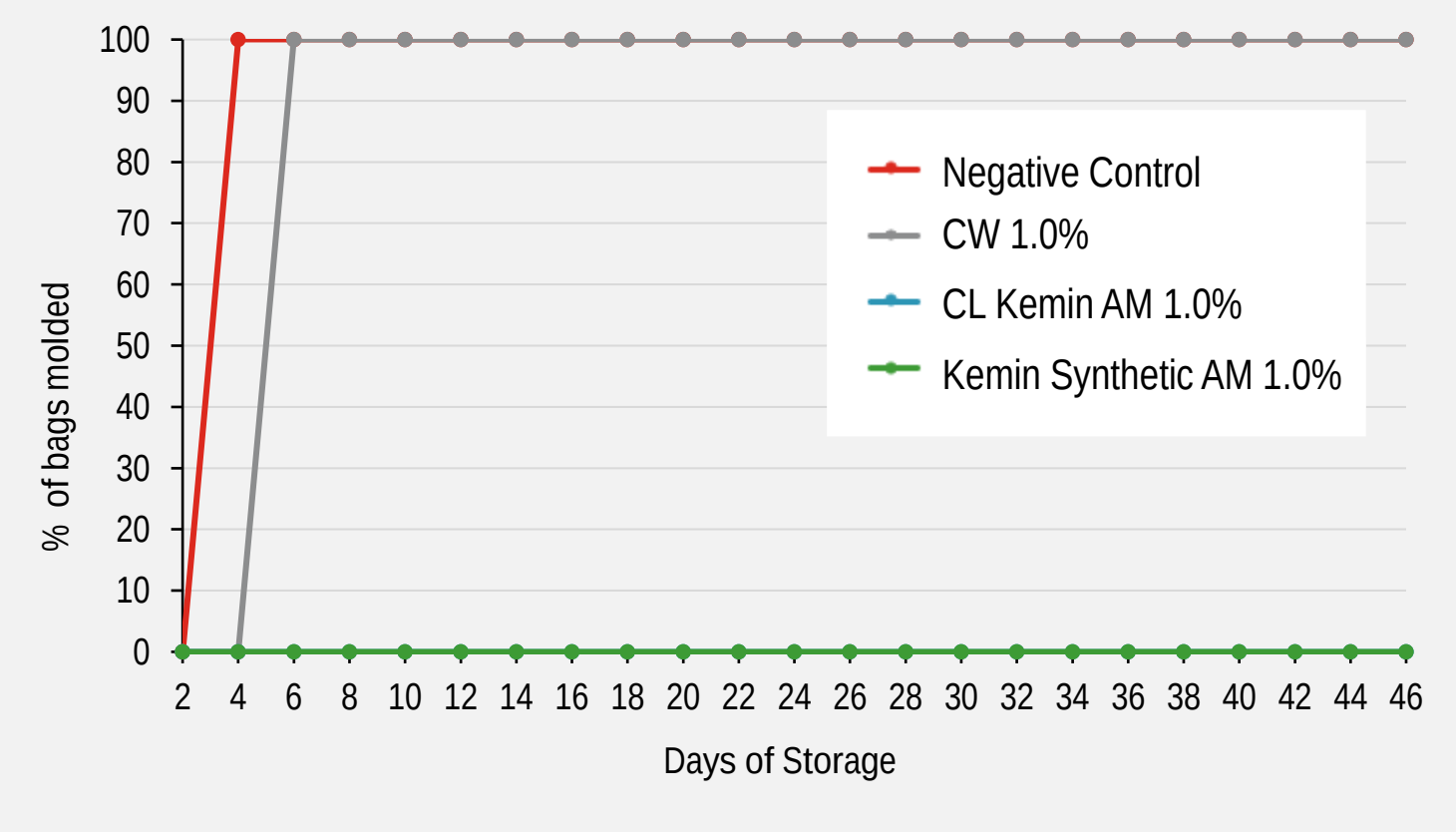
EFFICACY IN CORN TORTILLA



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

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

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

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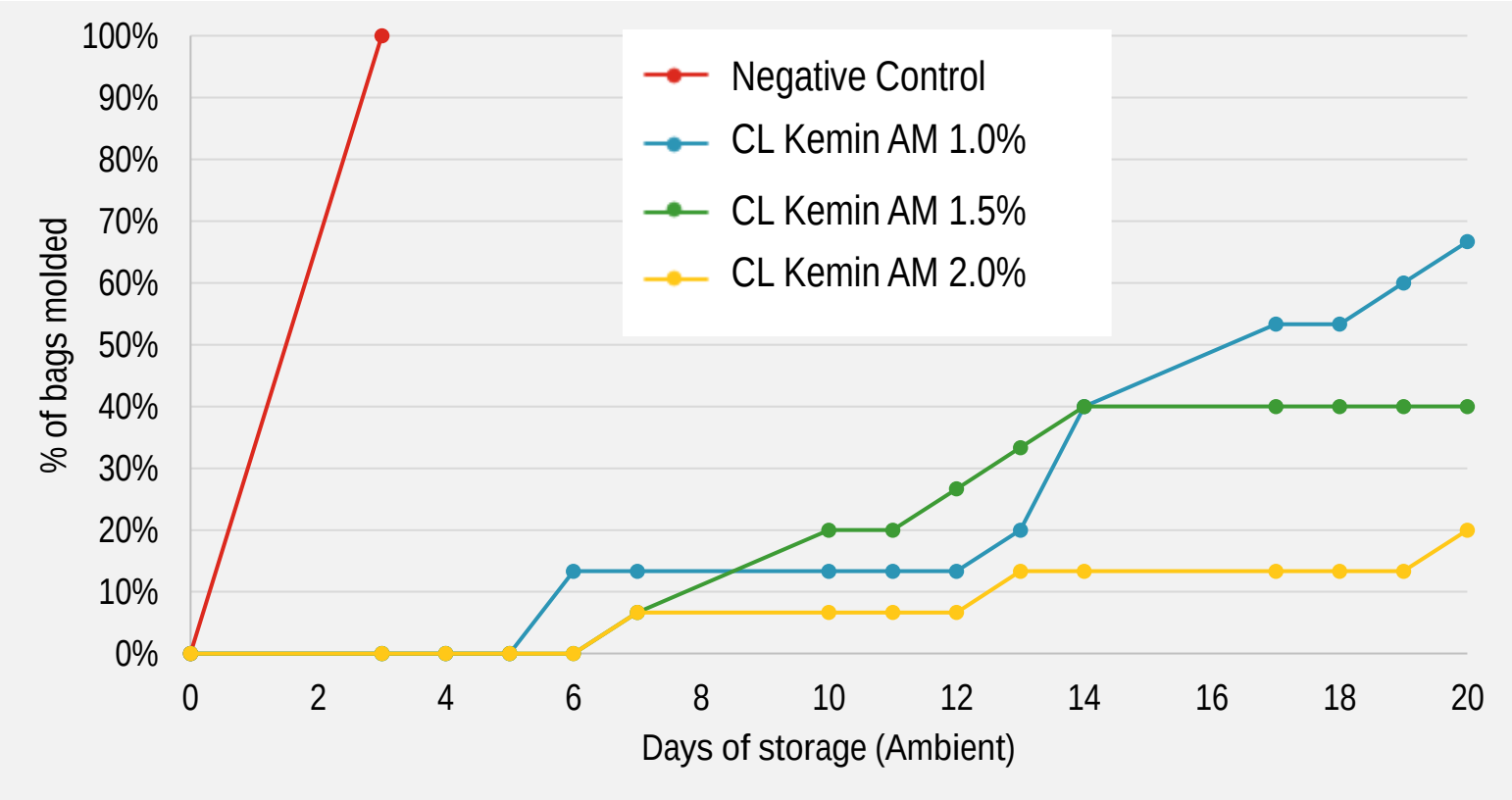
EFFICACY IN FLOUR TORTILLA



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.

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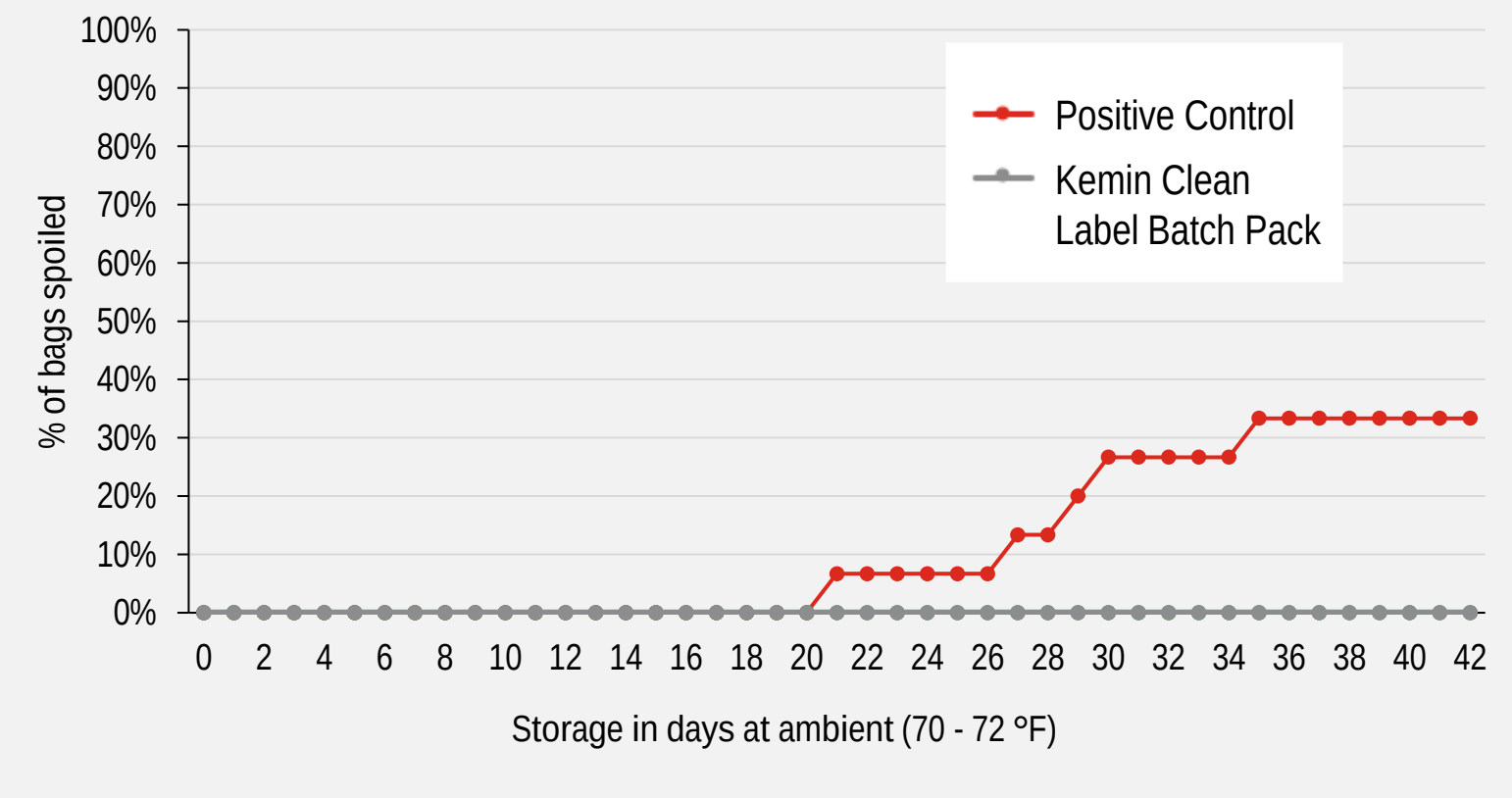
CLEAN LABEL BATCH PACKS - FLOUR TORTILLA



Positive control – Clean label batch pack

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

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CONCLUSIONS

- Clean label antimicrobials available – fermented products have limitations
- The available products vary hugely on efficacy, cost and sensory impact
- Efficient products with high active content, no sensory or textural impact are available
- Appropriate selection and testing is required to choose the most suitable product for your matrix