

A FINAL REPORT TO THE
AMERICAN MEAT INSTITUTE FOUNDATION

**ANTIMICROBIAL EFFECTS OF SURFACE
TREATMENTS AND INGREDIENTS ON CURED
RTE MEAT PRODUCTS**

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EXECUTIVE SUMMARY

Two frankfurters formulations were manufactured under commercial processing conditions to contain no KL (Control) or 3.3% KL. After cooking, chilling and peeling, each batch was divided into inoculated (four strain *Listeria monocytogenes* cocktail) and non-inoculated groups. After 60 min, each group was then treated 30 sec. with one of four different dips: Control (saline solution), acidified calcium sulfate (SWPA), 3.3% potassium lactate (KL) or 3.4% lactic acid (LA). The franks were vacuum packaged, stored under refrigeration (4.5°C) and evaluated at two-week intervals (0, 2, 4, 6, 8, 10, 12). Proximate composition, process yield, vacuum-package purge, a_w , residual nitrite, sodium content, insoluble components (calcium and phosphorus), pH, objective color, sensory evaluation and microbiological shelf-life (APCs) were determined on non-inoculated samples. *L. monocytogenes* counts were determined on inoculated frankfurters. SWPA and LA dips were effective at reducing *L. monocytogenes* on the surface of franks. A residual listericidal and listeristatic effect of the SWPA dip was observed on *L. monocytogenes* counts over storage. The most significant observations were that *L. monocytogenes* numbers were reduced by 5.8 logs on the surface of franks treated with SWPA dip and that after dip treatment *L. monocytogenes* counts remained at the minimum level of detection (1.7 logs) over the 12 weeks storage period. The addition of KL did not affect fat, protein, ash, process yield, sodium, calcium, phosphorus, vacuum-package purge, pH, a_w , objective color and lactate values; except that percent moisture was slightly lower. Proximate composition of frankfurters was not affected by dip treatments. Vacuum-package purge was slightly higher in samples treated with SWPA dip and pH of the SWPA franks was 0.83 units lower. Only slight changes in

surface and internal color were noted for the SWPA dip. A slight increase in calcium content of franks dipped with SWPA was detected. Descriptive attribute sensory panel results indicated little effect on the sensory properties of the frankfurters containing KL and dipped in antimicrobial solutions.

INTRODUCTION

Listeria monocytogenes is a foodborne pathogen of significant public health concern due to its virulence in susceptible individuals, and as a consequence has received a presidential mandate for reduction to decrease the incidence of foodborne illness. The estimated annual incidence of listeriosis in the United States is 1850 cases resulting in 425 deaths. Although foodborne listeriosis is rare, the associated mortality rate is as high as 20% among those at risk (FDA 2001). On May 5, 2000, President Clinton issued a directive to the Department of Health and Human Services (HHS) in cooperation with United States Department of Agriculture-Food Safety Inspection Service (USDA-FSIS) to conduct a risk assessment of *Listeria monocytogenes*. The Administration also proposed a goal of cutting the number of *Listeria* caused illness in half by year 2010. In response to the President's directive, the National Advisory Committee on Meat and Poultry Inspection (NACMPI) produced an issue paper recommending a revised action plan for control of *L. monocytogenes* to prevent food-borne listeriosis in meat and poultry products.

L. monocytogenes is a facultative, intracellular gram-positive, nonsporeforming and psychrotrophic bacterium that causes the disease called listeriosis. Immunocompromised individuals, infants, pregnant women and elderly persons are the most at risk. In humans, the primary manifestations of listeriosis are meningitis, abortion and prenatal septicemia (FDA 2001). The infectious dose of *L. monocytogenes* is unknown. It is an ubiquitous organism able to survive and multiply at refrigeration temperatures in the presence or absence of oxygen, and can tolerate a range of pHs and concentrations of up to 12-13% salt. Moreover, some strains may grow at a water activity

(a_w) as low as 0.9 and at a pH value as low as 4.4 (Walker and others 1990; Farber and Peterkin 1991, Miller 1992).

Ready-to-eat (RTE) products, such as hot dogs, lunchmeats, smoked fish, and certain types of soft cheeses, are among the foods most commonly associated with food-related listeriosis. Thus, a “zero tolerance” for *L. monocytogenes* in RTE foods has been specified by FDA based on the characteristics of this microorganism and the reported cases of listeriosis (Ryser and Marth, 1999). Contamination of RTE food products with *L. monocytogenes* primarily occurs post-processing and prior to consumption of these products. Even though cured RTE meat products contain sodium chloride and nitrite salts in their formulations that possess antimicrobial properties, they are not able to inhibit the growth of *L. monocytogenes* under refrigerated storage conditions (Mbandi and Shelef, 2002).

The safety of RTE meat products, which may be consumed without additional heat treatment, can be enhanced by adding substances to serve as microbiological hurdles and suppress the growth of *L. monocytogenes*. Such hurdles include pH lowering substances such as lactic acid and other organic compounds. Antilisterial effects of organic acids, their salts or combinations have been examined in several types of meat products. Shelef and Yang (1991) showed growth suppression of *L. monocytogenes* by lactate (4%) in sterile broth, and on chicken and beef. Chen and Shelef (1992) studied the relationship between water activity (a_w), salts of lactic acid, and growth of *L. monocytogenes* strain Scott A in a meat model system. They found that lactate concentrations less than 4% were not listeristatic, but combinations of 2 or 3% lactate with 2% NaCl inhibited the growth of *L. monocytogenes*. Sodium lactate (3 or 4%) was found to be effective against the growth

of *L. monocytogenes* in cooked beef stored at 10°C when compared to 0 or 2% (Miller and Acuff, 1994). Artificial contamination of frankfurters with *L. monocytogenes* followed by a 2 min dip in 1% lactic, acetic, tartaric, or citric acids resulted in a 1-2 log kill of the bacteria (Palumbo and Williams 1994). However, surviving bacteria recovered and began to grow during refrigerated storage. By dipping in 5% acetic or lactic acid, *L. monocytogenes* was not only killed, but also prevented from growing during 90 days of refrigerated storage. Mbandi and Shelef (2001) found enhanced inhibition of *L. monocytogenes* Scott A in sterile comminuted beef at 5 and 10°C using a combination of sodium lactate (2.5%) and sodium diacetate (0.2%). They also, evaluated the inhibitory effect of these salts alone and in combination in RTE meat inoculated with single strain or a cocktail of six strains of *Listeria*. These salts delayed growth of listeriae at 5°C and the effect of their combination was listericidal for *L. monocytogenes* Scott A and listeristatic for the six-strain mixture (Mbandi and Shelef, 2002).

Sodium and/or potassium lactate (Purasal®, PURAC America, Inc., Lincolnshire, IL) at levels of 2 to 4% have been shown to act as bacteriostatic agents against pathogenic bacteria such as *L. monocytogenes*, *E. coli* O157:H7 and *Salmonella* when incorporated into a variety of RTE meat products (Houstsma et al 1996; Murano and Rust 1995; Nerbrink and others 1999; Shelef 1994; Stekelenburg and Kant-Muermans, 2001). Sodium or potassium lactate is available commercially as a neutral aqueous solution (60%), and approved for use as a flavoring agent at levels of up to 4.8% in emulsified products (9 CFR, 424.21, 2002) such as frankfurters, bologna and wieners. Both may be used at concentrations up to 4.8% (or a concentration of 2.9% of a 100% solution) as a secondary ingredient to inhibit the growth of pathogenic bacteria in refrigerated, RTE,

hermetically packaged, cooked, uncured and cured meats. Therefore, the incorporation or a surface application of lactate could potentially afford protection against pathogen outgrowth in or on RTE products and provide additional protection to consumers.

Safe₂O™ (Mionix Corporation, Naperville, IL), an organic acid, calcium sulfate preservative, has been shown in preliminary tests to dramatically reduce the total numbers of aerobic bacteria on the surface of frankfurters. All Safe₂O™ ingredients are affirmed as GRAS (generally recognized as safe) under the FDA Code and their bacteriostatic effect is thought to be due to hydronium ions that inactivate bacterial membrane proton pumps. For these reasons, Safe₂O™ may have tremendous potential as an effective bacteriostatic preservative against pathogenic microorganisms such as *L. monocytogenes*. Thus, this preservative, when incorporated into or applied to the surface of RTE products, may afford a degree of protection against pathogens that has not been demonstrated by other products. The objective of this study was to test the effectiveness of preservative compounds applied to the surface of frankfurters immediately prior to packaging in concert with a selected preservative (potassium lactate) added as an ingredient. The outcome of this project would play an important role in reducing the risk of pathogen contamination in RTE products and provide a means of achieving the former President's Healthy People Goal by 2005 rather than by 2010.

OBJECTIVES

1. To determine the preservative properties of potassium lactate (KL) as an ingredient for preserving quality and extending the shelf-life of vacuum packaged, cured, ready-to-eat (RTE) frankfurters.

2. To evaluate the effectiveness of a surface application of either lactic acid (LA), Safe₂OTM + propionic acid (SWPA) or potassium lactate (KL) on frankfurters in combination with KL as an ingredient for suppressing the outgrowth of *Listeria monocytogenes*.
3. To evaluate the sensory, physical and chemical properties of vacuum-packaged frankfurters containing KL with a surface application of LA, SWPA or KL.

MATERIALS AND METHODS

Potassium lactate (3.3%, 60% concentration) solution was incorporated into 22.7 kg (50 lbs) of a standardized frankfurter formulation and compared to a control formulation without the ingredient. Each batch was divided into an inoculated (*L. monocytogenes*) and non-inoculated group. Then, each group was dipped into a saline solution (control), dilute Safe₂O™ with propionic acid (SWPA, 1:2 water pH = 1.64), 3.3% potassium lactate (KL, pH = 6.32) or 3.4% lactic acid (LA, pH = 2.16) (88% concentration), for 30 sec followed by vacuum packaging and storage under refrigeration (4.5°C). Evaluations were performed at two-week intervals (0, 2, 4, 6, 8, 10, 12) for 12 weeks. The following analytical procedures were performed on the treatments of non-inoculated samples: vacuum purge release, water activity, residual nitrite, salt content, total lactate, insoluble components (calcium and phosphate), pH, objective color, sensory evaluation and microbiological shelf-life (Aerobic Plate Counts). Some analyses were only performed one time per replication such as process yield, proximate analysis (percentage of moisture, fat, protein and ash), salt content and insoluble components (calcium and phosphorus). Residual nitrite was determined three times per replicate after 0, 8 and 12 weeks of storage, respectively. *L. monocytogenes* counts were determined on inoculated frankfurters at each storage period. A total of two replications were performed for this study.

Frankfurter Preparation

Frankfurters were prepared in a state inspected (Texas Department of Health), commercial-scale pilot plant located in the Rosenthal Meat Science and Technology Center at Texas A&M University. Fresh and/or frozen lean beef trimmings and pork fat

trimmings (-2° to 3°C) were selected, coarse ground through a 1.27 cm (1/2”) plate, reground through a 0.48 cm (3/16”) plate, analyzed for fat content and formulated to achieve a 20% fat endpoint. A base formulation of raw materials, dry ingredients and water are shown in Table 1 and represent calculations to yield a finished product with the following specifications: 70 to 75% meat block, ~28% ether extractable fat based on the meat block or 20% fat based on the finished batch weight, 56 to 58% moisture, pH 6.0 to 6.3, 2.25 to 2.5% salt (sodium chloride), 0.35 to 0.5% sodium tripolyphosphate, and <156 mg/kg (ppm) of sodium nitrite (calculated on the meat component). An automated, staged standard processing cycle for frankfurters was selected to achieve an endpoint temperature of 71.1°C (160°F) in a commercial processing oven.

Table 1. Frankfurter formulation ingredients calculated on a raw batch weigh basis

Ingredients	Formulation Treatments	
	Control (%)	Potassium Lactate (%)
Meat Trimmings	74.1	71.7
Lean beef trim (85/15)	38.4	37.2
Pork fat trim (60/40)	35.7	34.6
Non-meat Ingredients	25.9	28.3
Salt*	1.66	1.61
Potassium Lactate (as specified)	-	3.3
Corn Syrup Solids (DE 42)**	1.48	1.43
HMP or HVP	0.74	0.72
Hydrolyzed Beef Stock	0.37	0.36
Sodium Tripolyphosphate	0.33	0.32
Spice/Seasoning	0.37	0.36
Sodium Erythorbate	0.037	0.036
Sodium Nitrite (cure blend)***	0.185	0.179
Added Water	13.3	12.9
10% Added Water (Cook Shrink)	7.4	7.2
Total (Batter)	100.0	100.0

*For each percent sodium lactate, sodium chloride was reduced 0.1%, for example: total NaCl was reduced to 2.05% with the addition of 2.0% K Lactate (100% basis) or 3.3% K Lactate (60% basis).

**DE = Dextrose Equivalent

***Cure blend contains 6.25% sodium nitrite bonded to 93.75% salt. Pure nitrite, if used, would be added at 0.011% while the salt would be increased to 1.84%.

Analytical Techniques

Process Yield

The percentage of process yield was determined, by dividing the cooked product weight by the raw uncooked weight product and multiplying by 100, using the following equation:

$$\text{Percent Process Yield} = \frac{\text{Finished Product Weight}}{\text{Raw Product Weight}} \times 100$$

Vacuum Purge Release

The percentage of vacuum purge released during storage was determined on vacuum packaged product (four franks/package from each treatment) at 14-day intervals, over a 12 week refrigerated (4°C) storage period. Each package was weighed to obtain a total package weight and opened. The contents (franks and package) were hand dried with paper towels and reweighed. Finally, the percent of vacuum purge release was determined using the following equation:

$$\text{Percent Vacuum Package Purge} = \frac{\text{Purge Weight}}{\text{Product Weight} + \text{Purge Weight}} \times 100$$

Proximate Analysis

Percentages of moisture, fat, protein and ash were determined on the cooked frankfurters according to AOAC (2000) procedures. Eight franks per treatment were homogenized in a food processor (Cuisinart Inc., Model DLC-8M, Norwich, CT) before sampling. Proximate analysis was performed on frankfurters assigned to the week 0 and stored (-20°C) under frozen conditions until the day of analysis. Moisture content (%)

was determined by microwave drying using the CEM SMART TRAC™ System 5 moisture analyzer (CEM Corporation, Matthews, NC). Fat content (%) was determined by methylene chloride extraction of the dried samples (CEM fat analyzer, Model FAS-9001, CEM Corporation, Matthews, NC). Crude protein percentage was determined by Dumas sample combustion method to release gaseous N₂ in a Leco FP-528 Protein Analyzer (St. Joseph, MO). The procedure was standardized using ethylenediamine tetraacetic acid (EDTA) (Leco Lot # 1030, %N = 9.56 ± 0.03) and Orchard leaves (Leco Lot # 1005, %N = 2.55 ± 0.05). Percent crude protein was calculated as 6.25 times the percent nitrogen. Ash content was determined by the muffle furnace method. Analysis of the samples was performed in duplicate.

Water Activity

Water activity (a_w) values were determined using an Aqua Lab™ (model series 3, Decagon Devices Inc., Washington, USA). Eight franks per treatment were taken at each test week (0, 2, 4, 6, 8, 10, 12) and homogenized in a food processor (Cuisinart Inc., Model DLC-8M, Norwich, CT) before sampling. Approximately, 8 grams of homogenized sample were spread evenly on the bottom of an Aqua Lab sample cup, positioned inside the vapor chamber, and a reading was obtained after 3-5 min of equilibration. The a_w of each treatment was performed by triplicate over the 12 week storage period.

Residual Nitrite

Residual nitrite values of each treatment and at storage weeks 0, 8 and 12 were determined by a colorimetric method according to AOAC (2000) procedures. Residual

nitrite determinations were performed in duplicate and the results were reported as ppm (mg/kg).

Salt Content

Salt content was determined by potentiometry using an ion specific electrode (Model 86-11 combination electrode for sodium, Orion Research Inc., Beverly, MA) calibrated with dilutions of a 1000 ppm sodium standard (No 841108, Orion Research Inc., Beverly, MA), and a digital pH meter (Model 720A, Orion Research Inc., Beverly, MA). Determinations of salt content were performed on frankfurters assigned to week 0 and stored (-20°C) under frozen conditions until the day of analysis. Ten grams of thawed sample were homogenized for 30 sec. with 240 ml of distilled water (1:25 dilution) using a Waring® blender (Model 31BL92, Waring Products Division, Dynamics Corp. of America, New Hartford, CT). Finally, a 50 ml aliquot of the slurry was combined with 5 ml of sodium ionic strength adjustor (ISA, No 841111, Orion Research Inc., Beverly, MA), with continuous stirring, and the sodium electrode inserted into the slurry for a reading. A standard curve was developed with anchor points at 100 (minimum) and 1000 ppm (maximum) sodium Na⁺ standard (1000 ppm, No 841108, Orion Research Inc., Beverly, MA). Measurements were taken on duplicate slurry homogenates and the results were reported as percentage of sodium ion.

Total Lactate

Total lactate at each designated test week was determined by an enzymatic and colorimetric method using a Sigma Diagnostics Kit (Procedure No 500 Sigma Diagnostics, Inc., St. Louis, MO). Seven grams of sample were homogenized at three intermittent times for 30 sec. each with 35 ml of distilled water (1:5 dilution) using a

Waring® blender (Model 31BL92, Waring Products Division, Dynamics Corp. of America, New Hartford, CT). The slurry was placed in a plastic centrifuge tube and centrifuged at 30,000 x g for 15 min using a Beckman Avanti® J-25 centrifuge (Beckman Instruments, Inc., Palo Alto, CA). One milliliter aliquots were placed in 1.7 ml micro-centrifuge tubes (No 20170-610, VWR Scientific Products, West Chester, PA) and enzymatic and colorimetric procedures performed in duplicate. Absorbance readings were taken at 540 nm using a Cary 300 Bio UV-visible double-beam spectrophotometer (Varian, Optical Spectroscopy Instruments, Victoria, Australia). Results were reported as mg of lactate per gram of tissue.

Insoluble Components

Calcium content was determined by potentiometry using an ion specific electrode (Model 93-20 electrode for calcium, Orion Research Inc., Beverly, MA) attached to a pH meter (Model 720A, Orion Research Inc., Beverly, MA) and calibrated with 0.1 M Ca²⁺ standard (No 922006, Orion Research Inc., Beverly, MA). Thirty grams of homogenized sample was blended with 270 ml of double distilled water (1:10 dilution) for 30 sec in a Waring® Blendor (Model 31BL92, Waring® Products Division, Dynamics Corp. of America, New Hartford, CT). The slurry was filtered through cheesecloth mini-wipes (VWR Scientific Products, West Chester, PA) to remove particulates. Finally, 50 ml of slurry were combined with 1 ml of sodium ionic strength adjustor (ISA, No 841111, Orion Research Inc., Beverly, MA), stirred thoroughly, and the calcium and reference electrodes inserted into the slurry for a reading. A standard curve was developed with anchor points at 1 (minimum) and 10 ppm (maximum) of 0.1 M Ca²⁺ standard (No

922006, Orion Research Inc., Beverly, MA). Duplicate measurements were taken on slurried homogenates and the results reported as mg of calcium per 100 g of tissue.

Phosphorus content was determined in duplicate by the Inductively Coupled Plasma Method (Atomic Emission Spectrophotometry) at ABC Labs, Gainesville, FL. Duplicate measurements were taken on slurried homogenates and results reported as mg of phosphorus per 100 g of edible portion.

pH Determination

pH measurements of franks from each treatment were determined by the slurry method adapted for meat products utilizing an OrionTM (model 720A, Orion Research Inc., Beverly, MA) pH meter standardized with pH 4 and 7 buffers and fitted with a combination electrode. Thirty grams of homogenized sample was blended with 90 ml of double distilled water for 2 min in a Waring® Blendor (Model 31BL92, Waring® Products Division, Dynamics Corp. of America, New Hartford, CT) and the pH electrode inserted into the stirred slurry for a reading.

Objective Color

L*, a*, b* color space values of the frankfurter treatments were obtained by reflectance using a Minolta Colorimeter (model CR-200, Minolta Co., Ramsey, NJ), calibrated to a white tile standard surface (L* = 97.55, a* = -0.02, b* = 1.56) at channel 00. Two frankfurters per treatment at each test week were sliced in half longitudinally, the open face of the frankfurters covered with clean Saran® wrap, and random readings from the surface and inside of the franks taken at three locations. The results were expressed as L* (lightness), a* (redness) and b* (yellowness) values.

Sensory Evaluation

Frankfurter samples from each treatment (at each designated test week) were evaluated by a trained descriptive attribute sensory panel (4-7 members) at the Texas A&M Sensory Testing Facility. The panel was selected and trained according to procedures of Cross and others (1978), Meilgaard and others (1991) and AMSA (1995) guidelines. Training prior to testing was conducted by presenting reference samples to the panel to characterize the basic attributes of franks with the different treatments used in the study. The samples were evaluated for aromatics (overall meat flavor, fatty, smoke, spice complex, cardboard, painty, fishy, livery, caramelized, soured, soapy, musty and vinegar), feeling factors (astringent and metallic), basic tastes (salt, sour, bitter and sweet), aftertastes (fat mouthfeel, sour, spice, bitter, metallic, sweet, salty, vinegar and smoke) and texture (springiness, juiciness, hardness and cohesiveness of mass) using a 16-point Spectrum Universal intensity scale (Meilgaard and others, 1991) where 0 = absence of an attribute and 15 = extremely intense.

Samples of refrigerated franks from each treatment were evaluated at two weeks intervals (0, 2, 4, 6, 8, 10, 12) for twelve weeks. On each testing day, eight samples were evaluated per day during one session with four samples being served five minutes apart. The order of the treatments was randomized and a warm-up was presented to judges prior to sample evaluation to ensure that they had identified the treatment attribute to be tasted. The stimuli used for warm-up were franks from a control formulation with a dip containing saline solution (control). Samples of refrigerated precooked franks were steeped in boiling water for seven minutes. Three, warm cross-cut pieces (1.5 cm) from each treatment, randomly codified with three-digit codes, were served to each panelist.

Judges were seated in separated booths to avoid communication during the evaluation, and the samples were presented to panelists through stainless steel hoods adjacent to the preparation area. Testing was conducted under red filtered, incandescent lighting to mask color. Distilled water, unsalted crackers and ricotta cheese were given to judges to cleanse their mouths.

Subjective color of the surface and interior of longitudinally sliced franks were evaluated by the sensory panel. The uncooked franks were sliced in half longitudinally and presented to panelists under white incandescent lighting (Sylvania light bulbs incandescent, 150 W and 120 V., Sylvania Electronics, Danvers, MA) to simulate store conditions. Subjective color was determined using a 7-point scale for surface (1 = very dark reddish-brown; 7 = very pale pink) and interior views (7 = no cured color-gray; 1 = extremely pink color) of the samples in accordance with AMSA (1995) procedures.

Microbiological Analysis

Frankfurters (containing either no KL or 3.3% KL) were divided into inoculated and non-inoculated groups after the cooking, chilling and peeling operations. Designated samples were surface inoculated with a *Listeria monocytogenes* mixture (cocktail) and then treated with four different dip solutions. For microbiological shelf-life determinations of non-inoculated samples, Aerobic Plate Counts (APCs) were performed on the treatments (franks containing either no KL or 3.3% KL and treated with four different dip solutions).

Bacterial cultures and inoculum Preparation

Four strains of *L. monocytogenes* (ATCC 15313, 51414, 43256 and 49594) were used in this study. Cultures were grown in tryptic soy broth (TSB, Difco Laboratories, Detroit, MI) overnight at 35°C and then pooled immediately prior to use as an inoculum.

Twenty-one franks per treatment were transferred into sterile tubs and surface inoculated with 0.1ml of the *L. monocytogenes* mixture to give a final concentration of approximately 10^8 CFU/ml. One hour post inoculation, the inoculated samples were submerged in a dip containing saline solution (Control), SWPA, KL or LA for 30 sec. Samples then were placed in Cryovac® 10.16 cm x 30.48 cm (4 in x 12 in) bags (Type B540T, Cryovac® North America, Duncan, SC), vacuum packaged (KOCH Inc., Model X180, Germany), stored at 4.5°C (40°F) and evaluated at two-week intervals (0, 2, 4, 6, 8, 10 and 12) for twelve weeks.

Listeria monocytogenes

At each designated test interval, inoculated samples were analyzed in duplicate for recovery and enumeration of *L. monocytogenes*. One frank was aseptically removed from the package, transferred into a stomacher bag with 99 ml of 0.1% peptone water (Difco Laboratories, Detroit, MI) and pummeled by hand for 1 min. Decimal dilutions were prepared with 9 ml of 0.1% peptone water and surface plated on to Modified Oxford Medium (MOX) (Difco Laboratories, Detroit, MI). Thus, an aliquot (0.1ml) was placed on the agar plate and then uniformly spread on the agar surface with a sterile bent glass rod. Typical colonies were counted after incubation of the plates at 37°C for 48 h and then recorded as \log_{10} CFU/Frank.

Aerobic Plate Count

To evaluate microbiological shelf-life, Aerobic Plate Counts (APCs) were performed on the treatments for non-inoculated samples at each designated test week. One frank was aseptically removed from the packaging material, transferred into a stomacher bag with 99 ml of 0.1% peptone water and the mixture pummeled by hand for 1 min. Decimal dilutions were prepared and 1 ml of the appropriate dilution was placed onto Petrifilm™ Aerobic Count Plates (3 M, St. Paul, MN). Typical colonies were counted after incubation of plates at 25°C for 48 h, and then recorded as log₁₀ CFU/Frank.

Statistical Analysis

Data were analyzed as a split-plot design using the General Linear Model (GLM) procedures of the Statistical Analysis System (SAS, 1995). Analysis of variance was used to determine statistical differences among the main effects and their interactions with a significance level of $P < 0.05$. Least square means were used to identify significant treatment effects. For microbiological analysis, count data were transformed into base 10 logarithms. *L. monocytogenes* Log₁₀ reduction values were calculated by subtracting the Log₁₀ count of positive control and the Log₁₀ count of each treatment on inoculated frank surface. The experiment was replicated two times.

RESULTS AND DISCUSSION

Physical and Chemical Evaluations of frankfurters

Percentages of moisture, fat, protein and ash are shown in Table 2. No differences ($P > 0.05$) were observed in these variables between the control and the potassium lactate (KL) treatment, except that percent moisture was slightly lower in franks with KL. In general, the addition of KL did not change the chemical composition of the frankfurters. Likewise, no differences ($P > 0.05$) were observed in the percent moisture, fat, protein and ash of franks treated with different dips.

Results for percent process yield, percent sodium, calcium and phosphorus contents are presented in Table 3. Process yield and the sodium, calcium and phosphorus content of the franks were not affected ($P > 0.05$) by the addition of KL. However, Safe₂O™ (SWPA) increased ($P < 0.05$) calcium content by 2.46 mg/100g over the control. This was likely caused by the SWPA material which contains a calcium sulfate complex. There was also an ingredient treatment by dip interaction for calcium (Table 4) that showed calcium levels to increase by 2.88 and 2.04 mg/100 g in frankfurters containing either no KL or 3.3% KL, respectively, and dipped with SWPA. The increase in calcium content of the frankfurters could enhance their nutritional value through calcium enrichment.

As shown in Table 5, the vacuum-package purge, pH, objective color values (surface and internal) and lactate and residual nitrite content were not affected ($P > 0.05$) by the addition of KL. However, the water activity (a_w) was slightly lower ($P < 0.05$) in franks with KL. Vacuum-package purge (Table 5) was slightly higher (0.68%) in franks treated with SWPA dip when compared to ($P < 0.05$) the control or other dip treatments. Franks treated

Table 2. Least squares means for percent moisture, fat, protein and ash of frankfurters containing KL and dipped in antimicrobial solutions.

	Percent Moisture	Percent Fat	Percent Protein	Percent Ash
<u>Ingredient Treatment¹</u>				
Control	59.29 ^a	19.34 ^a	14.10 ^a	2.06 ^a
KL	57.91 ^b	21.32 ^a	13.12 ^a	2.55 ^a
Std. Error	0.07	0.41	0.12	0.08
<u>Dip Treatment²</u>				
Control	58.93 ^a	20.63 ^a	13.57 ^a	2.34 ^a
SWPA	58.39 ^a	20.09 ^a	13.58 ^a	2.24 ^a
KL	58.73 ^a	20.04 ^a	13.66 ^a	2.30 ^a
LA	58.34 ^a	20.56 ^a	13.60 ^a	2.34 ^a
Std. Error	0.27	0.24	0.08	0.09

^{ab} Means within a column per main effect with different superscript letters are significantly different ($P < 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

²Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

Table 3. Least squares means for percent process yield and sodium, calcium and phosphorus content of frankfurters containing KL and dipped in antimicrobial solutions.

	Percent Process Yield	Percent Sodium	Calcium (mg/100g)	Phosphorus (mg/100g)
<u>Ingredient Treatment¹</u>				
Control	90.20 ^a	0.77 ^a	4.15 ^a	209.63 ^a
KL	90.66 ^a	0.75 ^a	4.23 ^a	198.81 ^a
Std. Error ³		0.005	0.15	1.28
<u>Dip Treatment²</u>				
Control		0.78 ^a	3.48 ^{ab}	206.00 ^a
SWPA		0.75 ^a	5.94 ^c	206.25 ^a
KL		0.76 ^a	3.67 ^{ab}	203.00 ^a
LA		0.76 ^a	3.68 ^{bc}	201.63 ^a
Std.Error		0.01	0.05	1.80

^{abc} Means within a column per main effect with different superscript letters are significantly different ($P < 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

²Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

³Std Error LSMean for Process yield (%) - control = 0.33, KL = 0.35

Table 4. Least squares means for ingredient by dip interaction for calcium values

	<u>Ingredient Treatment</u> ¹	
	Control	KL
Dip ²		
Control	3.12 ^{ac}	3.84 ^{bc}
SWPA	6.00 ^{ad}	5.88 ^{ad}
KL	3.68 ^{ae}	3.67 ^{ae}
LA	3.82 ^{ae}	3.55 ^{bc}
Std Error	0.07	0.07

^{ab} Means with the same superscript letters across row are not different ($P > 0.05$).

^{cde} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution

²Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

with KL dip had the lowest percent vacuum-package purge or about 0.3% less than the control. No differences ($P > 0.05$) were observed between the control and LA dip.

The pH decreased ($P < 0.05$) by 0.83 units (Table 5) due to application of the SWPA dip while the LA treatment reduced the pH 0.12 unit less than the control. Moreover, no differences in pH were found in franks treated with KL when compared to control. The pH lowering effects of SWPA, as well as LA, were due to their naturally low acidity (pH = 1.64 and 2.16, respectively). Reductions in vacuum-package purge were also likely due to the increased acidity of the dips.

Among the dip treatments, only the franks treated with SWPA were slightly lower ($P < 0.05$) in a_w (Table 5).

Results for objective color (surface and internal) values are presented in Table 5. The addition of KL did not change the surface nor internal (L^* , a^* , b^*) color attributes of the franks. However, surface L^* (lightness) values of SWPA and LA dips were slightly lower ($P < 0.05$) indicating a darker frank when compared to the control. The KL dip had no effect on surface L^* value. Surface redness (a^*) increased slightly with the SWPA dip, but no differences ($P > 0.05$) were observed among the control, KL and LA treatments. Surface b^* (yellowness) values were not different ($P > 0.05$) among dip treatments.

Internal a^* values or redness (Table 5) were slightly lower ($P < 0.05$) in franks treated with KL and LA, but not different for the SWPA treatment. Franks treated with SWPA had lower b^* values (less yellow) than the control, but no differences ($P > 0.05$) in internal L^* values were observed among the other dip treatments. It can be concluded that the SWPA treatment made the surface of the franks slightly darker (lower L^* values), increased surface redness (higher a^* values) and decreased internal yellowness (lower b^* values). Although

Table 5. Least squares means for percent vacuum-package purge, pH, a_w , surface color values, internal color values, and lactate and residual nitrite content of frankfurters containing KL and dipped in antimicrobial solutions.

Ingredient Treatment ¹	Percent Vacuum-package Purge	pH	a_w	Surface Color Values			Internal Color Values			Lactate (mg/g tissue)	Residual Nitrite (ppm)
				L*	a*	b*	L*	a*	b*		
Control	1.84 ^a	6.05 ^a	0.978 ^a	63.45 ^a	18.69 ^a	13.21 ^a	68.30 ^a	15.53 ^a	9.81 ^a	4.56 ^a	1.55 ^a
KL	1.98 ^a	6.09 ^a	0.973 ^b	63.44 ^a	17.85 ^a	12.92 ^a	67.75 ^a	15.00 ^a	9.64 ^a	11.60 ^a	1.36 ^a
Std. Error	0.03	0.008	0.0003	0.46	0.23	0.41	0.84	0.06	0.28	1.16	0.13
<u>Dip Treatment²</u>											
Control	1.82 ^b	6.32 ^a	0.977 ^b	63.99 ^b	18.10 ^a	13.19 ^a	67.73 ^a	15.64 ^b	10.00 ^a	7.36 ^a	1.75 ^b
SWPA	2.48 ^a	5.49 ^c	0.975 ^a	62.70 ^a	19.10 ^b	13.06 ^a	68.32 ^a	15.40 ^{ab}	9.33 ^b	9.23 ^a	0.59 ^a
KL	1.52 ^c	6.26 ^{ab}	0.977 ^b	63.95 ^b	17.89 ^a	13.00 ^a	68.04 ^a	15.08 ^a	9.79 ^a	7.69 ^a	1.83 ^b
LA	1.80 ^b	6.20 ^b	0.976 ^{ab}	63.15 ^a	17.98 ^a	13.01 ^a	68.02 ^a	14.96 ^a	9.78 ^a	8.03 ^a	1.66 ^b
Std. Error	0.06	0.02	0.0004	0.19	0.16	0.14	0.19	0.13	0.09	0.71	0.08

^{abc} Means within a column per main effect with different superscript letters are significantly different ($P < 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

²Control = Saline solution; SWPA = SafeOTM with propionic acid; KL = Potassium lactate; LA = Lactic acid.

these differences were statistically significant, the color space value differences were of such small magnitude that they would not likely be detectable by consumers.

Lactate levels (Table 5) in the frankfurters were not affected ($P > 0.05$) by dip treatments in this study.

Residual nitrite of the SWPA dip was not affected ($P > 0.05$) by the addition of KL (Table 5). However, residual nitrite was lower ($P < 0.05$) by 1.16 ppm when compared to the control. No differences in residual nitrite contents were found in franks treated with KL or LA.

Storage had a small effect on pH, a_w , objective color (surface and internal), lactate concentration and residual nitrite values (Table 6). However, there was no change in percent vacuum-package purge due to storage under refrigerated conditions for 12 weeks.

The pH increased ($P < 0.05$) slightly during the 6th and 8th weeks of storage but returned to initial levels on weeks 10 and 12. Ingredient treatment (KL) by storage week and dip treatment by storage week interactions for pH are shown in Table 7 and 8. In general, the control pH decreased slightly with storage (Table 7) after week 10. Also, the inclusion of KL increased pH slightly in the frankfurters after week 10. According to dip by week interaction (Table 8), the application of the SWPA and LA dips decreased ($P < 0.05$) pH values in frankfurters when compared to the other dip treatments.

Water activity (a_w) increased slightly as storage progressed (Table 6) with the lowest a_w values observed at week 0. Moreover, there were ingredient by storage week (Table 9) and ingredient by dip by storage week (data not shown) interactions for a_w . Inclusion of KL as an ingredient (Table 9) kept the a_w lower throughout 12 weeks of simulated retail storage at 4.5°C, thus tending to provide some inhibition to pathogen growth. In general, a_w gradually

Table 6. Least squares means for percent vacuum-package purge, pH, a_w , surface color values, internal color values, lactate and residual nitrite of frankfurters stored under refrigerated conditions.

Storage Week	Percent Vacuum-package Purge	pH	a_w	Surface Color Values			Internal Color Values			Lactate (mg/g tissue)	Residual Nitrite (ppm)
				L*	a*	b*	L*	a*	b*		
				Week 0		6.03 ^a	0.974 ^a	62.23 ^a	18.82 ^b		
Week 2	1.93 ^a	6.04 ^a	0.975 ^b	63.66 ^{ab}	18.43 ^b	12.21 ^a	68.38 ^b	15.75 ^c	8.55 ^a	6.98 ^{ab}	
Week 4	1.91 ^a	6.08 ^{ab}	0.976 ^{bc}	63.81 ^b	18.83 ^b	13.07 ^{ab}	68.39 ^b	15.81 ^c	9.79 ^{bc}	9.14 ^{bc}	
Week 6	1.88 ^a	6.11 ^b	0.976 ^{bcd}	62.68 ^{ab}	18.80 ^b	13.15 ^b	67.82 ^{ab}	15.52 ^{bc}	9.85 ^{bc}	6.33 ^a	
Week 8	2.04 ^a	6.13 ^b	0.976 ^{bcd}	66.01 ^c	16.86 ^a	12.27 ^a	69.62 ^c	14.72 ^{ab}	9.38 ^b	8.10 ^{abc}	0.86 ^a
Week 10	1.91 ^a	6.04 ^a	0.977 ^{bcd}	62.45 ^{ab}	17.84 ^{ab}	13.64 ^{bc}	66.97 ^a	14.68 ^a	10.30 ^{cd}	7.92 ^{ab}	
Week 12	1.77 ^a	6.05 ^a	0.977 ^d	63.28 ^{ab}	18.31 ^b	14.02 ^c	68.18 ^b	14.76 ^{ab}	10.47 ^d	10.14 ^c	0.55 ^a
Std. Error	0.08	0.02	0.0004	0.52	0.36	0.30	0.36	0.29	0.19	0.76	

^{abcd} Means within a column with different superscript letters are significantly different ($P < 0.05$).

Table 7. Least squares means for ingredient by storage week interaction for pH values

	<u>Ingredient Treatment¹</u>	
	Control	KL
Storage Week		
Week 0	6.06 ^{ade}	6.00 ^{ac}
Week 2	6.03 ^{ade}	6.04 ^{acd}
Week 4	6.09 ^{ae}	6.07 ^{acd}
Week 6	6.10 ^{ae}	6.12 ^{ad}
Week 8	6.10 ^{ae}	6.15 ^{ad}
Week 10	5.95 ^{ac}	6.13 ^{bd}
Week 12	5.99 ^{acd}	6.10 ^{bd}
Std Error	0.03	0.03

^{ab} Means with the same superscript letters across row are not different ($P > 0.05$).

^{cde} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

Table 8. Least squares means for dip by storage week interaction for pH values.

	<u>Dip</u> ¹			
	Control	SWPA	KL	LA
<i>Storage Weeks</i>				
Week 0	6.32 ^{ade}	5.37 ^{cd}	6.24 ^{abe}	6.18 ^{bd}
Week 2	6.24 ^{ad}	5.52 ^{be}	6.22 ^{ae}	6.18 ^{ad}
Week 4	6.34 ^{ade}	5.49 ^{cd}	6.33 ^{afg}	6.19 ^{bd}
Week 6	6.36 ^{ade}	5.52 ^{ce}	6.36 ^{afg}	6.21 ^{bd}
Week 8	6.37 ^{ade}	5.53 ^{ce}	6.38 ^{ag}	6.23 ^{bd}
Week 10	6.25 ^{ade}	5.47 ^{bd}	6.24 ^{aef}	6.18 ^{ad}
Week 12	6.35 ^{ade}	5.53 ^{ce}	6.05 ^{bd}	6.25 ^{ad}
Std. Error	0.04	0.04	0.04	0.04

^{abc} Means with the same superscript letters across row are not different ($P > 0.05$).

^{defg} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = Saline solution; SWPA = SafeOTM with propionic acid; KL = Potassium lactate; LA = Lactic acid.

Table 9. Least squares means for ingredient by storage week interaction for water activity (a_w) values.

	<u>Ingredient Treatment</u> ¹	
	Control	KL
Storage Week		
Week 0	0.977 ^{ac}	0.971 ^{bc}
Week 2	0.978 ^{acd}	0.973 ^{bd}
Week 4	0.978 ^{acd}	0.974 ^{bd}
Week 6	0.979 ^{ad}	0.974 ^{bde}
Week 8	0.979 ^{ad}	0.974 ^{bde}
Week 10	0.979 ^{acd}	0.976 ^{bf}
Week 12	0.978 ^{acd}	0.975 ^{bef}
Std Error	0.0005	0.0005

^{ab} Means with the same superscript letters across row are not different ($P > 0.05$).

^{cdef} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

increased ($P < 0.05$) in frankfurters with KL over refrigerated storage but overall a_w levels of the KL treatment were lower than the control.

There were no clear patterns observed for changes in frankfurter surface and internal color values (Table 6) over a 12-week refrigerated (4°C) storage period. Overall, storage at 4°C for 12 weeks did not affect surface and internal lightness and redness in frankfurters.

In general, lactate content (Table 6) varied ($P < 0.05$) during the 12-week storage period. The highest values for lactate content were observed on weeks 4, 8 and 12. Residual nitrite values decreased ($P < 0.05$) during the 12-week storage period and tables 10 and 11 show the ingredient treatment by storage week and dip by storage week interactions.

Frankfurters made with KL had slightly lower ($P < 0.05$) residual nitrite as compared to the control at week 0. In Table 11, levels of residual nitrite for the SWPA dip were the lowest among all treatments and the LA dip tended to be lower at week 0. Lower levels of residual nitrite were likely caused by the lower pH of SWPA. Studies have shown that less than 10% of the sodium nitrite used in the curing process remains in the finished product (Cassens, 1997). Also, it has been reported that the levels of residual nitrite decrease in cured products with increased storage time (Woolford and Cassens, 1977; Cassens and others, 1974; Kemp and others, 1975). The use of nitrite in cured meat products has been challenged as a potential health risk to human beings and as a necessary component of nitrosamine formation. Thus, cured meat products with lower levels of residual nitrite might be advantageous.

Sensory Evaluation

Descriptive attribute sensory panel evaluations of frankfurters containing KL (Table 12) indicated that the inclusion of KL had some effect ($P < 0.05$) on aromatics, feeling factor,

Table 10. Least squares means for ingredient by storage week interaction for nitrite (ppm) values

	<u>Ingredient Treatment</u> ¹	
	Control	KL
Storage Week		
Week 0	3.36 ^{ac}	2.55 ^{bc}
Week 8	0.75 ^{ad}	0.98 ^{ad}
Week 12	0.56 ^{ad}	0.56 ^{ad}
Std Error	0.17	0.17

^{ab} Means with the same superscript letters across row are not different ($P > 0.05$).

^{cd} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution

Table 11. Least squares means for dip by storage week interaction for nitrite (ppm).

	<u>Dip</u> ¹			
	Control	SWPA	KL	LA
<i>Storage Weeks</i>				
Week 0	3.48 ^{ac}	0.97 ^{bc}	3.72 ^{ac}	3.64 ^{dc}
Week 8	1.01 ^{abd}	0.42 ^{ac}	1.26 ^{bd}	0.77 ^{adb}
Week 12	0.77 ^{ad}	0.40 ^{ac}	0.50 ^{ad}	0.57 ^{ad}
Std. Error	0.23	0.23	0.23	0.23

^{ab} Means with the same superscript letters across row are not different ($P > 0.05$).

^{cd} Means within a column with the same superscript letters are not different ($P > 0.05$).

¹Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

Table 12. Least squares means of descriptive attribute sensory panel scores for aromatics¹, feeling factors¹, basic tastes¹, aftertastes and textures¹ of frankfurters containing KL.

	Control ²	KL ²	Std. Error
<i>Aromatics</i>			
Overall Meat Flavor	6.25 ^a	6.44 ^a	0.030
Fatty	2.14 ^a	2.28 ^b	0.010
Smoke	2.07 ^a	2.13 ^a	0.010
Spice	2.96 ^a	3.02 ^a	0.040
Cardboard	0.03 ^a	0.02 ^a	0.020
Painty	0.00 ^a	0.01 ^a	0.004
Fishy	0.01 ^a	0.00 ^a	0.010
Caramelized	0.06 ^a	0.08 ^a	0.002
Vinegar	0.58 ^a	0.46 ^a	0.020
<i>Feeling Factors</i>			
Astringent	2.52 ^a	2.64 ^b	0.004
Metallic	2.14 ^a	2.21 ^a	0.005
<i>Basic Tastes</i>			
Salt	3.83 ^a	4.43 ^a	0.090
Sour	2.52 ^a	2.56 ^a	0.030
Bitter	2.39 ^a	2.43 ^b	0.002
Sweet	0.72 ^a	0.69 ^a	0.060
<i>Aftertastes</i>			
Fat Mouthfeel	0.97 ^a	1.05 ^a	0.030
Sour	0.91 ^a	0.86 ^a	0.005
Bitter	0.73 ^a	0.81 ^b	0.003
Salty	1.20 ^a	1.52 ^a	0.030
<i>Textures</i>			
Springiness	6.81 ^a	6.74 ^a	0.044
Juiciness	3.88 ^a	4.01 ^a	0.050
Hardness	4.85 ^a	4.72 ^a	0.090
Cohesiveness of mass	4.29 ^a	4.21 ^a	0.030

^{ab} Means in the same row with different superscript letters are different ($P < 0.05$).

¹ Based on a 16-point intensity scale (0 = absence of an attribute; 15 = extremely intense).

² Control = no KL, KL = 3.3% of a 60% solution.

basic taste and aftertaste attributes. The addition of KL slightly increased fatty, astringent, bitter and bitter aftertaste attributes while cardboard, painty and fishy flavor/aromatics, descriptors associated with warmed-over-flavor (WOF), were not affected by the addition of KL. No differences ($P > 0.05$) in springiness, juiciness, hardness and cohesiveness of mass were found in frankfurters with KL.

Antimicrobial dips applied on surface of frankfurters (Table 13) appeared to have little effect on the sensory properties of the frankfurters. Sensory panelists found a slight decrease ($P < 0.05$) in overall meat flavor (most dominant flavor) and sweet taste in franks treated with SWPA as compared to the other dip treatments. Moreover, some descriptive attributes such as caramelized and vinegar flavors, astringent feeling factor, sour, bitter, and sweet tastes, sour aftertaste and hardness were increased slightly by SWPA dip. Off-flavor notes were not affected ($P < 0.05$) by antimicrobial dips. Overall, SWPA dip had only a slight effect on some sensory properties of the frankfurters, but these differences were at minimal perception levels and would not likely be detected by consumers.

As shown in Table 14, storage caused only minor changes ($P < 0.05$) in sensory flavors, feeling factors, tastes, aftertaste and textural attributes. The descriptive attributes affected across storage periods were: overall meat flavor, fatty and caramelized flavors; astringent and metallic feeling factors; sour and sweet tastes; sour and vinegar aftertastes; and springiness, juiciness, hardness and cohesiveness of mass. Overall meat flavor score of franks increased ($P < 0.05$) slightly after 8 weeks of refrigerated storage, but after 12 weeks were not different from those at 0 week. Fatty flavor scores increased ($P < 0.05$) slightly after 2 weeks, but then declined slightly throughout the 12-week storage period.

Table 13. Least squares means of descriptive attribute sensory panel scores for aromatics¹, feeling factors¹, basic tastes¹, aftertastes¹ and textures¹ of frankfurters dipped in antimicrobial solutions.

	Control ²	SWPA ²	KL ²	LA ²	Std. Error
Aromatics					
Overall Meat Flavor	6.44 ^b	6.09 ^a	6.40 ^b	6.45 ^b	0.064
Fatty	2.24 ^a	2.12 ^a	2.23 ^a	2.24 ^a	0.030
Smoke	2.13 ^a	2.07 ^a	2.07 ^a	2.11 ^a	0.040
Spice Complex	2.97 ^a	3.03 ^a	2.92 ^a	3.03 ^a	0.030
Cardboard	0.02 ^a	0.01 ^a	0.07 ^a	0.01 ^a	0.030
Painty	0.01 ^a	0.00 ^a	0.00 ^a	0.00 ^a	0.005
Fishy	0.00 ^a	0.00 ^a	0.01 ^a	0.00 ^a	0.007
Caramelized	0.07 ^a	0.13 ^c	0.04 ^b	0.04 ^b	0.070
Vinegar	0.16 ^a	1.41 ^b	0.26 ^a	0.23 ^a	0.080
Feeling Factors					
Astringent	2.53 ^{ab}	2.74 ^c	2.47 ^a	2.57 ^b	0.030
Metallic	2.15 ^a	2.27 ^b	2.14 ^a	2.16 ^a	0.023
Basic Tastes					
Salt	4.13 ^a	4.10 ^a	4.09 ^a	4.18 ^a	0.104
Sour	2.37 ^a	2.84 ^b	2.49 ^a	2.46 ^a	0.059
Bitter	2.39 ^{ab}	2.45 ^c	2.38 ^a	2.43 ^{bc}	0.011
Sweet	0.81 ^b	0.57 ^a	0.80 ^b	0.65 ^{ab}	0.045
Aftertastes					
Fat Mouthfeel	1.03 ^a	0.97 ^a	1.01 ^a	0.99 ^a	0.035
Sour	0.62 ^a	1.27 ^b	0.83 ^a	0.82 ^a	0.074
Bitter	0.81 ^a	0.84 ^a	0.68 ^a	0.75 ^a	0.041
Salty	1.49 ^a	1.19 ^a	1.28 ^a	1.46 ^a	0.080
Textures					
Springiness	6.75 ^a	6.83 ^a	6.75 ^a	6.78 ^a	0.039
Juiciness	3.91 ^a	3.95 ^a	3.96 ^a	3.96 ^a	0.039
Hardness	4.72 ^{ab}	4.94 ^c	4.68 ^a	4.79 ^b	0.031
Cohesiveness of mass	4.27 ^a	4.23 ^a	4.24 ^a	4.26 ^a	0.027

^{abc} Means in the same row with different superscript letters are different ($P < 0.05$).

¹ Based on a 16-point intensity scale (0 = absence of an attribute; 15 = extremely intense).

² Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

Table 14. Least squares means of descriptive attribute sensory panel scores for aromatics¹, feeling factors¹, basic tastes¹, aftertastes¹ and textures¹ of frankfurters stored under at refrigerated conditions.

	Week 0	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12	Std. Error
Aromatics								
Overall Meat Flavor	6.25 ^{ab}	6.08 ^a	6.09 ^{ab}	6.28 ^{ab}	6.66 ^c	6.76 ^c	6.29 ^b	0.07
Fatty	2.25 ^{ac}	2.38 ^b	2.25 ^a	2.16 ^{ac}	2.14 ^c	2.10 ^c	2.16 ^{ac}	0.04
Smoke	1.94 ^a	2.12 ^a	2.07 ^a	2.22 ^a	2.17 ^a	2.07 ^a	2.10 ^a	0.06
Spice Complex	3.08 ^a	2.83 ^a	2.98 ^a	3.05 ^a	2.99 ^a	3.07 ^a	2.94 ^a	0.06
Cardboard	0.00 ^a	0.03 ^a	0.00 ^a	0.02 ^a	0.05 ^a	0.02 ^a	0.08 ^a	0.03
Painty	0.00 ^a	0.02 ^a	0.00 ^a	0.00 ^a	0.00 ^a	0.00 ^a	0.00 ^a	0.01
Fishy	0.00 ^a	0.00 ^a	0.01 ^a	0.00 ^a	0.03 ^a	0.00 ^a	0.00 ^a	0.01
Caramelized	0.02 ^{ab}	0.05 ^{ab}	0.00 ^a	0.10 ^{bc}	0.08 ^{ab}	0.16 ^c	0.08 ^{ab}	0.03
Vinegar	0.42 ^a	0.44 ^a	0.44 ^a	0.49 ^a	0.47 ^a	0.61 ^a	0.76 ^a	0.11
Feeling Factors								
Astringent	2.31 ^a	2.47 ^{ab}	2.47 ^{ab}	2.63 ^{bc}	2.57 ^b	2.80 ^c	2.79 ^c	0.06
Metallic	2.18 ^b	2.23 ^b	2.15 ^{ab}	2.19 ^b	2.05 ^a	2.21 ^b	2.24 ^b	0.04
Basic Tastes								
Salt	3.99 ^a	3.90 ^a	4.15 ^a	4.45 ^a	4.14 ^a	4.18 ^a	4.08 ^a	0.13
Sour	2.41 ^{ab}	2.32 ^a	2.49 ^{bc}	2.66 ^c	2.58 ^{bc}	2.66 ^c	2.68 ^d	0.06
Bitter	2.39 ^a	2.28 ^a	2.36 ^a	2.51 ^a	2.46 ^a	2.40 ^a	2.50 ^a	0.06
Sweet	0.86 ^c	0.82 ^{bc}	0.82 ^{bc}	0.70 ^{bc}	0.44 ^a	0.66 ^b	0.66 ^b	0.06
Aftertastes								
Fat Mouthfeel	0.99 ^a	1.04 ^a	0.98 ^a	1.07 ^a	0.93 ^a	0.90 ^a	1.15 ^a	0.09
Sour	0.55 ^a	0.49 ^a	0.81 ^b	0.82 ^b	0.95 ^b	1.25 ^c	1.34 ^c	0.09
Spice	1.77 ^b	1.58 ^{ab}	1.42 ^a	1.70 ^b	1.58 ^{ab}	1.79 ^b	1.24 ^a	0.08
Bitter	0.86 ^{bc}	0.63 ^{ab}	0.55 ^a	0.81 ^{abc}	0.81 ^{abc}	1.08 ^c	0.65 ^{ab}	0.10
Metallic	0.46 ^b	0.33 ^{ab}	0.13 ^a	0.49 ^b	0.31 ^{ab}	0.33 ^{ab}	0.20 ^a	0.08
Salty	1.60 ^{cd}	1.32 ^{abc}	1.68 ^d	1.50 ^{bcd}	1.21 ^{ab}	0.96 ^a	1.25 ^{ab}	0.11
Vinegar	0.05 ^a	0.04 ^a	0.06 ^a	0.02 ^a	0.00 ^a	0.00 ^a	0.19 ^b	0.04
Textures								
Springiness	6.51 ^a	6.66 ^a	6.80 ^{ab}	6.77 ^a	6.87 ^b	7.00 ^b	6.81 ^{ab}	0.07
Juiciness	4.20 ^b	3.95 ^b	3.40 ^a	3.89 ^b	4.08 ^b	4.20 ^b	3.91 ^b	0.11
Hardness	4.77 ^{bc}	4.53 ^a	4.70 ^{ab}	4.83 ^{bc}	4.84 ^{bc}	4.95 ^c	4.88 ^{bc}	0.07
Cohesiveness of mass	4.37 ^c	4.27 ^{bc}	4.31 ^{bc}	4.15 ^{ab}	4.18 ^{ab}	4.38 ^c	4.10 ^a	0.05

^{abcd} Means in the same row with different superscript letters are different ($P < 0.05$).

¹ Based on a 16-point intensity scale (0 = absence of an attribute; 15 = extremely intense).

² Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

Off-flavor was not affected ($P < 0.05$) by refrigerated storage period. Caramelized flavor scores were highest on week 6 and 10. Sensory panelists detected a higher astringent feeling for franks after 6 weeks and a lower metallic feeling on week 8. Sour taste tended to increase with storage and sweet taste was lower on week 8.

All aftertastes were detected at low levels but only sour and vinegar aftertastes were affected ($P < 0.05$) over storage. Sour aftertaste tended to increase with storage and vinegar slightly increased by week 12.

Springiness scores were higher on weeks 4, 8 and 10, and juiciness score was lower on week 4. Hardness decreased ($P < 0.05$) after 2 weeks, then increased slightly after 4 weeks, but at the end of the storage period was not different to week 0. Cohesiveness scores were lower on weeks 6, 8 and 10. Overall, storage for 12 weeks at 4.5°C had minimal effect on descriptive attributes of the frankfurters.

Comparative results of ingredient and dip treatments for subjective panel color and objective color determinations on the surface and the interior of franks are presented in Table 15. The addition of KL did not affect ($P < 0.05$) sensory or objective color of frankfurters, except that surface a^* values or redness were slightly lower in franks treated with KL. Franks treated with SWPA had slightly lower ($P < 0.05$) surface color scores as compared to the other antimicrobial dips. No differences in surface color scores were found in franks treated with SWPA and LA. SWPA dip made the surface of the franks slightly darker, increased surface redness and decreased internal yellowness. Overall, the SWPA dip appeared to enhance surface redness of franks. However, surface lightness and internal yellowness were slightly diminished.

Table 15. Least squares means of attribute sensory panel scores for color, and surface color values and internal color values of frankfurters containing KL and dipped in antimicrobial solutions.

<u>Ingredient Treatment</u> ¹	Surface Subjective Color Values ³	Internal Subjective Color Values ⁴	Surface Objective Color Values			Internal Objective Color Values		
			L*	a*	b*	L*	a*	b*
Control	4.18 ^a	4.24 ^a	63.59 ^a	18.52 ^a	12.91 ^a	68.44 ^a	15.21 ^a	9.63 ^a
KL	4.20 ^a	4.38 ^a	63.17 ^a	18.24 ^b	12.88 ^a	67.10 ^a	15.16 ^a	9.57 ^a
Std. Error	0.01	0.08	0.71	0.002	0.13	0.33	0.03	0.18
<u>Dip Treatment</u> ²								
Control	4.27 ^b	4.46 ^a	63.93 ^c	17.95 ^a	12.83 ^a	68.21 ^a	15.11 ^a	9.81 ^{bc}
SWPA	3.98 ^a	4.29 ^a	62.69 ^a	19.00 ^b	12.94 ^a	68.24 ^a	15.21 ^a	9.15 ^a
KL	4.36 ^b	4.43 ^a	63.51 ^{bc}	18.26 ^a	12.72 ^a	66.47 ^a	15.25 ^a	9.60 ^b
LA	4.16 ^{ab}	4.44 ^a	63.39 ^b	18.32 ^a	13.10 ^a	68.16 ^a	15.18 ^a	9.83 ^c
Std. Error	0.06	0.06	0.15	0.16	0.19	0.19	0.16	0.07

^{abc} Means within a column per main effect with different superscript letters are significantly different (P < 0.05).

¹Control = no KL, KL = 3.3% of a 60% solution.

²Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

³Based on a 7-point scale (1 = very dark reddish-brown; 7 = very pale pink).

⁴Based on a 7-point scale (1 = extremely pink color; 7 = no cured color-gray).

Sensory and objective color data over storage time (Table 16) indicated that surface color did not change and that internal color scores varied slightly with time. Surface and internal color L^* values were higher, and a^* and b^* values were lower on week 8. However, there were no clear patterns observed on internal color values for franks over storage time.

Microbial Evaluation of Frankfurters

Addition of KL did not affect ($P > 0.05$) APC and *L. monocytogenes* counts (Table 17) of the frankfurters. SWPA and LA dips tended to decrease ($P < 0.05$) APC counts slightly, but no statistical differences ($P > 0.05$) were observed in franks for either treatment. Franks treated with KL dip tended to have higher APC counts among the dip treatments, but KL was not different from the control.

Compared with the control (Table 17), *L. monocytogenes* on inoculated franks treated with SWPA and LA dips diminished significantly ($P < 0.05$) and especially with the SWPA. *L. monocytogenes* numbers were significantly reduced by 5.8 and 3.2 logs (Table 17), respectively, on frankfurters treated with SWPA and LA. These results demonstrate the antimicrobial effectiveness of these treatments for reducing on *L. monocytogenes* on the surface of frankfurters and therefore could afford considerable protection against this microorganism in RTE products. The mode action of organic acids for inhibiting microbial growth appears to be associated to proton donation, maintenance of acid-base equilibrium and production energy from the cells (Davidson and Branen, 1993).

Microbial evaluations of frankfurters during a 12-week refrigerated storage period are presented in the Table 18. APC counts (\log_{10} CFU/Frank) of non-inoculated samples increased approximately 1 to 1.5 logs over 12 weeks. Total log numbers of *L. monocytogenes* increased ($P < 0.05$) on the inoculated samples over a 12 week refrigerated (4°C) storage period but because the

Table 16. Least squares means of attribute sensory panel scores for color, and surface color values and internal color values of frankfurters stored under refrigerated conditions.

Storage Week	Surface Subjective Color Values ³	Internal Subjective Color Values ⁴	Surface Objective Color Values			Internal Objective Color Values		
			L*	a*	b*	L*	a*	b*
Week 0	4.16 ^a	4.48 ^{bc}	62.29 ^{ab}	19.02 ^b	12.65 ^{bc}	66.14 ^a	15.88 ^{bc}	9.29 ^{ab}
Week 2	3.95 ^a	4.25 ^{ab}	61.59 ^a	18.66 ^b	12.39 ^b	67.64 ^{ab}	14.60 ^{ab}	8.90 ^a
Week 4	4.30 ^a	4.09 ^a	63.67 ^b	19.16 ^b	13.46 ^{cd}	68.47 ^b	15.81 ^{bc}	9.90 ^{bc}
Week 6	4.33 ^a	4.78 ^c	63.13 ^{ab}	19.26 ^b	13.09 ^{bcd}	67.56 ^{ab}	16.03 ^c	9.75 ^b
Week 8	4.26 ^a	4.27 ^{ab}	67.39 ^c	15.24 ^a	11.19 ^a	70.53 ^c	13.44 ^a	8.90 ^a
Week 10	4.33 ^a	4.47 ^{bc}	62.37 ^{ab}	18.17 ^b	13.60 ^d	67.17 ^{ab}	14.89 ^{bc}	10.30 ^c
Week 12	3.99 ^a	4.48 ^{bc}	63.21 ^{ab}	19.16 ^b	13.90 ^d	66.89 ^{ab}	15.65 ^{bc}	10.14 ^{bc}
Std. Error	0.14	0.11	0.60	0.52	0.31	0.67	0.45	0.14

^{abc} Means within a column per main effect with different superscript letters are significantly different ($P < 0.05$).

¹ Based on a 7-point scale (1 = very dark reddish-brown; 7 = very pale pink).

² Based on a 7-point scale (1 = extremely pink color; 7 = no cured color-gray).

Table 17. Least squares means for APC, *Listeria monocytogenes* counts and log reduction on frankfurters containing KL and dipped in antimicrobial solutions.

	APC ³ (log ₁₀ CFU/Frank)	<i>Listeria monocytogenes</i> ⁴ (log ₁₀ CFU/Frank)	<i>Listeria monocytogenes</i> log reduction
<u>Ingredient Treatment¹</u>			
Control	3.1 ^a	5.2 ^a	3.0 ^a
KL	2.9 ^a	4.7 ^a	2.9 ^a
Std. Error	0.13	0.32	0.16
<u>Dip Treatment²</u>			
Control	3.6 ^{ab}	6.6 ^b	1.4 ^a
SWPA	1.8 ^a	1.7 ^c	5.8 ^c
KL	4.7 ^b	7.0 ^b	1.3 ^a
LA	2.0 ^a	4.5 ^a	3.2 ^b
Std. Error	0.61	0.44	0.40

^{abc} Means within a column and main effect with different superscript letters are significantly different ($P < 0.05$).

¹Control = no KL, KL = 3.3% of a 60% solution.

²Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

³APC counts for non-inoculated samples.

⁴*Listeria monocytogenes* counts for inoculated samples.

Table 18. Least squares means for APC, *Listeria monocytogenes* counts and log reduction of frankfurters stored under refrigerated conditions.

	APC ¹ (log ₁₀ CFU/Frank)	<i>Listeria monocytogenes</i> ² (log ₁₀ CFU/Frank)	<i>Listeria monocytogenes</i> log reduction
<u>Storage Week</u>			
Week 0	2.0 ^a	3.9 ^a	2.9 ^a
Week 2	2.7 ^b	3.9 ^a	3.3 ^a
Week 4	3.0 ^{bc}	4.4 ^{ab}	3.3 ^a
Week 6	3.5 ^c	4.9 ^b	3.2 ^a
Week 8	3.1 ^{bc}	5.8 ^c	2.7 ^a
Week 10	3.6 ^c	5.6 ^c	2.7 ^a
Week 12	3.3 ^c	6.1 ^c	2.5 ^a
Std. Error	0.24	0.23	0.24

^{abc} Means within a column and main effect with different superscript letters are significantly different (P < 0.05).

¹APC counts for non-inoculated samples.

²*Listeria monocytogenes* counts for inoculated samples.

treatment by storage weeks interaction was significant, the true effects are better presented in Table 19. *L. monocytogenes* counts increased ($P < 0.05$) on the frankfurter surface of all the dip treatments except SWPA. The log counts remained at the minimum detection level of approximately 1.7 logs over 12 weeks at 4.5°C for SWPA. These results are indicative of SWPA's listericidal and listeristatic effect since the initial inoculation level, prior to dipping, was 6.6 logs on the control. Thus, SWPA is an effective listericidal and listeristatic surface treatment for frankfurters stored under refrigerated conditions over a 12 weeks period.

Table 19. Least squares means for dip by storage week interaction for *Listeria monocytogenes* counts (log₁₀ CFU/Frank).

	<u>Dip¹</u>			
	Control	SWPA	KL	LA
<i>Storage Weeks</i>				
Week 0	5.2 ^{ae}	1.7 ^{ce}	5.1 ^{ae}	3.7 ^{bef}
Week 2	6.2 ^{ae}	1.8 ^{ce}	5.6 ^{ae}	3.3 ^{be}
Week 4	6.1 ^{ae}	1.7 ^{ce}	6.1 ^{ae}	3.8 ^{bef}
Week 6	5.7 ^{ae}	1.7 ^{de}	7.4 ^{bfg}	4.8 ^{cfg}
Week 8	8.1 ^{af}	1.7 ^{ce}	7.9 ^{ag}	5.4 ^{bg}
Week 10	7.8 ^{af}	1.8 ^{be}	8.2 ^{ag}	4.7 ^{af}
Week 12	8.3 ^{af}	1.7 ^{ce}	8.6 ^{ag}	5.8 ^{bg}
Std. Error	0.46	0.46	0.46	0.46

^{abcd} Means with the same superscript letters across row are not different (P > 0.05).

^{efg} Means within a column with the same superscript letters are not different (P > 0.05).

¹Control = Saline solution; SWPA = Safe₂O™ with propionic acid; KL = Potassium lactate; LA = Lactic acid.

SUMMARY AND CONCLUSIONS

The most significant observations in this study were that SWPA and LA dips were effective for reducing *L. monocytogenes* on the surface of franks just prior to vacuum packaging. SWPA counts remained at the minimum detection level of 1.7 logs immediately after dipping and throughout 12 weeks of storage at 4.5°C. *L. monocytogenes* increased on all other dips treated franks during storage, except SWPA. Thus, these results demonstrate a residual protective effect of the SWPA application over the storage studied.

Potassium lactate used as ingredient in frankfurters did not affect chemical composition (except percent moisture), process yield, sodium, phosphorus, vacuum-package purge, pH, a_w , lactate and objective color values.

Proximate composition of frankfurters was not affected by dip treatments. SWPA dip slightly increased vacuum-package purge and decreased pH by 0.83 units. Surface and internal color values were slightly affected in the franks treated with SWPA dip, but of such a small magnitude that may not be detectable by the consumers.

Trained panel sensory evaluations indicated that the addition of KL slightly increased fatty, astringent, bitter and bitter aftertaste attributes and that antimicrobial dips applied on the surface of frankfurters appeared to have little effect on the sensory properties of the franks, especially SWPA. Overall, sensory attributes were minimally affected by addition of KL to the frankfurters and antimicrobial dip treatments.

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APPENDIX

MIONIX Product Specification for Safe₂O™ brand Hot Dog Solution

Safe₂O™ is comprised of GRAS (Generally Recognized As Safe) food grade ingredients therefore legislatively allowed in food.

Product Name: Safe₂O™ -HD

Common Name: Highly Acidic Propionic Acid/Lactic Acid/Calcium Sulfate Complex

Chemical Name: Highly Acidic Propionic Acid/Lactic Acid/Calcium Sulfate Complex

Chemical Family: Organic Acid/ Calcium Salt

Physical Data:

Boiling Point:	104°C
Vapor Pressure:	Approximates water
Solubility in Water:	100%
Evaporation Rate (Butyl Acetate = 1.0):	~H ₂ O
Specific Gravity (H ₂ O = 1.0):	1.02-1.10
Vapor Density (Air = 1.0):	Approximates water
Odor:	0
Appearance:	Clear to slightly cloudy

Toxicity Information for Product Components: Same as Calcium Sulfate and Sulfuric Acid

Reactivity:	Stable
Conditions to avoid:	Boiling and Freezing
Materials to avoid:	None
Hazardous Decomposition Products:	None
Hazardous Polymerization:	No

Hazard Ratings (Least = 0, Slight = 1, Moderate = 2, High = 3, and Extreme = 4):

Health = 0
Fire = 0
Reactivity = 1

Application Instructions:

Product is concentrated
Mix Safe₂O™ with water in one (1) part Safe₂O™ Hot Dog solution with two (2) parts water.